



The 5th Congress of the Asia Pacific Initiative on Reproduction

4 - 6 April 2014 • Brisbane, Australia

in conjunction with FSA Annual Conference

FINAL PROGRAMME AND ABSTRACT BOOK



Organized by:

Aspire

Hosted by:



Held in:

brisbane
australia's new world city



www.aspirecongress.org

ACKNOWLEDGEMENTS

We wish to thank the following companies who, through their generosity, have helped make this Congress possible:

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WELCOME MESSAGE FROM THE PRESIDENT



Dear Colleagues,

It gives me great pleasure to welcome you to the 5th Congress of the Asia Pacific Initiative on Reproduction (ASPIRE 2014) held in conjunction with the Fertility Society of Australia Annual Conference from 4 to 6 April 2014 in Brisbane, Australia.

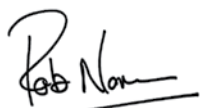
This congress themed “The Future of Reproductive Medicine in the Asian Century” will take a look at the future of science and practice in reproductive medicine. We are pleased to have highly respected and internationally renowned speakers who will share, discuss, debate, and dissect significant new developments and scientific advancements that will impact the future of fertility preservation, embryology, andrology, clinical trials, PCOS, PGS, IVM, and related fields.

The Fertility Society of Australia is proud to host this combined meeting of ASPIRE and FSA for the first time in the history of both societies. We extend a warm welcome to clinicians, researchers, academics, nurses, and other allied health professionals with interest in reproductive medicine and fertility from Asia Pacific and beyond.

Aside from the excellent scientific program, we have also prepared an exciting social program that will provide foreign delegates an opportunity to explore, indulge and relax during their stay in Brisbane. The Welcome Reception and Gala Dinner will give participants a chance to network among colleagues while enjoying the cuisine, culture, and warm hospitality that the new world city of Brisbane has to offer.

We wish everyone a fruitful meeting at ASPIRE and FSA Annual Conference 2014 and a memorable stay in Brisbane!

Sincerely,



Prof Robert Norman

President, ASPIRE

Organising Committee Chairman, ASPIRE 2014 and FSA Annual Conference 2014

WELCOME MESSAGE FROM THE LOCAL ORGANISING CHAIRPERSON



Dear Colleagues,

On behalf of the Local Organising Committee of ASPIRE 2014, welcome to Brisbane. In April, Brisbane is in a subtropical autumn and April is one of the best times to visit. The average daily temperature is 17°C to 26°C. Daylight hours are 5.30am until 5.45pm. Brisbane is the gateway to Queensland, Australia's playground and premier holiday destination.

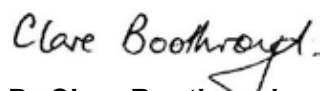
Brisbane is close to the Gold Coast, the Sunshine Coast, Moreton Bay, the Gold Coast hinterland with the rainforests of Lamington National Park and Mt Tambourine, vineyards and wineries of the apple growing areas of Stanthorpe. Further afield are whale watching, the Great Barrier Reef and rainforest. The World Heritage listed Fraser Island, with the Kingfisher Bay resort, is the world's largest sand island and is a four hour drive away with amazing native wildlife and unspoilt beaches. The Queensland coast abounds in island resorts offering diving, snorkeling, boating and fishing. Truly Queensland has something for everyone. You will find that many of the locals have not had the time to savour the many and varied delights this area has to offer. Either before or after the ASPIRE meeting I encourage you to see a bit of our beautiful state of Queensland.

There are short day trips within the city of Brisbane including river cruises, visits to native zoos and the Lone Pine koala sanctuary. For the fit and adventurous there is a climb of the Storey Bridge for breathtaking and windy view of Moreton Bay. For the cricket lovers there is the mandatory visit to the Gabba and maybe even a game of Australian Rules Football. The Gallery of Modern Art and Queensland Art Gallery located an easy walk from the convention centre and house a great collection of classic and modern Australian and Asian art. Something for everyone is here in Brisbane.

In addition to the local attractions we will have a world class entertainment and food for the conference which will provide an ideal opportunity to relax and exchange ideas with colleagues from other countries.

We look forward to your joining us for the 5th Annual Congress of ASPIRE 2014 and the Annual Conference of the Fertility Society of Australia and to experience Queensland.

Sincerely,



Dr Clare Boothroyd
 Local Organising Chair

ABOUT THE ORGANISERS



The Asia Pacific Initiative on Reproduction, was founded in 2001 to improve knowledge and awareness of ART and infertility-related services, with ultimate aim of improving the quality of patient care.

The objectives of ASPIRE are:

- To improve the quality of patient care
- To develop and advance fertility services in the Asia-Pacific region
- To provide a forum for professionals in this field to share experiences and information
- To form a cohesive group to promote infertility management across the region, and to form relations with local and international associations
- To raise awareness and understanding of treatment options for infertility amongst healthcare professionals and the public
- To assist healthcare professionals in motivating patients to receive treatment for infertility
- To facilitate the dissemination of accurate information about infertility to patients and the public
- To support the basic reproductive rights of all couples and individuals to decide freely and responsibly how many children to have and when to have them

Visit www.aspire-reproduction.org for more information.



The Fertility Society of Australia is the peak body representing scientists, doctors, researchers, nurses, consumers and counsellors in reproductive medicine in Australia & New Zealand. Each year the FSA holds a Scientific Meeting attracting experts in reproductive health from around the world to present research and discuss new technologies and treatments. In 2014, the FSA Annual Conference will be held alongside the 5th Congress of Asia Pacific Initiative on Reproduction (ASPIRE) from 4-6 April 2014 at the Brisbane Convention & Exhibition Centre.

Australia has an outstanding record in helping couples experiencing infertility. Thousands of women in Australia and around the world have conceived and given birth using pioneering in vitro fertilisation techniques developed and perfected in this country.

For more information, visit www.fertilitysociety.com.au.

ORGANISING COMMITTEE

ASPIRE Executive Board

President:	Prof Robert Norman	
Secretary General:	Prof Bruno Lunenfeld	
President-Elect:	Dr Jaideep Malhotra	
Treasurer:	Prof Young Min Choi	
Past President:	Prof Yoshiharu Morimoto	
Board Members:	Dr Andon Hestiantoro	Dr Haris Hamzah
	Dr Manh Tuong Ho	Dr Hsin-Fu Chen
	Dr Asma Munir	Dr Nao Suzuki
	Dr Hari Kishor Shrestha	

Advisor to Executive Board: Prof PC Wong

FSA Board Members

President:	Assoc Prof Mark Bowman	
Vice President:	Prof Michael Chapman	
Treasurer:	Mr Adnan Catakovic	
Secretary:	Ms Donna Close	
Board Members:	Dr Judith Applegarth	Dr Andrew Murray
	Ms Kate Bourne	Assoc Prof Luk Rombauts
	Dr Anne Clark	Prof Elizabeth Sullivan
	Ms Jacqui Irving	Ms Louise Younger

Local Organising Committee

Chair:	Dr Clare Boothroyd	
Members:	Ms Carmel Carrigan	Dr Julie Lindstrom
	Ms Donna Close	Dr Kee Ong
	Dr Ben Kroon	Ms Margie Parry

Scientific Programme Committee

International Scientific Board for ASPIRE 2014

Prof Dan Dumesic, <i>USA</i>	Prof Jie Qiao, <i>China</i>
Prof Human Fatemi, <i>Kuwait</i>	Assoc Prof Luk Rombauts, <i>Australia</i>
Prof Bart CJM Fauser, <i>The Netherlands</i>	Dr Alan Trounson, <i>USA</i>
Prof Ian Fraser, <i>Australia</i>	Prof PC Wong, <i>Singapore</i>
Prof Yoshiharu Morimoto, <i>Japan</i>	Prof Nick Macklon, <i>United Kingdom</i>
Prof Pasquale Patrizio, <i>USA</i>	

Scientific Advisory Committee for ASPIRE 2014

Prof Cindy Farquhar	Prof Robert McLachlan
Prof Karin Hammarberg	Dr Cecilia Sjoblom
Prof Roger Hart	Assoc Prof Jeremy Thompson
Prof Bill Ledger	Assoc Prof Sheryl de Lacey

GENERAL INFORMATION

Venue

Brisbane Convention & Exhibition Centre (BCEC)

Merivale St, South Brisbane QLD 4101, Australia

Telephone: +61 7 3308 3000

Website: www.bcec.com.au

About Brisbane, Australia

Brisbane, Australia's third largest city, is situated in the south-east of Queensland. This stylish and vibrant city has evolved around the large River Brisbane, which meanders through the centre, often attracting large black whaler and bull sharks.

Brisbane's impressive Central Business District (CBD) is dominated by the historic Brisbane City Hall and features many breathtaking skyscrapers, office blocks and modern buildings, with magnificent architecture. One of Australia's prominent financial, commercial and industrial cities, Brisbane is home to many major corporations, international companies and entertainment complexes, including the enormous Conrad Treasury Casino, which is situated next to the Brisbane River in the Central Business District (CBD). Many hotels are concentrated around the city centre.

Language

The official language of ASPIRE 2014 is English. No simultaneous interpretation is provided at this congress.

Weather

In April, Brisbane enjoys excellent weather with lows of 16.2°C and highs of 26°C.

Time Zone

Brisbane operates on Australian Eastern Standard Time – GMT plus ten hours. Daylight savings times do not apply in Queensland.

Currency

Decimal currency is used in Australia - units are dollars and cents. All major credit cards are widely accepted in Australia.

Banks

Normal banking hours are Monday to Thursday 9:30am-4:00pm and Fridays 9:30am-5:00pm, excluding public holidays. 24 Hour Automatic Teller Machines (ATMs) can be found throughout the city and shopping centres.

Taxes

A Goods and Services Tax (GST) of 10% applies to all consumer goods and is included in retail prices. All fees are quoted in Australian Dollars and are subject to GST.

Travelling to and from Brisbane Airport

Brisbane Airport (BNE) is located just 13 km / 8 miles to the north-east of Brisbane city centre. Taxi ranks are conveniently located at both the International and Domestic Terminals. At the Domestic Terminal, the taxi rank is located centrally in front of the terminal. At the International Terminal, the taxi rank is located at the northern end of Arrivals on Level 2.

For more information please contact:

Black & White Cabs

Phone: 133222

Website: www.blackandwhitecabs.com.au

Yellow Cab

Phone: 131924

Website: www.yellowcab.com.au

Alternatively, you may wish to take a 20-minute journey on the Airtrain from the Brisbane Airport to the city center and vice versa. The Airtrain stations at the Domestic and International Airports are located directly outside the terminals, with trains departing every 15 minutes during peak periods. The Brisbane Convention and Exhibition Centre is 2 minutes from South Brisbane Station. For more information, please visit www.airtrain.com.au.

Registration and Information Desks

Registration and information desks are located near the main entrance on the Foyer Level. Registration opening hours are as follows:

Day	Date	Registration Hours
Thursday	3 April	8:00am - 6:00pm
Friday	4 April	7:00am - 6:00pm
Saturday	5 April	6:30am - 5:00pm
Sunday	6 April	8:00am - 3:00pm

Exhibition Opening Hours

The Congress exhibition is located at the Great Hall (Door 1) on the Foyer Level. Please note that admission is by badge only. Exhibition opening hours are as follows:

Day	Date	Exhibition Hours
Thursday	3 April	5:30pm - 8:00pm
Friday	4 April	8:00am - 6:30pm
Saturday	5 April	8:00am - 6:00pm
Sunday	6 April	8:00am - 2:00pm

Internet Access

Complimentary WIFI is available within the Congress venue and no password is required. It is designed for web browsing and checking web-based email and not designed for accessing VPN's or downloading large files. Delegates who wish to upgrade their WIFI speed may approach the information counter near the entrance of BCEC on the Foyer Level to purchase upgraded access at AUD 10 per device for the duration of the Congress.

Tea Breaks and Lunch

Morning and afternoon tea, as well as lunch are served in the Exhibition Hall (Great Hall Door 1, Foyer Level) daily, and are included in the price of the meeting.

Welcome Reception, 3 April

All registered delegates are invited to join us for an evening of networking, drinks and exciting performances to kick off ASPIRE 2014 on 3 April, from 6:00pm to 8:30pm. The programme will begin at Great Hall Door 6 and 7 on the Mezzanine Level and followed by a reception in the Exhibition Hall (Great Hall Door 1, Foyer Level). Admission is by badge only.

Gala Dinner, 5 April

All delegates with a gala ticket are invited to enjoy a networking and cultural evening at the ASPIRE 2014 Gala Dinner ticket on Saturday, 5 April from 7:00pm at the Brisbane City Hall. Admission is by Gala Dinner ticket only.

Venue address:

Epicure Brisbane City Hall
64 Adelaide St, Brisbane QLD 4000

Return coach transfers are available at the Brisbane Convention & Exhibition Centre to bring guests to and from Brisbane City Hall. Delegates who are attending this function are advised to gather at the Glenelg street bus stop outside the main entrance of BCEC based on the coach transfer schedule as follows:

Time	Route
6:45pm	BCEC to Brisbane City Hall
7:05pm	BCEC to Brisbane City Hall
10:00pm	Brisbane City Hall to BCEC
10:30pm	Brisbane City Hall to BCEC

The venue is a short five minute taxi ride away from BCEC for delegates who may wish to make their own way there or miss the coach transfers.

Post-Congress Tours

Post-Congress tours are available for delegates to sign up at a separate fee. Delegates may approach the Tour Desk near the main entrance on the Foyer Level for more information.

Speakers Preparation Room

Upon registration, all speakers are requested to proceed to the Speakers Preparation Room at M10 Speakers Presentation Centre located on the Mezzanine level, to submit your PowerPoint slide presentations. Please note that all PowerPoint slide presentations should be submitted at least 4 hours prior to your session.

Please note the opening hours of the Speakers Preparation Room as follows:

Day	Date	Speakers Preparation Room Opening Hours
Thursday	3 April	10:00am – 6:00pm
Friday	4 April	7:00am – 6:00pm
Saturday	5 April	6:30am – 5:00pm
Sunday	6 April	8:00am - 3:00pm

ASPIRE 2014 Awards

Five ASPIRE 2014 Awards will be presented to authors of the best papers and poster presentations at the Closing Ceremony, to be held in Great Hall (Door 6) on Sunday, 6 April from 2:30pm to 3:00pm.

FSA 2014 Awards

Five FSA 2014 Awards will be presented to FSA members who are authors of the best papers and poster at the Gala Dinner, to be held on Saturday, 5 April at the Brisbane City Hall.

Certificate of Attendance

Certificates of Attendance are available for collection at the pre-registration counters located near the main entrance of BCEC on the Foyer Level from Saturday, 5 April, 3:00pm onwards.

Liability and Insurance

The Congress Secretariat and Organisers cannot accept liability for personal accidents or loss of or damage to private property of participants and accompanying persons. Participants are advised to take out their own personal travel and health insurance for their trip.

Messages/Medical Assistance/Lost & Found

Please approach the Information Desk for assistance.

Important Numbers

Ambulance	PH 13 12 33
Police	PH 07 3364 6464
Ambulance / Fire Brigade / Police (Emergencies only)	PH 000

NOTE: It is advisable to always have the telephone number and the address of your embassy or consulate with you.

Congress Organisers

For post-Congress enquiries and assistance, please contact the Congress Secretariat:



20 Kallang Avenue
 Pico Creative Centre, Singapore 339411
 Tel: +65 6295 6984 / +65 6393 0202
 Fax: +65 6292 4721 / +65 6292 7577
 Email: aspirecongress@kenes.com

REGISTRATION INFORMATION

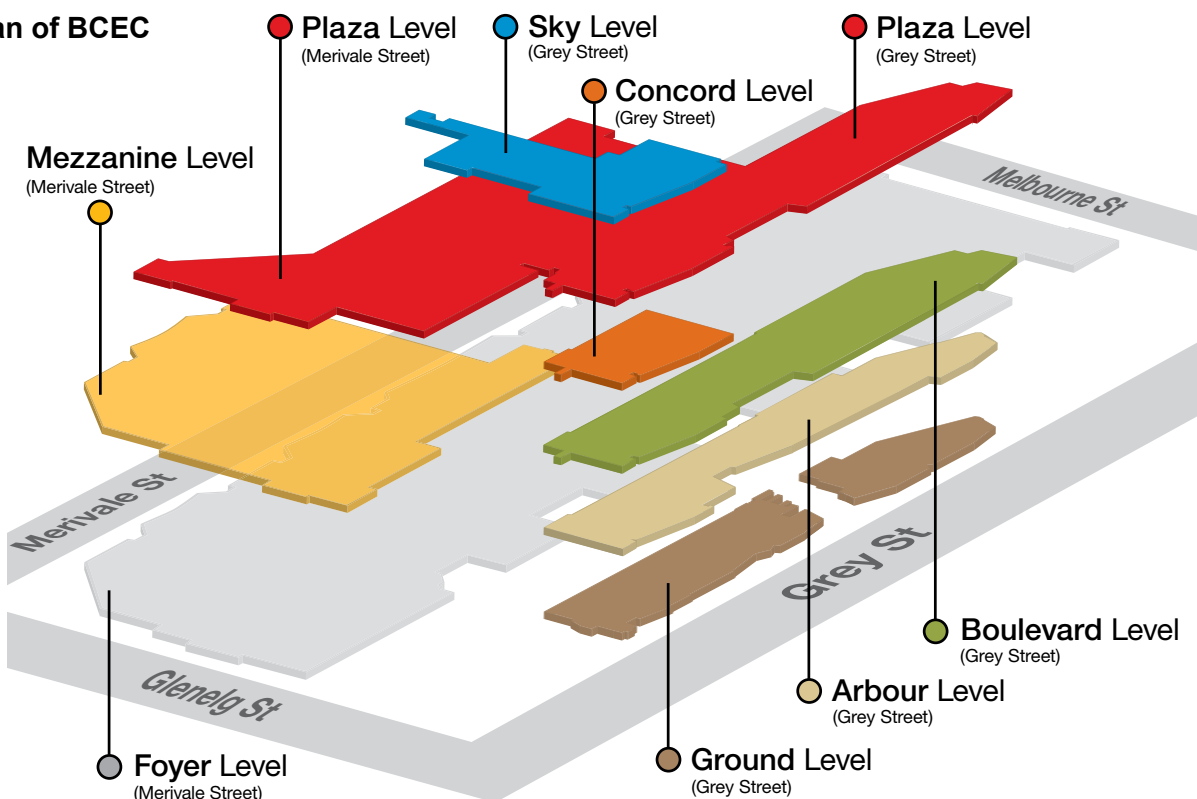
Registration Category	Onsite Rates* (04-06 Apr 2014)
Delegate (with Gala Dinner ticket)	AUD 968
Delegate (without Gala Dinner ticket)	AUD 850
Student/Trainee	AUD 400+
Day Registration	AUD 500
Accompanying Person	AUD 250
Gala Dinner Ticket	AUD 118

*Kindly note that delegates who register onsite may not receive all Congress materials but will be entitled to lunches, tea breaks, as well as entry to the Congress exhibition. Prices include GST.

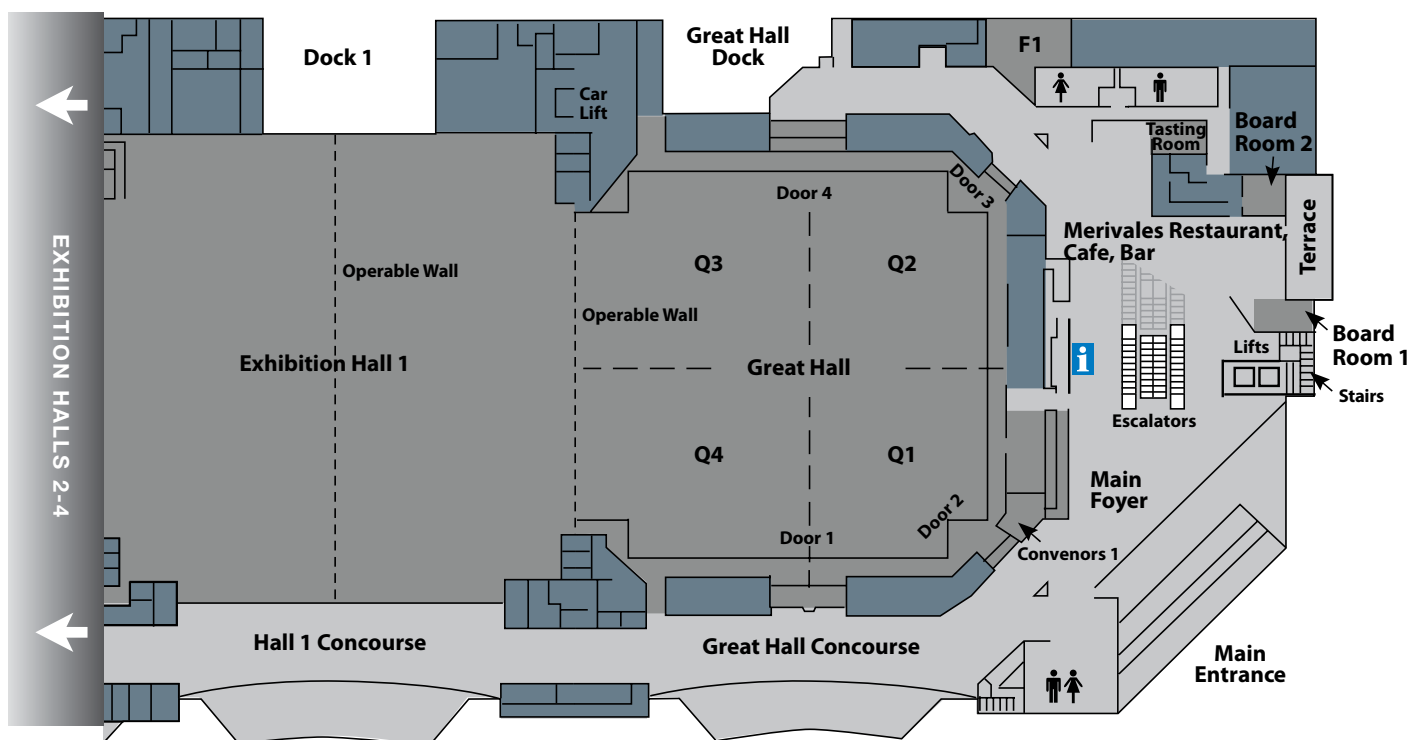
+Proof of student or trainee status must be produced.

VENUE FLOORPLANS

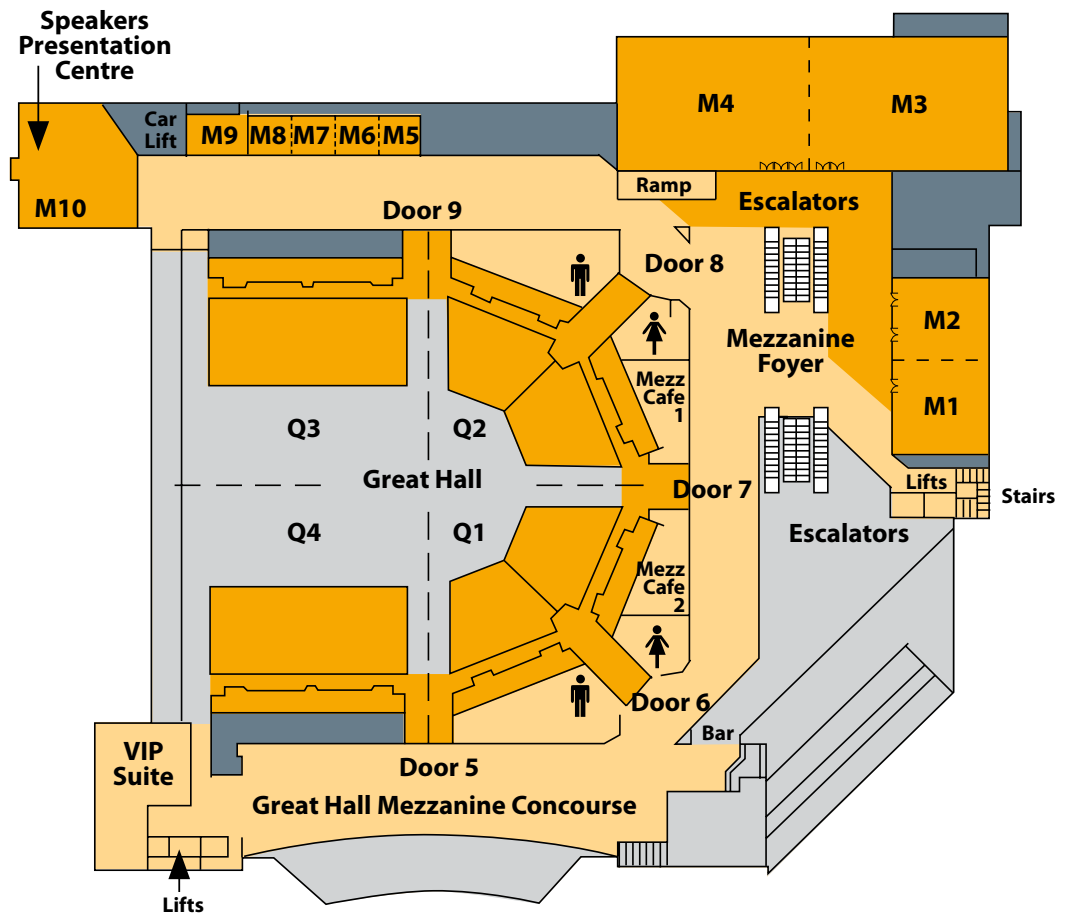
Overall plan of BCEC



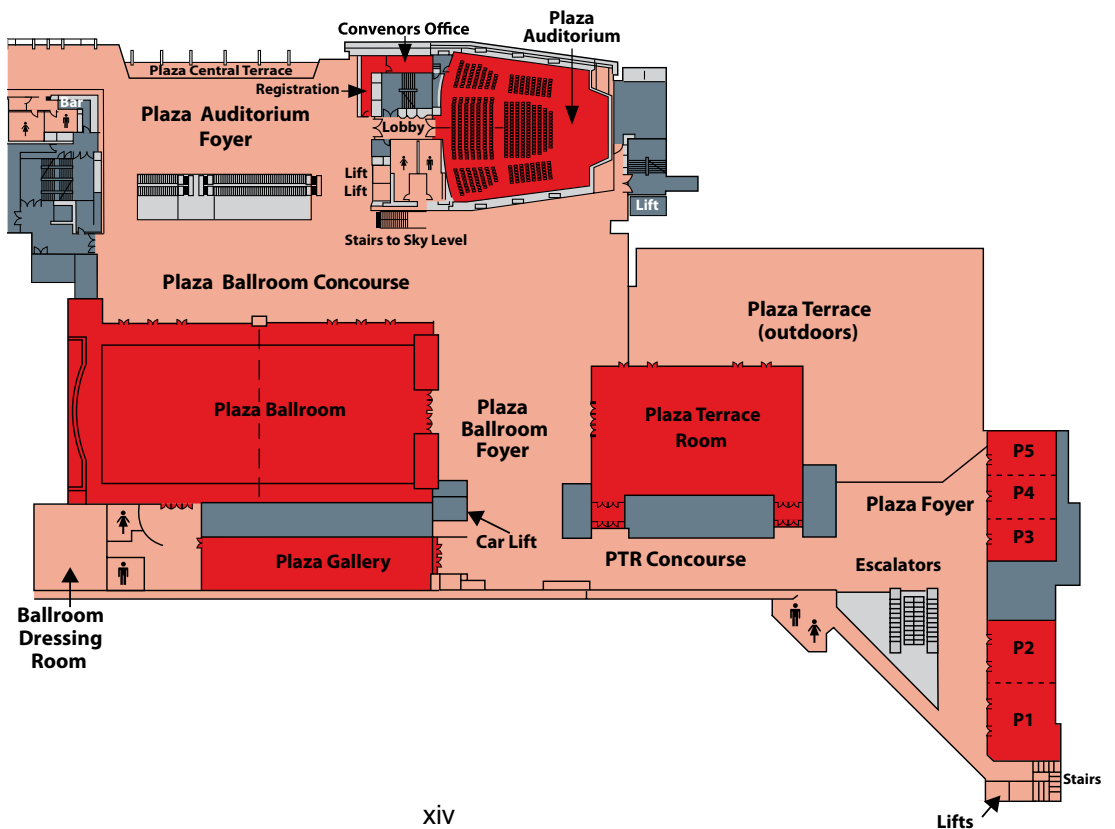
Foyer Level



Mezzanine Level



Plaza Level



PROGRAMME AT A GLANCE

TIME	THURSDAY, 03 APRIL	
08:30-09:00	Merck Serono Symposium Pushing Boundaries: Reproductive Care Across Borders (Held at Rydges South Bank Brisbane.) (Registration is separate from ASPIRE 2014)	
09:00-09:30		
09:30-10:00		
10:30-11:00		
11:30-12:00		
12:00-12:30	Lunch	
12:30-13:30		
13:30-14:00	Merck Serono Symposium (cont'd) (Held at Rydges South Bank Brisbane.) (Registration is separate from ASPIRE 2014)	Industry Sponsored Symposium 1 New Perspectives on IVM Sponsored By: Cook Medical <i>Chair and Speaker: A Prof Jeremy Thompson (AUS)</i> <i>Speakers: A Prof Rob Gilchrist (AUS);</i> <i>Dr Michel de Vos(BEL)</i> (Venue: Meeting Room M1)
14:30-15:00		
15:00-15:30		
15:30-16:00		
16:00-16:30		
16:30-17:00		
18:00-20:30	Social Function: Welcome Reception (Venue: Great Hall Door 6 & 7, Mezzanine Level)	

TIME	FRIDAY, 04 APRIL					
08:30-08:50	Opening Ceremony: Introduction <i>Prof Robert Norman (AUS) and Dr Clare Boothroyd (AUS)</i> (Venue: Great Hall Door 6 & 7)					
08:50-09:30	Opening Lecture: <i>Chairs: Prof Robert Norman(AUS); Prof Jaideep Malhotra(IND)</i> Vaccines and Reproduction: Where are we heading? <i>Prof Ian Frazer (AUS)</i> (Venue: Great Hall Door 6 & 7)					
09:30-10:00	Keynote Lecture: The Future of Stem Cells <i>Prof Alan Trounson (USA)</i> (Venue: Great Hall Door 6 & 7)					
10:00-10:30	Plenary Lecture 1: Infertility As A Global Health Problem <i>Prof Chittaranjan Purandare (IND)</i> (Venue: Great Hall Door 6 & 7)					
10:30-11:00	Coffee Break and Exhibition Viewing (Venue: Great Hall Door 1)					
11:00-11:30	Concurrent Session 1: The Future of Genetics and Environment in Determining Disease in Reproduction <i>Chairs: Prof Shin Yong Moon (KOR); Prof Linda Giudice (USA)</i> GWAS and post-GWAS of PCOS <i>Prof Zi-Jiang Chen (CHN);</i> Genetics of Menopause <i>Prof Bart Fauser (NET);</i> Pharmacogenetics of ART <i>Prof Young Min Choi (KOR)</i> (Venue: Great Hall Door 6)	Industry Sponsored Symposium 2: Cracking the Code: Biomarkers in Embryo Selection and Uterine Receptivity (Sponsored by Merck Serono) <i>Chair: Prof David K. Gardner</i> Innovation in ART <i>Dr Diego Ezcurra (IND);</i> Utilization of Electrochemiluminescence immunoassay (ECLIA) for the detection of early embryonic biomarkers for embryo selection <i>Prof Jie Li(CHN);</i> Uterine receptivity: the final hurdle in IVF <i>Prof Lois Salamonsen (AUS)</i> (Venue: Great Hall Door 8)	Free Communication 1: ART Basic, Embryology and Laboratory <i>Chairs: Ms Helena Jericho (AUS); Dr Cecilia Sjoblom (AUS)</i> (Venue: Meeting Room P1)	Free Communication 2: Nursing Counselling, Ethics, Psychology <i>Chairs: Ms Donna Close (AUS); Dr Petra Thorn (GER)</i> (Venue: Meeting Room P2)	Free Communication 3: Contraception, Endocrinology, Laboratory <i>Chairs: Dr Anna Smirnova (RUS); Dr Robert Lahoud (AUS)</i> (Venue: Meeting Room M2)	"How to ... Session 1": 11:00-11:45 How to Plan and Participate in a Clinical Trial <i>A/Prof Neil Johnson (NZ)</i> <i>Chair: Prof Ernest Ng (HK)</i> (Venue: Meeting Room M1)
11:30-12:00						
12:00-12:30						"How to ... Session 2": 11:45-12:30 How to Understand GnRH Control <i>Prof Bruno Lunenfeld (ISRAEL)</i> <i>Chair: Dr Louise Hull (AUS)</i> (Venue: Meeting Room M1)

PROGRAMME AT A GLANCE

12:30-13:30	Lunch and Poster Session / Mini Oral Presentations 1 <i>Chairs: Prof Gab Kovacs (AUS); Prof Young Min Choi (KOR)</i> (Venue: Great Hall Door 1)			FSA IVF Directors' Meeting (By Invitation) (Venue: Meeting Room M2)		
13:30-14:00	Concurrent Session 2: The Future Of Embryology <i>Chairs: Dr Colin Howles (SWITZ); Dr Phillip Matson (AUS)</i> Video Microscopy <i>Dr Yasuyuki Mio (Japan); Videomicroscopy - Clinical Results</i> <i>Dr Marcos Meseguer (SPN); Latest in Embryo Metabolomics</i> <i>Dr Hsin-Fu Chen (Taiwan)</i> (Venue: Great Hall Door 6)	Industry Sponsored Symposium 3: All women at 30 yo that are not in a relationship should freeze their eggs (Sponsored by MSD) <i>Chair: Prof Bart Fauser(NET)</i> Debaters: <i>Dr Devora Lieberman (Aus); Dr Kelton Tremellen (Aus)</i> (Venue: Great Hall Door 8)	Free Communication 4: Nursing Counselling, Ethics, Psychology <i>Chairs: Mrs Carmel Carrigan (AUS); Mrs Helen Alvino (AUS)</i> (Venue: Meeting Room P1)	Free Communication 5: Ovary <i>Chairs: Prof Kyung-Ah Lee (KOR); Prof Ray Rodgers (AUS)</i> (Venue: Meeting Room P2)	Free Communication 6: Fertility Preservation, Male Fertility <i>Chairs: Dr Hari Kishor Shrestha (NPL); Dr K.K. Ishwaran (MYL)</i> (Venue: Meeting Room M2)	"How to ... Session 3": 13:30-14:15 How to Diagnose PCOS in Adolescence <i>Dr Delfin A.Tan (PHIL)</i> <i>Chair: Prof Helena Teede (AUS)</i> (Venue: Meeting Room M1)
14:00-14:30						
14:30-15:00						"How to ... Session 4": 14:15-15:00 How to present a paper at a conference when English is not your first language <i>Prof Robert Norman (AUS)</i> <i>Chair: Prof PC Wong (SG)</i> (Venue: Meeting Room M1)
15:00-15:30	Coffee Break and Exhibition Viewing (Venue: Great Hall Door 1)					
15:30-16:00	Concurrent Session 3: The Future of Stem Cells in Reproduction <i>Chairs: Dr Sheila Loh Kia Ee (SG); Dr Budi Wiweko (INDO)</i> Just How Promiscuous Are Cells In Vitro? <i>Prof Gerald Schatten (USA); Making Oocytes and Babies from Stem Cells</i> <i>A/Prof Katsuhiko Hayashi (JPN); Aspect of Stem Cell-Based Therapies</i> <i>Dr Kwang Yul Cha (KOR)</i> (Venue: Great Hall Door 6)	Special Symposium 1: ASRM Symposium Precision <i>Chairs: Prof Bruno Lunenfeld (ISR); Prof Yoshiharu Morimoto (JPN)</i> A Systems Biology Approach to Diagnosing and Staging Endometriosis <i>Prof Linda Giudice (USA); The Male with Infertility or Reproductive Failure</i> <i>Dr Craig Niederberger (USA); Management of Hypothalamic Amenorrhea</i> <i>Dr Richard Reindollar (USA)</i> (Venue: Great Hall Door 8)	Free Communication 7: Nursing Counselling, Ethics, Psychology <i>Chairs: Ms Louise Younger (AUS); Ms Cailin Jordan (AUS)</i> (Venue: Meeting Room P1)	Free Communication 8: Genetics <i>Chairs: Dr Michelle Fraser (AUS); Prof Don Leigh (AUS)</i> (Venue: Meeting Room P2)	Free Communication 9: ART Basic, Embryology and Laboratory <i>Chairs: Dr Osamu Okitsu (JPN); Ms Jacquelyn Irving (AUS)</i> (Venue: Meeting Room M2)	"How to ... Session 5": 15:30-16:15 How to do IVF Successfully <i>Dr Ho Manh Tuong (VIET)</i> <i>Chair: Prof Roger Hart (AUS)</i> (Venue: Meeting Room M1)
16:00-16:30						
16:30-17:00						"How to ... Session 6": 16:15-17:00 How to Assess Ovarian Reserve <i>Dr Andon Hestiantoro (INDON)</i> <i>Chair: Prof William Ledger (AUS)</i> (Venue: Meeting Room M1)
17:00-17:30	Plenary Lecture 2: Ian Johnston Memorial Lecture <i>Chairs: Dr Mark Bowman (AUS); Dr Lyndon Hale (AUS)</i> The Future of Clinical Trials <i>Dr Richard Legro (USA)</i> (Venue: Great Hall Door 6 & 7)					
17:30-18:30	Poster Session / Mini Oral Presentations 2 <i>Chairs: Prof Nao Suzuki (JAP); Dr Clare Boothroyd (AUS)</i> (Venue: Great Hall Door 1)			Panel Session: Individualized Management in Patients Undergoing ART (By Invitation) <i>Chair: Prof YM Choi (KOR)</i> <i>Panelists:</i> <i>IVM in PCOS - Dr YS Kim, Dr MJ Kim; COS in PCOS - Dr NY Kim, Dr AM Park; COS in Endometriosis - Dr JS Koo, Dr YB Choi; COS in Poor Responder - Dr CY Hur, Dr HY Kim; Repeated Implantation Failure - Dr KS Lim</i> (Venue: Meeting Room M5)		
19:00-21:30	Social Function: Faculty Reception (By Invitation only) (Venue: Queensland Art Gallery, Gallery of Modern Art (GOMA))					

PROGRAMME AT A GLANCE

TIME	SATURDAY, 05 APRIL			
07:00-08:15	Breakfast Symposium Local Experience & Patient Acceptance of Elonva in Clinical Practice (Sponsored by: MSD) <i>Chair: Dr Mary Birdsall (NZ)</i> <i>Dr Devora Lieberman (AUS)</i> (Venue: Meeting Room M1)		FSA Preconception Health SIG Meeting (No Breakfast will be served) (Venue: Meeting Room M2)	
08:30-09:00	Concurrent Session 4: The Future Of Fertility Interventions <i>Chairs: A Prof Anusch Yazdani (AUS);</i> <i>Dr Craig Niederberger (USA)</i> Are We Doing Too Much IVF? <i>Dr Ben Willem Mol (NL)</i> (Venue: Great Hall Door 6)	Concurrent Session 7: Development of Larger IVF Organisations. Good or Bad? <i>Chairs: Dr Andrew Murray (NZ);</i> <i>Prof Vladislav Korsak (RUS)</i> An Indian Perspective <i>Dr Manish Banker(IND);</i> A Malaysian Perspective <i>Dr Haris Hamzah (MALAYSIA);</i> An Australian Perspective from a Large Organisation <i>A/Prof Peter Illingworth (AUS);</i> An Australian Perspective from a Small Organisation <i>Dr Clare Boothroyd(AUS)</i> (Venue: Great Hall Door 8)	Free Communication 10: Ovary & Art <i>Chair: Prof Hyuck Dong Han (KOR);</i> <i>Dr Debra Gook (AUS)</i> (Venue: Meeting Room M2)	"How to ... Session 7": 09:00-09:45 How to Determine the Best Culture Medium <i>Dr Cecilia Sjoblom (AUS)</i> <i>Chair: Prof Marcos Meseguer (SPN)</i> (Venue: Meeting Room M1)
09:00-09:45	Concurrent Session 5: The Future Of Uterine Transplantation <i>Prof Mats Brännström (SWE)</i> (Venue: Great Hall Door 6)			
09:45-10:30	Concurrent Session 6: The Future Of Psychosocial Interventions In Infertility <i>Dr Petra Thorn (GER)</i> (Venue: Great Hall Door 6)			"How to ... Session 8": 09:45-10:30 How to Evaluate Success with PGS <i>Prof Don Leigh (AUS)</i> <i>Chair: Dr Colin Lee (MY)</i> (Venue: Meeting Room M1)
10:30-11:00	Coffee Break and Exhibition Viewing (Venue: Great Hall Door 1)			
11:00-11:30	Concurrent Session 8: The Future Of Contraception <i>Chairs: Dr Devora Lieberman (AUS);</i> <i>Prof Gab Kovacs (AUS)</i> Reducing unintended pregnancies in Australia- Leveraging UK experience with Long Acting Reversible Contraceptives <i>Dr Paula Briggs (UK);</i> Physiology of reproduction/ biochemistry of steroids in the Pill <i>Prof Gab Kovacs (AUS);</i> Vaginal contraception-back to the future <i>Dr Terri Foran (Aus)</i> (Venue: Great Hall Door 6)	Industry Sponsored Symposium 4: Patient Centered ART: Individualising Treatment to Optimise Outcomes (Sponsored by Ferring) <i>Chairs: Prof Pak Chung Ho (HK);</i> <i>Prof Robert Norman (AUS)</i> AMH: The Beginning of the Start <i>Dr Richard Fleming (UK);</i> Biomarkers, Evolution of use in Ovarian Stimulation - The Asia-Pacific Experience <i>Dr Vuong Thi Ngoc Lan (VIET);</i> Biomarkers, My Use and Your Use to Individualise Ovarian Stimulation <i>Dr Richard Paulsen (USA)</i> (Venue: Great Hall Door 8)	Free Communication 11: Male Fertility <i>Chairs: Dr Aisaku Fukuda (JPN);</i> <i>Dr Virgilio (Joy) Novero (PHIL)</i> (Venue: Meeting Room P1)	Free Communication 12: Endometriosis <i>Chairs: Dr Asma Munir (UAE);</i> <i>Dr Colin Howles (SWITZ)</i> (Venue: Meeting Room P2)
11:30-12:00				"How to ... Session 9": 11:00-11:45 How to Set Up a TQM Programme <i>Dr John Peek (NZ)</i> <i>Chair: Dr Su Ling Yu (SG)</i> (Venue: Meeting Room M1)
12:00-12:30				"How to ... Session 10": 11:45-12:30 How to Set Up a Fertility Register in Emerging Markets <i>Dr Narendra Malhotra (IND)</i> <i>Chair: Prof Elizabeth Sullivan (AUS)</i> (Venue: Meeting Room M1)
12:30-13:30	Lunch and Poster Sessions / Mini Oral Presentations 3 <i>Chairs: Dr Narendra Malhotra (IND);</i> <i>Dr Devora Lieberman (AUS)</i> (Venue: Great Hall Door 1)		FSA REACT Group SIG Meeting (Venue: Meeting Room M2)	LIGHT Study Update Meeting 12:30 - 13:00 (Venue: Meeting Room M1)

PROGRAMME AT A GLANCE

13:30-14:00	Concurrent Session 9: The Future Of Fertility Preservation <i>Chairs: Prof Mats Brännström (SWE); Prof Nao Suzuki (JAP)</i> Current topics on fertility preservation of cancer patient- ovarian tissue vitrification <i>Dr Nao Suzuki (JPN); Ovarian Stimulation Protocols in the Cancer Patients</i> <i>Dr Jung Ryeol Lee (KOR); Australian Experience</i> <i>Dr Kate Stern (AUS)</i> (Venue: Great Hall Door 6)	Concurrent Session 10: The Future of Reproductive Sciences <i>Chairs: Prof Jie Qiao (CHN); Prof Chii-Ruey Tzeng (TAIWAN)</i> New Concepts on the Origin of PCOS <i>Prof Ray Rodgers (AUS); Exosomes: A New Paradigm for Embryo-Maternal Interactions in Establishing Pregnancy</i> <i>Prof Lois Salomonsen (AUS); A Mechanism Explaining Why Peri-Conception Maternal Hyperglycaemia Affects Oocytes and Embryos</i> <i>A Prof Jeremy Thompson (AUS)</i> (Venue: Great Hall Door 8)	Free Communication 13: Nursing Counselling, Ethics, Psychology <i>Chairs: Dr Michael Condon (AUS); A Prof Sheryl de Lacey (AUS)</i> (Venue: Meeting Room P1)	Free Communication 14: ART, Clinical <i>Chairs: Dr Kanthi Bansal (IND); Dr Pratap Kumar (IND)</i> (Venue: Meeting Room P2)	Free Communication 15: Genetics <i>Chairs: Dr Julia Loginova (RUS); Dr Richard Reindollar (USA)</i> (Venue: Meeting Room M2)	"How to ... Session 11": 13:30-14:15 How to Lower Costs in an IVF Programme <i>Dr David Molloy (AUS)</i> <i>Chair: Prof Alan Trounson (USA)</i> (Venue: Meeting Room M1)
14:00-14:30						
14:30-15:00						"How to ... Session 12": 14:15-15:00 How to do Double Stimulation and Egg Recovery for Poor Responders <i>Dr Kuang Yanping (CHN)</i> <i>Chair: A/Prof John McBain (AUS)</i> (Venue: Meeting Room M1)
15:00-15:30	Coffee Break and Exhibition Viewing (Venue: Great Hall Door 1)					
15:30-16:00	Special Symposium 2: Access and iCSI Symposium - The Future of Patient Centred ART <i>Chairs: Prof Linda Giudice (USA); Ms Sandra Dill (AUS)</i> Patient-Centred ART - A Japanese Clinic Perspective <i>Prof Yoshiharu Morimoto (JAP); Patient-Centred ART - A Japanese Consumer Perspective</i> <i>Ms Mikae Ueda (JAP); Patient-Centred ART - A Male Consumer Perspective</i> <i>Mr David Rawlings (AUS); Patient Centred ART in Resource-Constrained Settings</i> <i>Prof Alan Trounson (USA)</i> Panel Discussion (Venue: Great Hall Door 6)	Special Symposium 3: ISAR Symposium - Endoscopy: Ultrasound at its best in Fertility Enhancement <i>Chairs: Dr Manish Banker (IND); Prof Narendra Malhotra (IND)</i> Laparoscopy <i>Dr Prakash Trivedi (IND); Hysteroscopy</i> <i>Dr Sunita Tendulwadkar (IND); "Which Patient? Which Surgery? For Maximising the Reproductive Outcome"</i> <i>Moderator: Prof Jaideep Malhotra (IND)</i> <i>Panelists: Dr Rekha Kurien (IND); Dr Kaberi Bannerji (IND); Dr Kuldeep Jain (IND); Dr Sonal Panchal (IND); Dr Sudha Prasad (IND); Dr Kanthi Bansal (IND)</i> (Venue: Great Hall Door 8)	Free Communication 16: ART Basic, Embryology and Laboratory <i>Chairs: Mrs Carolyn Hills (AUS); Dr Keith Harrison (AUS)</i> (Venue: Meeting Room P1)	Free Communication 17: ART, Clinical <i>Chairs: Dr Bhola Rijal (NPL); Dato' Dr Prashant Nadkarni (MYL)</i> (Venue: Meeting Room P2)	Free Communication 18: Female Infertility, Fertility Preservation <i>Chairs: Dr Haroon Latif (PAK); Dr Graeme Thompson (AUS)</i> (Venue: Meeting Room M2)	"How to ... Session 13": 15:30-16:15 How to do Ovulation Induction in Low Weight Individuals <i>Dr Vuong Thi Ngoc Lan (VIET)</i> <i>Chair: Dr Pratap Kumar (IND)</i> (Venue: Meeting Room M1)
16:00-16:30						
16:30-17:00						"How to ... Session 14": 16:15-17:00 How to Maximise Pregnancy Rates from an Embryo Transfer <i>Prof Teraporn Vutyavanich (THL)</i> <i>Chair: A/Prof Kelton Tremellen (AUS)</i> (Venue: Meeting Room M1)
17:00-18:00	ASPIRE Annual General Meeting (Venue: Great Hall Door 6 & 7)					
19:00-22:30	Social Function: ASPIRE Gala Dinner (*Ticketed) (Venue: Brisbane City Hall)					

PROGRAMME AT A GLANCE

TIME	SUNDAY, 06 APRIL		
08:30-09:00	Concurrent Session 11: The Future Of PCOS <i>Chairs: Dr Benjamin Kroon (AUS); Prof Helena Teede (AUS)</i> PCOS in Asian Patients; <i>Prof Jie Qiao (CHN); Health Implication of PCOS in Australia; Prof Roger Hart (AUS); PCOS in South Asians; Dr Abha Majumdar (IND)</i> (Venue: Great Hall Door 6)	Concurrent Session 12: The Future For Preconception Planning: What The Patient Should Know About Preconception Care <i>Chairs: Ms Louise Johnson (AUS); Dr Karin Hammarberg (AUS)</i> Preconception health – what the clinician should talk about with patients <i>Prof Tony Chung (HK);</i> Preconception health – research and action at a popula- tion level <i>Dr Karin Hammarberg (AUS); Dr Kamini Rao (IND);</i> Preconception Care in Developing Countries <i>Panel Discussion Prof Tony Chung (HK); Dr Karin Hammarberg (AUS); Dr Kamini Rao (IND); Ms Cailin Jordan (AUS); Dr Antony Lighten (AUS)</i> (Venue: Great Hall Door 8)	Free Communication 19: ART, Clinical <i>Chairs: Dr Sabina Shrestha (NPL); Prof Tongtis Tonyai (THAI)</i> (Venue: Meeting Room M1)
09:00-09:30			
09:30-10:00			
10:00-10:30	Coffee Break and Exhibition Viewing (Venue: Great Hall Door 1)		
10:30-11:00	Concurrent Session 13: The Future Treatment Of Endometriosis <i>Chairs: Dr David Molloy (AUS); Prof Zi-Jiang Chen (CHN)</i> Advances in Clinical Management <i>A/Prof Luk Rombauts (AUS); Biomarkers of Endometriosis Prof Chii-Ruey Tzeng (TAIWAN); Genetics of Endometriosis Dr Jenny Fung (AUS)</i> (Venue: Great Hall Door 6)	Concurrent Session 14: The Future Of Andrology Symposium <i>Chairs: Dr Derek Lok (AUS); Dr Craig Niederberg (USA)</i> Assessing the Male <i>Dr Craig Niederberg (USA); Sperm Extraction in ART Dr Ju Tae Seo (KOR); Hormonal Approaches to Male Fertility Regulation Prof Robert McLachlan (AUS)</i> (Venue: Great Hall Door 8)	Free Communication 20: ART Basic, Embryology and Laboratory <i>Chairs: Dr Aisaku Fukuda (JPN); Dr Deirdre Zander-Fox (AUS)</i> (Venue: Meeting Room M1)
11:00-11:30			
11:30-12:00			
12:00-13:00	Lunch and Poster Session (Venue: Great Hall Door 1)		
13:00-14:00	Late-Breaking Lectures What is the most cost-effective gonadotropin treatment for poor responders? <i>Dr Virochana Kaul (AUS);</i> Genome and transcriptome analyses of single human oocytes and preimplantation embryos <i>Prof Jie Qiao (CHN)</i> (Venue: Great Hall Door 6 & 7)		
14:00-14:30	Special Symposium 4: ASPIRE in 2014 onwards <i>Chairs: Prof Bruno Lunenfeld (ISRAEL); Prof PC Wong (SG)</i> <i>Dr Jaideep Malhotra (IND)</i> (Venue: Great Hall Door 6 & 7)		
14:30-15:00	Closing Ceremony (Venue: Great Hall Door 6 & 7)		

MINI ORAL AND POSTER PRESENTATIONS

All mini oral and poster presenters are required to display a paper poster. Posters are displayed as per the poster numbers indicated in the programme. Poster presenters are requested to stand by their posters during tea breaks and on Friday evening during the dedicated poster viewing time. Poster presenters are to adhere to the poster set up and dismantling schedule below. Please remove your posters within the specified dismantling period. Organisers will not be responsible for posters that are not removed on time.

Location: Poster area located in Exhibition Hall (Great Hall Door 1)
Poster Set-up: Friday, 4 April 2014, 8:00am onwards
Poster Dismantling: Sunday, 6 April 2014, from 1:00pm – 3:00pm

For mini oral presenters, kindly report to the mini oral presentation area located in the Exhibition Hall (Great Hall Door 1) at least 30 minutes before your allocated time slots to submit your slides.

SCIENTIFIC PROGRAMME

Thursday-Friday, 3-4 April 2014





From Conception to Delivery

At Ferring Pharmaceuticals, we believe in the power of research and sharing of knowledge. Our research-driven approach and ongoing commitment has led to a portfolio of Reproductive Health products that can assist couples to conceive and complete a successful pregnancy. We are passionate about every stage of the reproductive cycle...from conception to delivery.



Reproductive Health
INFERTILITY, OBSTETRICS AND GYNAECOLOGY

Thursday, April 03, 2014

Industry Sponsored Symposium 1 - New Perspectives on IVM (Cook Medical)

13:30 - 16:00

Meeting Room M1

Chair: Jeremy Thompson (Australia)

Speakers: Jeremy Thompson (Australia)

Rob Gilchrist (Australia)

Michel De Vos (Belgium)

Welcome Reception

18:00 - 20:30

Great Hall Door 6,7; Mezzanine Level

Friday, April 04, 2014

Opening Ceremony

08:30 - 08:50

Great Hall Door 6 & 7

08:30 **Introduction**

Speaker: Robert Norman (Australia)

08:40 **Introduction**

Speaker: Clare Boothroyd (Australia)

Opening Lecture

08:50 - 09:30

Great Hall Door 6 & 7

Chairs: Robert Norman (Australia)

Jaideep Malhotra (India)

08:50 **Vaccines and Reproduction: Where are we heading?**

Speaker: Ian Frazer (Australia)

Vaccination to prevent infection in the genital tract has been predicted to be a challenge, as protective mucosal immunity is held to be short lasting. However, the field effectiveness over 8 years of the HPV vaccines in prevention of HPV associated disease in Australia suggests that other genital infections including Herpes and Chlamydia may also be susceptible to preventative immunisation. Vaccination to control fertility in animals has proven effective but acceptability in humans depends on regulation of duration of control, which is not yet feasible. Therapeutic vaccines for persisting genital tract infections are proving less tractable, with no successes from over 20 years of research effort. Possible reasons for this will be discussed.

Keynote Lecture

09:30 - 10:00

Great Hall Door 6 & 7

Chairs: Robert Norman (Australia)
 Jaideep Malhotra (India)

09:30 **The Future of Stem Cells**
 Speaker: Alan Trounson (USA)

Stem cell discoveries have revolutionized human medicine. Already there is a raft of cell therapies in clinical trials that provide benefit for blood and immune disorders, bone repair, osteoarthritis, graft versus host disease, cardiovascular disease and many others. Trials that are also moving forward through translational medicine research include demyelination diseases in children, cures for genetic diseases such as β thalassemia hemophilia and sickle cell disease, reversal of blindness, cure for HIV AIDS, Cure for type I diabetes, repair of damaged heart tissue, development of hyaline articulated cartilage for joint repair and many others. Destruction of dangerous cancer stem cells using small biologics, monoclonal antibodies and T cell activation strategies are changing oncology options and treatments. Stem cells will also help define the genetic cause for disease and susceptibility to pathogenic infections using induced pluripotent stem cells (iPSCs). Human disease modeling using iPSCs will enable new targets and drugs to be developed for many conditions. It is expected that universal donor cells and tissues will be available for transplantation, using HLA haplotyped iPSCs and genetically modified cell types that escape immune rejection. This has all happened in a very brief decade of research that has made a revolution in regenerative medicine.

Plenary Lecture

10:00 - 10:30

Great Hall Door 6 & 7

Chairs: Robert Norman (Australia)
 Jaideep Malhotra (India)

10:00 **Infertility As A Global Health Problem**
 Speaker: Chittaranjan Purandare (India)

No Summary Provided

Coffee Break and Exhibition Viewing

10:30 - 11:00

Great Hall Door 1

Concurrent Session 1 - The Future of Genetics and Environment in Determining Disease in Reproduction

11:00 - 12:30

Great Hall Door 6

Chairs: Shin Yong Moon (Korea)
 Linda Giudice (USA)

11:00 GWAS and post-GWAS of PCOS
 Speaker: Zi-Jiang Chen (China)

As a complex disorder, PCOS is present with heterogeneous characteristics originating from the combined effect of both environmental and genetic factors. On the genetic aspects, a series of family and twin studies have demonstrated the heritable nature of PCOS. Meanwhile, several researchers have tried to find the link between certain genes and PCOS. Recently, the whole-genome association studies (GWASs) of PCOS provide more reliable and unbiased results and represent a milestone for genetic studies in PCOS. However, the greatest challenges of the post-GWAS are just in the front to explore the functional consequences of the GWAS susceptibility loci.

11:30 Genetics of Menopause
 Speaker: Bart Fauser (Netherlands)

Age at menopause appears to a remarkably robust phenomenon when different populations around the world or possible differences over time are studied, suggesting a strong genetic component. Despite the major individual variability in normal menopause between 40 and 60 years of age, a strong association of menopausal age between mother and daughter has also been demonstrated.

Individual variability in age of menopause has been demonstrated to be associated with age of preceding decreased natural fertility, along with implications for long-term health such as breast cancer risk and changes for cardiovascular disease, Alzheimer's disease and stroke.

Age of menopause should be considered a complex genetic trait, and a recent meta-analysis of 22 genome wide association studies in almost 40,000 women identified 13 relevant loci in genes also involved in more general mechanisms such as DNA repair and immune function. Such technologies may also help to disclose novel information regarding the genetic background of menopause before age 40 (premature ovarian insufficiency (POI)).

12:00 Pharmacogenetics of ART
 Speaker: Young Min Choi (Korea)

In spite of attempts to standardize controlled ovarian hyperstimulation (COH) regimens for women undergoing ART procedure, we commonly experience either low ovarian response or high ovarian response leading to ovarian hyperstimulation syndrome. This individual variability in ovarian response necessitates the use of predictive markers of ovarian hyper- or hypo-response of corresponding COH cycles. In animal studies, it has been shown that ovarian response to gonadotropin stimulation is influenced by genetic factors. Recently, several researchers have tried to reveal genetic factors involved in the COH for ART. Several gene polymorphisms, including FSH receptor (FSHR), and estrogen receptor alpha gene, have been reported to be associated with the outcomes of COH for ART, with the inconsistent findings in different studies. Further studies using a larger number of patients in diverse ethnic populations are needed to elucidate the genetic factors involved in the COH for ART.

Industry Sponsored Symposium 2 - Cracking the Code: Biomarkers in Embryo Selection and Uterine Receptivity (Merck Serono)

11:00 - 12:30

Great Hall Door 8

Chair: David K. Gardner (Australia)

11:05 Innovation in ART

Speaker: Diego Ezcurra (India)

11:25 Utilization of Electrochemiluminescence immunoassay (ECLIA) for the detection of early embryonic biomarkers for embryo selection

Speaker: Jie Li (China)

11:55 Uterine receptivity: The Final Hurdle in IVF

Speaker: Lois Salamonsen (Australia)

Free Communication 1 - ART Basic, Embryology and Laboratory

11:00 - 12:30

Meeting Room P1

Chairs: Helena Jericho (Australia)

Cecilia Sjoblom (Australia)

11:00 FC001

ANALYSIS OF BOTH MORPHOKINETICS AND BLASTOCYST METABOLISM TO DEVELOP A COMBINED QUANTITATIVE PREDICTOR OF VIABILITY

Y.S. Lee, G.A. Thouas, D.K. Gardner (Australia)

11:15 FC002

DNA DAMAGE AND VACUOLES: WHEN DOES SPERM VACUOLATION BECOME IMPORTANT?

C. Knight, S. Cooke, P. Illingworth, M. Chapman (Australia)

11:30 FC003

USE OF HIGH CALCIUM MEDIUM ON PATIENTS FOLLOWING POOR OR FAILURE TO FERTILIZE ICSI CYCLES.

J. Robertson, B. Podsiadly, N. Hobson, S. McArthur (Australia)

11:45 FC004

HIGH FERTILIZATION AND LOW ZYGOTE ARREST RATES ARE INDICATORS OF GOOD LABORATORY PRACTICE IN ASSISTED REPRODUCTION TECHNOLOGY

J. Ali (Malaysia)

12:00 FC005

GETTING TO ELECTIVE SINGLE EMBRYO TRANSFER WITH BLASTOCYST CULTURE AND PREIMPLANTATION GENETIC SCREENING FOR PATIENTS OF ALL AGES

J. Conaghan, E. Fischer (USA)

12:15 FC006

TIME LAPSE IMAGING AND GENE EXPRESSION PROFILING IN CUMULUS CELLS AS PREDICTORS OF BLASTOCYST QUALITY

E. Hammond, B. Stewart, R. Dominick, L. Cree (New Zealand)

Free Communication 2 - Nursing Counselling, Ethics, Psychology

11:00 - 12:30

Meeting Room P2

Chairs: Donna Close (Australia)
Petra Thorn (Germany)

- 11:00 **FC007**
MOTIVATIONS AND EXPERIENCES OF PATIENTS SEEKING CROSS BORDER REPRODUCTIVE CARE: THE AUSTRALIAN AND NEW ZEALAND CONTEXT
I. Rodino, S. Goedeke, S. Nowowski (Australia)
- 11:15 **FC008**
AN EXAMINATION OF RECIPIENTS' EXPERIENCE OF INTRA-FAMILIAL SPERM DONATION
C. Singleton, E. Zwahlen (Australia)
- 11:30 **FC009**
HEALTH NUMERACY AND RISK AVERSION IN INFERTILE PATIENTS
L.J. Rombauts, L. Larmour, E. Wallace, A. Aliabadi, C. Motteram, D. De Guingand (Australia)
- 11:45 **FC010**
ASSISTED CONCEPTION, MATERNAL AGE AND BREASTFEEDING: AN AUSTRALIAN COHORT STUDY
J. Fisher, K. Hammarberg, K. Wynter, J. McBain, F. Gibson, J. Boivin, C. McMahon (Australia)
- 12:00 **FC011**
INFERTILITY INFORMATION: ACCESS, ACCURACY AND LIABILITY IN INDONESIA, NOVEMBER 2013
I. Kusumaningtyas (Indonesia)
- 12:15 **FC012**
DISTRIBUTION OF LEVEL OF STRESS AMONG INFERTILITY PATIENTS
B. Wiweko, U. Anggraheni, S.D. Erfira (Indonesia)

Free Communication 3 - Contraception, Endocrinology, Laboratory

11:00 - 12:30

Meeting Room M2

Chairs: Anna Smirnova (Russia)
Robert Lahoud (Australia)

- 11:00 **FC013**
THE IMPACT OF ORAL CONTRACEPTIVES ON COGNITION
A. Warren, C. Gurvich, R. Worsley, J. Kulkarni (Australia)
- 11:15 **FC014**
CLOMIPHENE CITRATE, METFORMIN OR THE COMBINATION OF BOTH, AS FIRST LINE OVULATION INDUCTION DRUG IN POLYCYSTIC OVARIAN SYNDROME ; A RANDOMISED CONTROLLED TRIAL.
S. Kar (India)
- 11:30 **FC015**
EFFECT OF SECOND GENERATION COCS ON SERUM LIPID PROFILES, FASTING BLOOD SUGAR, BLOOD PRESSURE AND BMI IN CHILD BEARING AGE WOMEN IN KHYBERPUKHTUNKHWA PROVINCE-PAISTAN.
R. Nazli, N. Sher Mohammad, M. Khan, T. Akhtar, Z. Zafar (Pakistan)
- 11:45 **FC016**
HYPOTHALAMIC GHRH AND PITUITARY GH GENES EXPRESSION LEVELS IN NEONATAL INHIBIN-IMMUNONEUTRALIZED FEMALE RATS
J. Al-Sa'aidi, H. Thanoon (Iraq)

- 12:00 **FC017**
SERUM SAMPLE PREPARATION FOR AMH MEASUREMENT WITHIN AN EXTERNAL QUALITY ASSURANCE (EQA) SCHEME: SAMPLE STABILITY UNDER DIFFERENT STORAGE CONDITIONS.
E. Zuvela, P. Matson (Australia)
- 12:15 **FC018**
IS PREMIXING OF SERUM SAMPLES NECESSARY TO OBTAIN REPRODUCIBLE AMH RESULTS USING THE ANSHLAB ASSAY
M. McShane, X. Han, C. White, W. Ledger (Australia)

"How to ... Session 1" - How to Plan and Participate in a Clinical Trial

11:00 - 11:45

Meeting Room M1

Chair: Ernest Ng (Hong Kong, China)

11:00 **How to Plan and Participate in a Clinical Trial**

Speaker: Neil Johnson (New Zealand)

How should we plan and participate in a clinical trial in reproductive medicine in 2014? The short answer is "Collaboratively"!

Key factors in the planning phase are to choose a clinical question of irrepressible interest to a wide audience, for which there is individual and collective equipoise. Multidisciplinary input and international consensus are helpful in refining specific trial proposals. It is crucial that any trial should have adequate power to answer the clinical question and a high feasibility of completion. The CONSORT Statement outlines the markers of trial quality and these are as important for participating researchers as for those assessing the worth of the research once completed. Participation is governed by numerous factors, but a critical component of successful trial participation is that work in the clinical trial must fit satisfactorily into the research collaborator's work pattern. The many determinants of success or failure of a trial, from the planning and participation perspectives, will be discussed.

11:30 **Discussion**

"How to ... Session 2" - How to Understand GnRH Control

11:45 - 12:30

Meeting Room M1

Chair: Louise Hull (Australia)

11:45 **How to Understand GnRH Control**

Speaker: Bruno Lunenfeld (Israel)

Gonadal sex steroids provide feedback signals to the brain-pituitary axis to maintain gonadotropin secretion within homeostatic boundaries, which supports normal gonadal function. Emerging evidence supports the idea that KiSS-1 peptide, the natural ligand of GPR54 stimulates LH and FSH secretion by governing GnRH pulse amplitude and frequency. The *Kiss1* gene is a target for regulation by gonadal steroids (e.g., estradiol and testosterone) as well as to changes in energetic resources (e.g. fasting) or signals of energy availability (e.g. leptin) and allows for adjustments of the kisspeptin in response to real-time changes in energy availability. *Kiss1* neurons in the arcuate nucleus (ARC) are implicated in the sex steroid-dependent negative feedback control of gonadotropin secretion, whereas *Kiss1* neurons in the anteroventral periventricular nucleus (AVPV) may be involved in generating the preovulatory GnRH/LH. In infertility due to hypothalamic amenorrhoea acute administration of kisspeptin results in stimulation of reproductive hormones.

12:15 **Discussion**

Lunch and Poster Sessions / Mini Oral Presentations 1

12:30 - 13:30

Great Hall Door 1

Chairs: Gab Kovacs (Australia)
Young Min Choi (Korea)

12:45 **M001**

NATURAL KILLER CELL ANALYSIS: FROM BLOOD, UTERUS, BOTH OR NOT AT ALL?

G. Sacks, N. Varnier, P. Russell (Australia)

12:50 **M002**

EPIDEMIOLOGICAL ANALYSIS OF THE RISK FACTORS AND THE PERIOD FOR ONSET OF OVARIAN INSUFFICIENCY AFTER SURGERY

S. Takae, K. Kawamura, Y. Sato, C. Nishijima, N. Yoshioka, Y. Sugishita, Y. Horage, M. Tanaka, B. Ishizuka, N. Suzuki (Japan)

12:55 **M003**

CLEAVAGE KINETICS ANALYSIS IS HELPFUL TO PREDICT THE BLASTOCYST TRACEABILITY

T. Akiyoshi, S. Okamoto, K. Matsuo, Y. Mizobe, S. Minami, M. Nishigaki, A. Matsuzaki, A. Yamaguchi (Japan)

13:00 **M004**

DEVELOPMENT OF A 5-POINT EMBRYOSCOPE ALGORITHM PREDICTIVE OF GOOD BLASTOCYST GRADE.

A. Storr, S. Cooke, L. William (Australia)

13:05 **M005**

MORPHOLOGICAL DIFFERENCES BETWEEN EMBRYOS TREATED WITH IN VITRO MATURATION AND ROUTINE IVF DETECTED WITH TIME LAPSE MONITORING INCUBATOR

P.A. Iffanolida, Y. Handoko, K. Mutia, A. Bowolaksono, E. Mansyur, B. Wiweko (Indonesia)

13:10 **M006**

LIVE BIRTH RATES OF EARLY STAGE BLASTOCYSTS CULTURED OVERNIGHT FOLLOWING VITRIFICATION AND WARMING.

B. Podsiadly, T. Roy, K. Waite, S. McArthur, M. Bowman (Australia)

Meeting: FSA IVF Directors' Meeting (By Invitation)

12:30 - 13:30

Meeting Room M2

Concurrent Session 2 - The Future Of Embryology

13:30 - 15:00

Great Hall Door 6

Chairs: Colin Howles (Switzerland)
Phillip Matson (Australia)

13:30 **Video Microscopy**

Speaker: Yasuyuki Mio (Japan)

Since our original development of the in vitro culture system for time-lapse cinematography (TLC) one decade ago, a number of studies have been analyzed. 1) the dynamic morphology from the fertilization process up to the hatched blastocyst stage 2) the demonstration of the precise time course of the fertilization process and thereafter embryonic development, 3) the appearance of the fertilization cone at the sperm entry point, 4) the way of fragmentation 5) the discovery of several novel phenomena: the extrusion of the third polar body involved in zygotes with single pronucleus or uneven two pronuclei; the splitting of ICM (Strand) during blastocyst stage; and the disturbance of trophoctoderm related to blastocoel collapse. Recently, we have found that there were two patterns in the hatching (inward and outward). In this presentation, I would like to focus on the hatching pattern of expanded blastocysts and the relationship between the hatching pattern and the blastocoel collapse.

14:00 **Videomicroscopy - Clinical Results**

Speaker: Marcos Meseguer (Spain)

We have been working in the last 5 years determining if incubation in the integrated EmbryoScope(TM) time-lapse monitoring system and selection supported by the use of a multivariable morphokinetic model improves the reproductive outcome in comparison with incubation in a standard incubator embryo culture and selection based exclusively on morphology. We present results from our randomized controlled trial over 900 patients where ongoing pregnancy rate was significantly increased, supporting the strategy of culturing and selecting embryos in the time-lapse monitoring system. The main limitation in this trial was that we did not know how much of this improvement was due to the culture conditions since the system does not allow us to differentiate. We carried out the study conscientiously and assuming that the improvement was down to both culture conditions and the selection model. The present study prospectively demonstrated the improvement in the reproductive outcome by using this time-lapse system and a set of selection and deselection criteria based on embryo morphokinetics.

14:30 **Latest in Embryo Metabolomics**

Speaker: Hsin-Fu Chen (Taiwan)

In assisted reproduction, selection of good embryos for transfer are in most centers based on the morphology of embryos. Although usually faithfully serving as an indicator for overall quality of the embryo and also the IVF lab, it is accepted that morphology alone may not be sensitive enough to identify the most optimal embryo, especially for the purpose of eSET. In recent years, other novel methods including metabolomic profiling of spent medium, time-lapse imaging of embryo growth and invasive procedures as preimplantation genetic screening/diagnosis (PGS/PGD) of the embryos are proposed to be potentially useful strategies to improve the overall efficiency of embryo selection. Among them, the clinical role of embryo metabolomics has remained highly disputable. In this talk I will present some of the data we obtained recently about this issue and will discuss in detail how we should look at the practical role of embryo metabolomics at this moment.

Industry Sponsored Symposium 3 - All Women At 30 YO That Are Not In A Relationship Should Freeze Their Eggs (MSD)

13:30 - 15:00

Great Hall Door 8

Chair: Bart Fauser (Netherlands)

Debaters: Devora Lieberman (Australia)

Kelton Tremellen (Australia)

Free Communication 4 - Nursing Counselling, Ethics, Psychology

13:30 - 15:00

Meeting Room P1

Chairs: Carmel Carrigan (Australia)

Helen Alvino (Australia)

13:30 **FC019**

HEALTH NUMERACY IN FERTILE AND INFERTILE POPULATIONS

L. Larmour, E. Wallace, A. Aliabadi, C. Motteram, D. De Guingand, L.J. Rombauts (Australia)

13:45 **FC020**

GAMETE DONORS' EXPECTATIONS AND EXPERIENCES OF CONTACT WITH THEIR DONOR OFFSPRING

M. Kirkman, K. Bourne, L. Johnson, J.R.W. Fisher, K. Hammarberg (Australia)

14:00 **FC021**

GENETICS, DUPED DADS AND DISCARDED CHILDREN

S. Dill (Australia)

14:15 **FC022**

OLDER MATERNAL AGE, ASSISTED CONCEPTION AND OBSERVED PARENTING QUALITY IN MOTHERS OF TODDLERS.

C.A. McMahon, S. Berry, F.L. Gibson (Australia)

14:30 **FC023**

IS THE BEST INTERESTS OF THE CHILD A USEFUL PRINCIPLE? EMERGING ISSUES IN ART PRACTICE.

S. De Lacey, J. McMillan (Australia)

14:45 **FC024**

"HOW MUCH DO THEY KNOW WHAT THEY DO?" A LOCAL PERCEPTION STUDY TARGETED ON PATIENTS OF ASSISTED REPRODUCTIVE TECHNIQUE (ART) IN HK

L. Chan, M. Chan, C. Louis (Hong Kong, China)

Free Communication 5 - Ovary

13:30 - 15:00

Meeting Room P2

Chairs: Kyung-Ah Lee (Korea)
Ray Rodgers (Australia)

- 13:30 **FC025**
ANDROGEN-INDUCED MOUSE MODELS FOR POLYCYSTIC OVARY SYNDROME - WHICH ONE IS BEST?
K.A. Walters, A.S.L. Caldwell, L.J. Middleton, C.M. Allan, D.J. Handelsman (Australia)
- 13:45 **FC026**
THE EFFECTIVENESS OF HERBAL MEDICINE PLUS A LIFESTYLE INTERVENTION FOR OLIGO/AMENORRHOEA IN WOMEN WITH PCOS; A RANDOMISED CONTROLLED TRIAL.
S. Arentz, C.A. Smith, J. Abbott, A. Bensoussan (Australia)
- 14:00 **FC027**
A CROSS SECTIONAL STUDY OF POLYCYSTIC OVARIAN SYNDROME (PCOS) AMONG ADOLESCENT AND YOUNG GIRLS IN MUMBAI, INDIA
B. Joshi, S. Mukherji, A. Patil, A. Purandare, S. Chauhan, R.A.M.A. Vaidya (India)
- 14:15 **FC028**
SEX HORMONE BINDING GLOBULIN (SHBG) AS A PREDICTOR OF RESPONSE TO METFORMIN THERAPY IN POLYCYSTIC OVARIAN SYNDROME (PCOS)
K. Guleria, S. Miglani, S.V. Madhu, B.D. Banerjee, A. Suneja, N.B. Vaid (India)
- 14:30 **FC029**
DETERMINATION OF OVARIAN FOLLICLE LOCALIZATION IN PATIENTS WITH PRIMARY OVARIAN INSUFFICIENCY (POI)
T. Haino, W. Tarumi, T. Harada, K. Sugimoto, A. Okamoto, K. Kawamura, M. Ikegami, N. Suzuki (Japan)
- 14:45 **FC030**
HOW DOES FOLLICULAR GROWTH RATE AT DIFFERENT TYPE AND DOSES OF OVARIAN STIMULATION DIFFER? A RETROSPECTIVE STUDY
R. Pandurengan, N. Razali, J. Ali, M. Hamdan (Malaysia)

Free Communication 6 - Fertility Preservation, Male Fertility

13:30 - 15:00

Meeting Room M2

Chairs: Hari Kishor Shrestha (Nepal)
Iswaran Kumar Kularatnam (Malaysia)

- 13:30 **FC031**
REACTIVE OXYGEN SPECIES (ROS) LEVELS IN SEMEN FROM INFERTILE COUPLES
T.H.V. Huynh, T.C. Tran, T.L.T. Nguyen, T.M. Nguyen, T. Ho (Vietnam)
- 13:45 **FC032**
THE ANALYSIS OF APOPTOSIS OF FRESH VERSUS THAWED-VITRIFIED ISOLATED PRE-ANTRAL FOLLICLES
B. Wiweko, K. Mutia, E. Mansyur, A. Aulia, S. Soebijanto, B. Affandi, A. Boediono (Indonesia)
- 14:00 **FC033**
EFFECTS OF THREE DIFFERENT TYPES OF ANTIFREEZE PROTEINS ON MOUSE OVARIAN TISSUE CRYOPRESERVATION
J. Lee, E.J. Kim, H.S. Kong, H.W. Youm, J.R. Lee, C.S. Suh, S.H. Kim (Korea)
- 14:15 **FC034**
GENETIC VARIANTS IN THE ETV5 GENE IN FERTILE AND INFERTILE MEN WITH NON-OBSTRUCTIVE AZOOSPERMIA ASSOCIATED WITH SERTOLI CELL ONLY SYNDROME
D. Jamsai, P. Stahl, P. Schlegel, R. McLachlan, M. O'Bryan (Australia)

- 14:30 **FC035**
ALCOHOL CONSUMPTION AND SEMEN QUALITY: A RETROSPECTIVE, CROSS-SECTIONAL STUDY OF MEN PRESENTING FOR FERTILITY ANALYSIS.
R. Warren, P. Burton, T. Brett, M. Bulsara (Australia)
- 14:45 **FC036**
SPERM ANTIBODY INCIDENCE AND CYCLE OUTCOMES IN 1086 SAMPLES FROM COUPLES PRESENTING WITH INFERTILITY
H. Wills, J. Osborn, S. McDowell, B. Kroon, E. Ford, K. Harrison, J. Wolyncevic, J. Esler, A. Yazdani (Australia)

"How to ... Session 3" - How to Diagnose PCOS in Adolescence

13:30 - 14:15

Meeting Room M1

Chair: Helena Teede (Australia)

13:30 Speaker: Delfin A. Tan (Philippines)

Polycystic ovary syndrome (PCOS) is frequently diagnosed during adolescence. The clinical impact of adolescent PCOS is amplified by the recent trend of increasing obesity among teenagers. Affected girls are as insulin-resistant as their adult counterparts and are also at increased risk later in life for cardiovascular and metabolic disease. Early treatment may prevent disease progression. The diagnosis of PCOS in adolescence could be difficult because some features of the syndrome are physiologic at this age. None of the different definitions of PCOS (NICHD 1990, Rotterdam 2003, AE-PCOS Society 2009) has proposed criteria for the diagnosis of PCOS in the adolescent. It is suggested that menstrual irregularity for over two years, reduced reliance on ultrasound diagnosis of polycystic ovarian morphology, and accurate assessment of hyperandrogenic and metabolic features are suitable strategies for the diagnosis of PCOS in the adolescent.

14:00 **Discussion**

"How to ... Session 4" - How to Present a Paper at a Conference When English is Not Your First Language

14:15 - 15:00

Meeting Room M1

Chair: PC Wong (Singapore)

14:15 Speaker: Robert Norman (Australia)

As the quality of science and clinical investigation improves in our region, researchers will want to present more often at international conferences where the sole language is English. This is a challenge to many people whose first language is not English because of concerns about their ability to communicate in an understandable way. As a result, they often don't present at all or use a poster as a preference. In my talk, I will encourage you to be bold and think of applying for talks where you have good data. I will discuss how to present, what your slides should look like and how to anticipate and field questions that arise. Some obvious errors will be discussed and there will be opportunity for interactive discussion.

14:45 **Discussion**

Coffee Break and Exhibition Viewing

15:00 – 15:30

Great Hall Door 1

Concurrent Session 3 - The Future of Stem Cells in Reproduction

15:30 - 17:00

Great Hall Door 6

Chairs: Sheila Loh (Singapore)
 Budi Wiweko (Indonesia)

15:30 Just How Promiscuous Are Cells In Vitro?

Speaker: Gerald Schatten (USA)

With advances in cancer therapies, survival rates in prepubescent patients have steadily increased. However a number of these surviving patients have been rendered sterile. In addition, men and women, who are genetically fertile, can become infertile due to immune suppression treatments, exposure to environmental and industrial toxicants, and injury. Notwithstanding the great emotional burden from an inability to conceive a child with their partner, the financial burdens for testing and treatment are high. Recent advances in pluripotent stem cell differentiation and the generation of patient-specific, induced pluripotent stem cells indicate that stem cell replacement therapies or in vitro differentiation may be on the horizon. Here, we discuss these recent advances, their relevance to treating male-factor and female-factor infertility, and what experimental procedures must be carried out before these exciting new treatments can be used in a clinical setting.

16:00 Making Oocytes and Babies from Stem Cells

Speaker: Katsuhiko Hayashi (Japan)

Pluripotent stem cells, such as embryonic stem cells (ESCs) and induced pluripotent stem cells (iPSCs), are able to differentiate into all cell lineages of the embryo proper, including germ cells. Establishing the germ cell lineage from ESCs/iPSCs is the key biological subject, since it would contribute to production of unlimited numbers of functional gametes *in vitro*. Toward this goal, we recently established a culture system that induces functional mouse primordial germ cells (PGCs) from ESCs/iPSCs. PGCs produced from ESCs/iPSCs are fully potent, since they differentiate into oocytes, which in turn give rise to healthy individuals. There are, however, many obstacles to be overcome for the robust generation of mature gametes or for application of the culture system to other species, including humans and livestock. In the meeting, I will discuss recent data and perspectives of germ cell production *in vitro*.

16:30 Aspect of Stem Cell-Based Therapies

Speaker: Kwang Yul Cha (Korea)

CHA Stem Cell Institute (CHA Health Systems, Korea) has put an effort into developing an applicable human embryonic (hES)- and several adult stem cell therapies for a number of clinical studies with leading research institutes and companies in ACT. Inc and Pluristem Co., USA. About 20 clinical trials including Age Related Macular Degeneration and Stargardt's disease using hES-RPEs, Parkinson's disease and Amyotrophic Lateral Sclerosis using fetal midbrain derived neuronal progenitor cells (hNPCs), Cerebral Palsy and Muscle marasmus using umbilical cord blood cells (UCBCs), Intermittent Claudication using PLX-PAD cells produced from Pluristem Co., are going on in Korea or in some cases, both USA and in Korea. In addition to the above-mentioned trials, CHA Health Systems have developed Pla-and Cord-Stem, products consisting of expanded placenta and cord stem cells for allogeneic transplantation, which has been regarded as the effective treatment for various neural diseases, arthritis, and anti-aging. I will introduce ongoing clinical trials in CHA Stem Cell Institute.

Special Symposium 1 - ASRM Symposium Precision

15:30 - 17:00

Great Hall Door 8

Chairs: Bruno Lunenfeld (Israel)
 Yoshiharu Morimoto (Japan)

15:30 A Systems Biology Approach to Diagnosing and Staging Endometriosis
 Speaker: Linda Giudice (USA)

Millions of women and teens worldwide suffer from endometriosis, which is associated with pelvic pain and infertility and has a mean lag time to diagnosis of 7.8 years. Currently, surgery under general anesthesia and histologic evaluation of lesions biopsied at surgery comprise the gold standard of diagnosis. High throughput technologies involving proteomic analysis of blood, urine and endometrium, and the transcriptomic analysis of eutopic endometrium with subsequent bioinformatics and statistical analyses and programs have opened the door to non-surgical diagnoses and staging of peritoneal disease. This lecture will summarize the current state of the art towards non-surgical diagnostics and staging of endometriosis in 2014.

16:00 The Male with Infertility or Reproductive Failure
 Speaker: Craig Niederberger (USA)

The evaluation of and therapy for male reproductive dysfunction has accelerated markedly in the last two decades. An understanding of underlying pathophysiological mechanism including endocrinopathy has opened new horizons for tailored interventions for individual patients. Advancing technologies for interrogating sperm have provided details of sperm structure and function that were previously unknown. Elegant and effective surgical techniques are now available for conditions that were previously considered untreatable. However, with an abundance of innovations, it can be confusing which assessment should be applied for whom; how results should be interpreted to care for an individual male; and what therapy should be employed in specific cases. This talk aims to review innovations in the assessment of and therapy for male reproductive dysfunction and to identify targeted strategies for precision care.

16:30 Management of Hypothalamic Amenorrhea
 Speaker: Richard Reindollar (USA)

Historically, hypothalamic causes of reproductive failure have been considered separately for: (1) delayed puberty / primary amenorrhea, and (2) secondary amenorrhea. Studies have occurred independently and parallel; the former group being considered as having irreversible hypogonadotropic hypogonadism (HH) and the later labeled with reversible HH or hypothalamic amenorrhea. Molecular studies of patients with delayed puberty (and low gonadotropins) have revealed that a number of genes are involved in reproductive function, and, mutations in them can cause this seeming irreversible disorder. Interestingly, however, some of these patients with gene mutations and delayed puberty have been found to have reversible disease later in life. Also, others (both men and women) have been found to initiate puberty and normal reproductive function (cyclic menses for women) and develop HH later in life; the disorder for them being an associated similar gene mutation. Implications of recent findings in these disorders for management will be discussed.

Free Communication 7 - Nursing Counselling, Ethics, Psychology

15:30 - 17:00

Meeting Room P1

Chairs: Louise Younger (Australia)
Cailin Jordan (Australia)

- 15:30 **FC037**
OBESITY HAS LIMITED PSYCHOLOGICAL IMPACT IN PATIENTS UNDERGOING FERTILITY TREATMENT
I. Rodino, S. Byrne, K. Sanders (Australia)
- 15:45 **FC038**
THE NIPT EXPERIENCE: REVIEW OF OVER 250 CASES FROM A SINGLE PROVIDER
B. Peach, S. McDowell, B. Sutton, S. Sinnott, A. Yazdani (Australia)
- 16:00 **FC039**
THE PERCEPTIONS AND MOTIVATIONS OF POTENTIAL ANONYMOUS SPERM DONORS IN QUEENSLAND
A. Arnold, K. Harrison, L. Wilson, C. Carrigan, S. Fruk (Australia)
- 16:15 **FC040**
WHAT DO PATIENTS UNDERSTAND ABOUT THEIR PARTICIPATION IN THE IVF ACUPUNCTURE MULTICENTRE RANDOMISED CONTROLLED TRIAL?
C. Smith, S. Fogarty (Australia)
- 16:30 **FC041**
SEXUAL FUNCTION AND HEALTH-RELATED QUALITY OF LIFE IN WOMEN WITH CLASSICAL BLADDER EXSTROPHY
R. Deans, L. Liao, D.A.N. Wood, C. Woodhouse, S. Creighton (Australia)
- 16:45 **FC042**
MATERNAL ANXIETY, ART OUTCOME AND EARLY CHILDHOOD TEMPERAMENT: A THREE-YEAR FOLLOW-UP STUDY FROM CHINA
L.T. Chen, F. Qu, F.F. Wang, F.H. Zhang, W.W. Su, L.F. Xing, H.F. Huang (China)

Free Communication 8 - Genetics

15:30 - 17:00

Meeting Room P2

Chairs: Michelle Fraser (Australia)
Don Leigh (Australia)

- 15:30 **FC043**
ANEUPLOIDY PATTERNS IN BLASTOCYSTS.
K. Sorby, E. Osborne, T. Osianlis (Australia)
- 15:45 **FC044**
PREDICTING SEGREGATION MODES FOR AUTOSOMAL RECIPROCAL TRANSLOCATIONS AT PREIMPLANTATION GENETIC DIAGNOSIS
C.E. Lillee, E. Osborne, J. Ryan, T. Osianlis (Australia)
- 16:00 **FC045**
DOES FEMALE AGE, AMH, CYCLE TYPE, AND FSH DOSING IMPACT ON THE NUMBER OF DAY 5/6 BLASTOCYSTS SUITABLE FOR BIOPSY AND SUBSEQUENT ANEUPLOIDY RATES?
A. Marren, M.A.R.K. Bowman, M. Bowman, K. Emmett, B. De Vries, M. Traversa (Australia)
- 16:15 **FC046**
NEXT GENERATION SEQUENCING-BASED ANEUPLOIDY DETECTION
T. Hardy, T. Fullston, M. Lane, W. Ledger (Australia)

- 16:30 **FC047**
A HIGH-THROUGHPUT AND ROBUST BAC-ON BEADS BASED COMPREHENSIVE CHROMOSOME SCREENING (KARYOLITE) TO IMPROVE THE SUCCESS OF PREIMPLANTATION GENETIC SCREENING
K. Choy, C. Wong, Q. Yeung, W. Chong, J.P. Chung, C.J. Haines, G. Kong (Hong Kong, China)
- 16:45 **FC048**
SELECTING A FERTILITY SPECIALIST: WHOSE OPINION MATTERS?
K. Lambert, S. Fruk, C. Carrigan, S. McDowell, A. Yazdani (Australia)

Free Communication 9 - ART Basic, Embryology and Laboratory

15:30 - 17:00

Meeting Room M2

Chairs: Osamu Okitsu (Japan)
 Jacquelyn Irving (Australia)

- 15:30 **FC049**
CHANGES IN THE SURFACE AREA OF HUMAN OOCYTES FOLLOWING ICSI: OBSERVATIONS MADE USING THE EMBRYOSCOPE™ TIME-LAPSE SYSTEM.
Y. Liu, V. Chapple, P. Roberts, J. Ali, P. Matson (Australia)
- 15:45 **FC050**
MEIOTIC SPINDLE NORMALITY PREDICTS LIVE BIRTH IN PATIENTS WITH RECURRENT IVF FAILURE
S. Kilani, M.G. Chapman (Australia)
- 16:00 **FC051**
EXPANDED BLASTOCYST MORPHOLOGY ASSESSMENT AND THE DECISION TO TRANSFER: RESULTS FROM AN EXTERNAL QUALITY ASSURANCE PROGRAMME.
E. Zuvela, P. Matson (Australia)
- 16:15 **FC052**
EFFECT OF GONADOTROPINS ON EPIGENETICS AND GENOMIC IMPRINTING IN OOCYTES: ART SAFETY IN VITRO MODEL
H. Feng, T. Tsai, J. Qiao (USA)
- 16:30 **FC053**
THE EFFECTIVE OF PLATELET LAYSET (PL) ON PROGESTERONE (P4) SECRETION FROM MOUSE PRE-ANTRAL FOLLICLES AND CONSEQUENCE OVULATION
H. Pazoki, H. Eimani, F. Farokhi, A.H. Shahverdi, B. Ebrahimi, R. Salman Yazdi, L.S. Tahaei (Iran)
- 16:45 **FC054**
CORRELATION OF OXIDATIVE STRESS BIOMARKERS IN HUMAN FOLLICULAR FLUID WITH OUTCOME IN ASSISTED REPRODUCTION CYCLES
T. Nishihara, N. Shimizu, S. Hashimoto, T. Amano, Y. Nakaoka, Y. Hosoi, K. Matsumoto, Y. Morimoto (Japan)

"How to ... Session 5" - How to do IVM Successfully

15:30 - 16:15

Meeting Room M1

Chair: Roger Hart (Australia)

15:30 How to do IVM Successfully

Speaker: Manh Tuong Ho (Vietnam)

In vitro maturation (IVM) involves collecting oocytes from small follicles in unstimulated ovaries. The collected oocyte then can be matured in-vitro. The major drawbacks of current IVF protocol result from controlled ovarian stimulation (COS), including drug costs, burden on patients, and the risk of ovarian hyperstimulation syndrome. IVM has been applied as an alternative to eliminate the drawbacks of COS. However, IVM has remained empirical and relatively inefficient. Recently, improvements in IVM protocols have led to better treatment outcomes. We have applied IVM since 2006 for different indications, including PCO/PCOS patients, very high risk of OHSS during COS, steady response to COS... Almost 2,000 IVM cycles have been done and more than 500 children have been born. Our recent results demonstrate that IVM is an efficient and safe treatment alternative, especially for PCO patients. Current treatment protocols and results of IVM are presented.

16:00 Discussion

"How to ... Session 6" - How to Assess Ovarian Reserve

16:15 - 17:00

Meeting Room M1

Chair: William Ledger (Australia)

16:15 How to Assess Ovarian Reserve

Speaker: Andon Hestiantoro (Indonesia)

The main function of ovaries is producing mature and viable oocytes, with regards to fertilization process, embryo formation and embryo implantation. Assessment of ovarian reserve is one of critical point in counting of reproductive life of women, as well as the likely success of assisted reproductive techniques (ART). Many ways and marker have been developed to determine a woman's ovarian reserve in daily practice, such as, assessing ovarian volume, basal antral follicle count, gonadotropin hormone and AMH levels measurement, and others. So far, none of those ovarian reserve tests directly measure total number of actual oocytes; rather, assuming the number of recruit able oocytes, which is associated with the rest of oocyte pool. In addition, besides predicting the remaining number of oocytes in advanced age women, the real function of oocytes would be something that is important to find the answer.

16:45 Discussion

Plenary Lecture 2 - Ian Johnston Memorial Lecture

17:00 - 17:30

Great Hall Door 6 & 7

Chairs: Mark Bowman (Australia)

Lyndon Hale (Australia)

17:00 **The Future of Clinical Trials**

Speaker: Richard Legro (USA)

Clinical trials in infertility are extraordinarily complex as they involve a mother, father and hopefully an eventual fetus and infant, all of whom must be accommodated in the study design. Further multiple treatment cycles lend to higher dropout rates and difficulty in data analysis. Infertility trials often fail to report key outcomes of interest to patients and the public health. These included adverse events including multiple pregnancy, live births, and congenital and neonatal abnormalities. However the quality of infertility trials is improving based on tracking of quality of trials. Further we have seen a number of high quality randomized trials in infertility that have changed practice and given important insight into infertility and reproductive endocrine disorders.

Poster Sessions / Mini Oral Presentations 2

17:30 - 18:30

Great Hall Door 1

Chairs: Nao Suzuki (Japan)

Clare Boothroyd (Australia)

17:45 **M007**

COMPARISON OF PREGNANCY RATES BETWEEN IVF CYCLES WITH AND WITHOUT MATURE OOCYTES (MII) AT COLLECTION

H.A. Le, T.X. Pham, A.T. Lam, M.T. Ho (Vietnam)

17:50 **M008**

RETROSPECTIVE ANALYSIS OF SERUM ANTI-MULLERIAN HORMONE LEVEL IN A COHORT OF PATIENTS WITH RECURRENT MISCARRIAGE

K.Y. Kong, N. Varnier, G. Sacks (Australia)

17:55 **M009**

OXIDANT AND ANTIOXIDANT STATUS IN NON- OBESE WOMEN WITH AND WITHOUT POLYCYSTIC OVARY SYNDROME

S. Aali (Australia)

18:00 **M010**

REGULATION OF SERUM AMH IN PATIENTS WITH PCOS: RELEVANCE OF INSULIN RESISTANCE INDICES

H. Asakura, K. Nishio (Japan)

18:05 **M011**

METABOLIC RISKS OF THE LEAN PCOS WOMAN.

S. Kar (India)

18:10 **M012**

IMPAIRED FASTING GLUCOSE (IFG) AND GLUCOSE TOLERANCE TEST (IGT) OCCUR ACROSS THE SPECTRUM OF BMI AND PHENOTYPES OF PCOS PATIENTS

M. Maidarti, B. Wiweko (Indonesia)

Panel Session: Individualized Management in Patients Undergoing ART (By Invitation)

17:30 - 18:30

Meeting Room M5

Chair: YM Choi (Korea)

IVM in PCOS

Panelist: YS Kim (Korea)

IVM in PCOS

Panelist: MJ Kim (Korea)

COS in PCOS

Panelist: NY Kim (Korea)

COS in PCOS

Panelist: AM Park (Korea)

COS in Endometriosis

Panelist: JS Koo (Korea)

COS in Endometriosis

Panelist: YB Choi (Korea)

COS in Poor Responder

Panelist: CY Hur (Korea)

COS in Poor Responder

Panelist: HY Kim (Korea)

Repeated Implantation Failure

Panelist: KS Lim (Korea)

Social Function: Faculty Reception (By Invitation)

19:00 – 21:30

Queensland Art Gallery, Gallery of Modern ART

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Saturday, April 05, 2014

Breakfast Sponsored Symposium - Local Experience & Patient Acceptance of Elonva in Clinical Practice (MSD)

07:00 - 08:15

Meeting Room M1

Chair: Mary Birdsall (New Zealand)

Speaker: Devora Lieberman (Australia)

Meeting: FSA Preconception Health SIG Meeting

07:00 - 08:00

Meeting Room M2

Concurrent Session 4 - The Future Of Fertility Interventions

08:30 - 09:00

Great Hall Door 6

Chairs: Anusch Yazdani (Australia)

Craig Niederberger (USA)

08:30 Are We Doing Too Much IVF?

Speaker: Ben Willem Mol (Netherlands)

One million babies were born in the first 25 years of IVF between 1978 and 2003. It took only two more years for the tally to reach two million in 2005, with over five million estimated to have been born by the end of 2013.³ In developed countries with public health systems 2-3% of the births each year are through IVF, rising as high as 5% in Denmark and Belgium.⁴

Since the birth of the first baby by in vitro fertilisation (IVF) in 1978, the technique has earned its reputation as a major medical breakthrough of the 20th century. IVF was developed for women with tubal disease, but its indications soon began to grow. In the 1990s intracytoplasmic sperm injection was developed to treat couples in which the man has poor semen quality, which like tubal infertility prevents sperm from coming into close proximity with an egg. In recent years, however, IVF has been applied to other types of subfertility such as mild male subfertility, endometriosis, and unexplained subfertility. The birth of many healthy children has enhanced provider and patient confidence in the safety of IVF. But does applying IVF to wider forms of infertility result in overtreatment of couples who had a reasonable chance of conceiving naturally? Is it equally effective in these conditions? And, as more is understood about the adverse health outcomes in IVF children can the risks of IVF be justified for these more liberal applications?

Concurrent Session 5 - The Future Of Uterine Transplantation

09:00 - 09:45

Great Hall Door 6

Chairs: Anusch Yazdani (Australia)

Craig Niederberger (USA)

09:00 Speaker: Mats Brännström (Sweden)

One of last frontiers to conquer in infertility treatment is absolute uterine factor infertility (AUI), which affects around one in every 500 women. Our team initiated animal-based research on UTx in 1999, and have then by a systematic step-by-step procedure taken the procedure from rodents, to large animals (sheep, pig) and non-human primates. We are presently performing the world's first clinical trial on uterine transplantation, including 9 women (mostly MRKH) receiving uterine transplants from live donors, with a majority being mothers. The surgical techniques and the 6-month outcome of the entire cohort and up to 12 months for some patients will be presented. The important end result and definition of a successful transplantation is birth of a live baby. Embryo transfers have started in our study population.

Concurrent Session 6 - The Future Of Psychosocial Interventions In Infertility

09:45 - 10:30

Great Hall Door 6

Chairs: Anusch Yazdani (Australia)

Craig Niederberger (USA)

09:45 Speaker: Petra Thorn (Germany)

Psychosocial infertility counselling has evolved and changed dramatically over the last 20 years. Initially, it focussed mainly on the female receiving medical treatment, in the last years, infertility counselling aimed to support the couple as an entity. Counselling in the area of third party reproduction has also included the perspective of the child to be conceived. Currently, counsellors are not only supporting these parties, but also donors and surrogates before, during and after treatment. In the future, counsellors are likely to have to embrace an additional task: supporting linking and contact between offspring, their families and donors and surrogates. This task is likely to require additional skills and interventions, which are currently being developed and debated.

Concurrent Session 7 - Development of Larger IVF Organisations. Good or Bad?

08:30 - 10:30

Great Hall Door 8

Chairs: Andrew Murray (New Zealand)
 Vladislav Korsak (Russia)

08:30 An Indian Perspective

Speaker: Manish Banker (India)

The term corporatization means Private Companies listed or backed by private equity or venture funds providing IVF services as opposed to individual doctors. In the IVF field, most of the medical advances have been undertaken at educational and corporate entities with institutions like University of Brussels or Corporates like IVI being at the forefront of developing science; however the services have generally been provided by individual doctors. Corporate entities have access to capital and resources; this generally leads to creation of better equipments and facilities alongwith effective marketing to create awareness. They are able to get economies of scale and hence lower the cost of advanced treatments. The infertile couple is best served by healthy and ethical Corporates that enable doctors to do what they do best i.e. impart the best medical services in the most advanced facilities, using the best equipment and following internationally accepted protocols.

09:00 A Malaysian Perspective

Speaker: Haris Hamzah (Malaysia)

Malaysia has a 3.5 million population of potential mothers (Age 20-44). Assuming a 15% subfertility rate (525,000 women) and if 5% in this group require ART, this would result in 26,250 potential ART treatment cycle per year. The current number of ART cycles is 4500 (17%) annually.

The pioneer ART unit was established in 1986 and currently there are 35 IVF units,

The potential number of treatment cycles is attractive for its financial potential both to local as well as foreign investment.

While presently unregulated, the Malaysian Ministry of Health has established regulations for ART which are expected to be made into legislation in the next 2 years.

Corporatisation by local and international investors may avail some advantages to ART practice.

09:30 An Australian Perspective from a Large Organisation

Speaker: Peter Illingworth (Australia)

No Summary Provided

10:00 An Australian Perspective from a Small Organisation

Speaker: Clare Boothroyd (Australia)

No Summary Provided

Free Communication 10 - Ovary and ART

08:30 - 10:30

Meeting Room M2

Chairs: Hyuck Dong Han (Korea)
Debra Gook (Australia)

- 08:30 **FC055**
ADMINISTRATION OF SAI-REI-TOU AND CABERGOLINE IS EFFECTIVE TO PREVENT SEVERE OHSS
K. Ito, H. Iwahata, R. Nakahira, T. Himeno, T. Inoue, S. Hashimoto, Y. Nakaoka, Y. Morimoto (Japan)
- 08:45 **FC056**
ENDOMETRIAL RECEPTIVITY OF ENDOMETRIOSIS PATIENTS MOLECULAR BIOLOGY STUDY ON POLYMORPHISM OF MUC-1 AND COX-2
U. Budihastuti, D. Dasuki, A. Sadewa (Indonesia)
- 09:00 **FC057**
CORRELATION BETWEEN LEVELS OF HIGH-DENSITY LIPOPROTEIN LEVEL IN SERUM AND FOLLICULAR FLUID WITH OOCYTE QUALITY IN PCOS PATIENTS UNDERGOING IVF
S. Soebijanto, M. Natadisastira, R. Isnaei (Indonesia)
- 09:15 **FC058**
INFLUENCE OF THE STATUS OF FALLOPIAN TUBE ON OVARIAN RESERVE AND PREGNANCY RATE OF IVF-ET
H. Matsumoto, S. Mizuno, M. Ida, A. Fukuda, Y. Morimoto (Japan)
- 09:30 **FC059**
AN INDIRECT MECHANISM TO EXPLAIN THE BENEFICIAL EFFECT OF GROWTH HORMONE ON HUMAN OOCYTE QUALITY IN POOR RESPONDER PATIENTS.
G. Almahbobi, S. Regan, J. Stanger, J. Yovich (Australia)
- 09:45 **FC060**
A DIRECT MECHANISM FOR THE PUTATIVE ROLE OF GROWTH HORMONE IMPROVING OOCYTE QUALITY FROM POOR RESPONDER PATIENTS.
G. Almahbobi, B. Weall, J. Yovich (Australia)
- 10:00 **FC061**
EMBRYO MORPHOKINETIC DEVELOPMENT IS THE SAME FOR PATIENTS WITH AND WITHOUT POLYCYSTIC OVARIES AND IS NOT IMPAIRED BY IVM TREATMENT.
M. Walls, J. Ryan, R. Hart (Australia)
- 10:15 **FC062**
THE ROLE OF SERUM ANTI-MULLERIAN HORMONE (AMH) IN PREDICTING POLYCYSTIC OVARIAN SYNDROME (PCOS)
H. Giang, T. Le, T. Vo, T. Vuong, M. Ho (Vietnam)

"How to ... Session 7" - How to Determine the Best Culture Medium

09:00 - 09:45

Meeting Room M1

Chair: Marcos Meseguer (Spain)

09:00 Speaker: Cecilia Sjoblom (Australia)

The IVF laboratory and embryology play a key role in the success of IVF, contributing over 70% to the overall results. Embryology is developing fast with new formulations of embryo culture medium continuously introduced.

The talk will cover not only the components of the culture medium but also other aspects having an impact on the 'best' culture medium such as sequential / single step, quality control, manufacturing, packaging, cold chain and customer service.

Learning objectives;

To give an overview of the development, components and functions of the modern embryo culture medium.

To critically review the approaches of different medium producers

To apply evidence-based knowledge to determine the best embryo culture medium.

09:30 **Discussion**

"How to ... Session 8" - How to Evaluate Success with PGS

09:45 - 10:30

Meeting Room M1

Chair: Colin Lee (Malaysia)

09:45 Speaker: Don Leigh (Australia)

Success can be evaluated differently by the various participants in the IVF process. For the patient it may simply be having a healthy baby, preferably on the first attempt. The definition may extend further to having several successful pregnancies and births, a family, in a timely manner. Failure can have several measures- no pregnancy, miscarriage or a child with a syndrome. For the IVF doctor the meaning of success will overlap with the above patient's measures but could also include successful clinical management of many different types of patients with a diversity of fertility issues, management of patient expectations, recurrent miscarriage, recurrent implantation failure and avoidance of genetic disease. The laboratory will have their own goals and measures of success, typically embryo implantation rates, reduced miscarriage rates and optimal use of laboratory and clinic resources. Is this achievable in today's IVF?

10:15 **Discussion**

Coffee Break and Exhibition Viewing

10:30 - 11:00

Great Hall Door 1

Concurrent Session 8 - The Future Of Contraception

11:00 - 12:30

Great Hall Door 6

Chairs: Devora Lieberman (Australia)
 Gab Kovacs (Australia)

11:00 Reducing unintended pregnancies in Australia- Leveraging UK experience with Long Acting Reversible Contraceptives

Speaker: Paula Briggs (United Kingdom)

Over the last 12 years, the UK has seen a 24% reduction in unplanned pregnancy in the under 18s. This mirrors an increase in Long Acting Reversible Contraception (LARC) provision. Dr Briggs lecture will describe the UK experience of LARC to reduce the risk of unplanned pregnancy.

11:30 Physiology of Reproduction/ Biochemistry of Steroids in the Pill

Speaker: Gab Kovacs (Australia)

This presentation will review the development of hormonal contraception commencing with the pioneering work of Marker, in Mexico City, who co-formed the company Syntex and combined with Carl Djerassi to manufacture the first orally acting progestin, norethisterone in 1951, to the recent development and incorporation of natural progestins in oral contraceptives. The lecture will include a review the biochemistry of steroid hormones as is relevant to clinicians. Finally it will provide some practical guidelines on the prescription of oral contraceptives, including an update on risks and complications.

12:00 Vaginal Contraception- Back to the Future

Speaker: Terri Foran (Australia)

Vaginal contraception has a time-honoured feminine tradition which was largely superseded with the advent of the oral contraceptive Pill in the 1960s. The Pill offered a level of effectiveness and convenience previously undreamt of and vaginal contraception came to be regarded as somewhat 'messy' and unfashionable. However the quest for alternative delivery systems for hormonal contraception saw the development of the hormonal contraceptive ring and there is no doubt that it offers some unique advantages in terms of convenience, side-effects and cycle control for the intending user. This presentation seeks to explore the pros and cons of this method as well as to address some of the misconceptions and controversies surrounding it. The presentation will also examine the place of an old favourite- the occlusive cap- in a world where women in both developed and developing countries are demanding a wider range of options to meet their varying contraceptive needs.

Industry Sponsored Symposium 4 - Patient Centred ART: Using Biomarkers To Optimise Ovarian Stimulation (Ferring)

11:00 - 12:30

Great Hall Door 8

Chairs: Robert Norman (Australia)
Pak Chung Ho (Hong Kong, China)

- 11:00 **Welcome and Introduction**
Speaker: Robert Norman (Australia)
- 11:10 **The Start of the Beginning**
Speaker: Richard Fleming (United Kingdom)
- 11:40 **Biomarkers, Evolution of use in Ovarian Stimulation - The Asia-Pacific Experience**
Speaker: Vuong Thi Ngoc Lan (Vietnam)
- 12:05 **Biomarkers, My Use and Your Use to Individualise Ovarian Stimulation**
Speaker: Richard Paulsen (USA)
- 12:30 **Closing**
Speaker: Pak Chung Ho (Hong Kong, China)

Free Communication 11 - Male Fertility

11:00 - 12:30

Meeting Room P1

Chairs: Aisaku Fukuda (Japan)
Virgilio Novero (Philippines)

- 11:00 **FC063**
HAVE SPERM CHARACTERISTICS CHANGED OVER TIME IN A POPULATION OF QUEENSLAND MEN? AN ANALYSIS OF 1799 SAMPLES FROM 2003 TO 2012.
H. Wills, J. Osborn, S. McDowell, B. Kroon, E. Ford, K. Harrison, J. Wolyncevic, J. Esler, A. Yazdani (Australia)
- 11:15 **FC064**
DYNAMIC EXPRESSION OF SEPT12 AFFECTS THE INTEGRATION OF NUCLEAR ENVELOPE DURING HUMAN SPERMIOGENESIS
Y.H. Lin, Y.Y. Wang, P.L. Kuo (Taiwan)
- 11:30 **FC065**
COMPARISON OF CYCLE NUMBER TO SUCCESSFUL IVF OUTCOME AFTER ICSI FROM EJACULATE SPERMATOCYTES, PERCUTANEOUS EPIDIDYMAL SPERM ASPIRATION AND TESTICULAR SPERM EXTRACTION IN SEVERE OLIGOZOOSPERMIA PATIENTS
H. Morita, K. Uchiyama, T. Okimura, J. Fukuda, K. Kato (Japan)
- 11:45 **FC066**
MESA: DOES IT CONTINUE TO IMPACT ICSI RESULTS?
H. Hibi, T. Ohori, M. Sumitomo, N. Fukunaga, Y. Asada (Japan)
- 12:00 **FC067**
A GLOBAL PROTEOMIC APPROACH FOR ELUCIDATING THE MECHANISM OF THE ACTION OF AIRE IN GERM CELLS
K. Radhakrishnan, K.P. Bhagya, P.G. Kumar (India)
- 12:15 **FC068**
IS MICRO-DISSECTION TESTICULAR SPERM EXTRACTION THE MINIMUM STANDARD OF CARE FOR NON-OBSTRUCTIVE AZOOSPERMIC MEN?
A. Talmor, K. Nowak, V. McLachlan, L. Rombauts, C. Motteram, R. McLachlan (Australia)

Free Communication 12 - Endometriosis

11:00 - 12:30

Meeting Room P2

Chairs: Asma Munir (United Arab Emirates)
Colin Howles (Switzerland)

- 11:00 **FC069**
PAIN PATHOGENESIS IN ENDOMETRIOSIS
K.M. Peters, I.S. Fraser, P.J. Wrigley (Australia)
- 11:15 **FC070**
IMPLEMENTATION OF ROBOTIC IN REPRODUCTIVE SURGERY IN INDONESIA
I. Sini, I. Suheimi (Indonesia)
- 11:30 **FC071**
ADIPONECTIN SERUM LEVELS IN ENDOMETRIOSIS
R. Anwar, F. Fahdiansyah, F.F. Wirakusumah, D.S. Nataprawira (Indonesia)
- 11:45 **FC072**
THE EFFECT OF INCREASING DOSE OF CURCUMIN SUPPLEMENTATION ON EXPERIMENTAL ENDOMETRIOSIS PROGRESSIVITY IN MICE
J. Annas, H. Hendarto, Widjiati (Indonesia)
- 12:00 **FC073**
THE EFFECTIVENESS OF DLBS 1442 IN ALLEVIATING ENDOMETRIOSIS- AND/OR ADENOMYOSIS-RELATED PAIN
B. Wiweko, C.G. Puspita, H. Situmorang, A.K. Harzif, G. Pratama, K. Sumapraja, M. Natadisastra, R. Tjandrawinata, A. Hestiantoro (Indonesia)
- 12:15 **FC074**
FACTORS ASSOCIATED WITH SUCCESSFUL PREGNANCY AFTER CONSERVATIVE SURGERY FOR ADENOMYOSIS.
Y. Kishi, M. Yabuta (Japan)

"How to ... Session 9" - How to Set Up a TQM Programme

11:00 - 11:45

Meeting Room M1

Chair: Su Ling Yu (Singapore)

11:00 Speaker: John Peek (New Zealand)

Total Quality Management (TQM) is an organisation-wide approach to quality in order to provide safe and effective services, an organisation that prospers, and compliance to regulatory requirements. This presentation will lead you through the 6 steps to set up TQM for your clinic. 1) Define your goals, such as pregnancy rates, acceptable levels of complications, and levels of patient satisfaction. 2) Measure actual performance against your target using Key Performance Indicators (KPI) and share these with staff. 3) Capture any problem when it occurs with an incidence reporting system, and address the root cause of the problem. 4) Anticipate problems by setting up a risk register of things that could go wrong. 5) Introduce strategies to eliminate or reduce each risk. 5) Undertake internal audit to check that your risk reduction strategies are in place and working.

11:30 **Discussion**

"How to ... Session 10" - How to Set Up a Fertility Register in Emerging Markets

11:45 - 12:30

Meeting Room M1

Chair: Elizabeth Sullivan (Australia)

11:45 Speaker: Narendra Malhotra (India)

The market of ART in the world is growing and is currently estimated at 5 billion dollars. Clinics in Asia and developing countries are now offering all quality ART services at very economical rates and with good success.

These countries in the developing world, today lack proper LAWS and proper Guidelines for the regulation of ART. This has led to many mushrooming of clinics and some unethical commercialisation of the treatment. To combat this we need to set up FERTILITY REGISTER specially in the emerging markets of ART. We feel all the developing and emerging ART countries like China, Indonesia, Nepal and others should make effort to set up basic data collection registry. It's absolutely important and should be mandatory to set up ART registries, specially in emerging markets and developing countries, which still do not have any Laws.

12:15 **Q&A**

Lunch and Poster Sessions / Mini Oral Presentations 3

12:30 - 13:30

Great Hall Door 1

Chairs: Narendra Malhotra (India)

Devora Lieberman (Australia)

12:45 **M013**

THE EFFICACY OF EMBRYO CULTURE IN MEDIUM SUPPLEMENTED WITH GRANULOCYTE MACROPHAGE-COLONY STIMULATING FACTOR (GM-CSF) ON OUTCOMES IN REPEATED IMPLANTATION FAILURE (RIF) PATIENTS

H.G. Bao, N.T.T. Lan, D.Q. Vinh, H.M. Tuong (Vietnam)

12:50 **M014**

ENDOMETRIOSIS: HOW MUCH DOES IT COST AUSTRALIA?

K.M. Peters, I.S. Fraser (Australia)

12:55 **M015**

DEVELOPMENT OF INFANTS DERIVED FROM METAPHASE II OOCYTES WITH SMOOTH ENDOPLASMIC RETICULUM CLUSTERS (SERC)

K. Kyono, Y. Nakamura, H. Hattori, Y. Nakajo, Y. Araki, M. Iwata, T. Takeuchi (Japan)

13:00 **M016**

INVESTIGATING THE POTENTIAL USE OF CHLAMYDIAL SEROLOGY DURING THE INITIAL INFERTILITY INVESTIGATION

S.H. Stansfield, S. Menon, J.A. Allan, G. Weston, L. Rombauts, W. Huston (Australia)

12:05 **M017**

BODY MASS INDEX (BMI) AS A PREDICTIVE FACTOR FOR MENARCHE IN INDONESIAN FEMALE STUDENTS

A.M.P. Iskandar, G.N. Hidayah, D.P. Wardhani, C.G. Puspita, D. Yusuf, P.A. Iffanolida, K. Sumapraja, M. Natadisastra, A. Hestiantoro, B. Wiweko (Indonesia)

Meeting: FSA REACT Group SIG Meeting

12:30 - 13:30

Meeting Room M2

Meeting: LIGHT Study Update Meeting

12:30 - 13:00

Meeting Room M1

Concurrent Session 9 - The Future Of Fertility Preservation

13:30 - 15:00

Great Hall Door 6

Chairs: Mats Brännström (Sweden)

Nao Suzuki (Japan)

13:30 Current Topics on Fertility Preservation of Cancer Patient - Ovarian Tissue Vitrification

Speaker: Nao Suzuki (Japan)

We have developed a new vitrification device for ovarian tissue, which achieves rapid cooling rates as the tissue sections are exposed directly to liquid nitrogen. Based on our positive results of vitrification from preclinical study using cynomolgus monkey, we started to use this technique for clinical application. Finally we have succeeded to have a live birth using ovarian tissue vitrification method first time in the world. In this lecture, I will talk about our recent data of ovarian tissue vitrification and transplantation from animal lab to clinical practice.

14:00 Ovarian Stimulation Protocols in the Cancer Patients

Speaker: Jung Ryeol Lee (Korea)

In female cancer patients, various strategies have been used for fertility preservation; ovarian stimulation followed by cryopreservation of embryo or oocytes, and ovarian tissue cryopreservation and transplantation. Among these, both embryo and oocyte cryopreservation after controlled ovarian stimulation (COS) are considered as established fertility preservation methods. In conventional COS regimen, ovarian stimulation started during early follicular phase of menstrual cycle. This may require up to 5 weeks of delay in starting cancer treatments. During COS, estradiol level is elevated to non-physiologic level and this phenomenon can give rise to safety concern in hormone dependent cancer patients. To overcome time constraint and safety concern, random-start COS and letrozole protocol have been suggested. In this lecture, COS regimens for cancer patients, such as random start protocol for reducing delay of chemotherapy and letrozole protocol for prevention of estradiol elevation are introduced. And other special issues for COS in cancer patients are discussed.

14:30 Australian Experience

Speaker: Kate Stern (Australia)

Current options to preserve fertility in women who undergo potentially gonadotoxic treatment include: preservation of embryos, oocytes, or ovarian tissue prior to cancer treatment, and ovarian protection with the use of GnRH analogues throughout the duration of treatment. For men, fertility preserving options include: sperm cryopreservation, testicular hormonal suppression, and testicular tissue cryopreservation. Given the gonadotoxic effects of chemotherapy agents, the availability of options to preserve and protect fertility of cancer patients is of great importance to young men and women and their families, in particular, allowing patients a possibility of having a family of their own.

Making sure patients have the opportunity for discussion about future fertility, increasing the success (without the risk) of preserving strategies, maximising accessibility for uptake of the options, and providing long term fertility care, are the goals of our fertility preservation programs in Australia and elsewhere.

Concurrent Session 10 - The Future of Reproductive Sciences

13:30 - 15:00

Great Hall Door 8

Chairs: Jie Qiao (China)

Chii-Ruey Tzeng (Taiwan)

13:30 New Concepts on the Origin of PCOS

Speaker: Ray Rodgers (Australia)

As the ovary develops, stroma from the underlying mesonephros penetrates the primordium, partitioning it into irregularly-shaped ovigerous cords composed of GREL (Gonadal Ridge Epithelial-like) cells and primordial germ cells (PGC)/oogonia (PLOS ONE 8(2):e55578). GREL cells are precursors to both granulosa cells and surface epithelial cells (PLOS ONE 8(2):e55578). The penetrating stroma expresses fibrillin 3 (FASEB J 25, 2256-2265), a candidate gene for polycystic ovary syndrome (PCOS). In general fibrillins regulate TGF β activity which in turn stimulates replication of fibroblasts and collagen deposition. The adult PCOS ovary has increased stroma and collagen. Thus these findings (FASEB J 25, 2256-2265) suggest a mechanism by which the genetics of PCOS, its fetal origins and its ovary phenotype could all be related and how perturbation of this system could bring about a predisposition to developing PCOS in later life.

14:00 Exosomes: A New Paradigm for Embryo- Maternal Interactions in Establishing Pregnancy

Speaker: Lois Salamonsen (Australia)

Exosomes/microvesicles (MV) are 50-150nm diameter vesicles of endocytic origin that are released by most cells upon fusion of multivesicular bodies with the plasma membrane. They are presumed to act as a vehicle for intercellular communication and carry cargo of proteins, lipids, mRNA and miRNA which they can transfer to other cells. We have identified exosomes in uterine fluid of women, and characterised exosomes released from the endometrial cell line ECC1 in terms of their miRNA content (1). A number of the miRNA identified in exosomes, are undetectable in the cells of origin. We propose that such exosomes, present in the microenvironment of implantation, present a new concept in the endometrial-blastocyst interactions essential for establishment of pregnancy.

14:30 A Mechanism Explaining Why Peri-Conception Maternal Hyperglycaemia Affects Oocytes and Embryos

Speaker: Jeremy Thompson (Australia)

Hyperglycemia is associated with poor fertility and increased incidence of fetal congenital abnormalities. The peri-conception period is known to be extremely sensitive to hyperglycemia; abnormalities include reduced cellular volume in oocytes, disrupted metabolic measures (e.g. mitochondrial function and glucose transport) in embryos; disrupted signalling pathways (such as PI3K/Akt) in embryos. We focus on the hexosamine biosynthesis pathway (HBP) that enables the post-translational protein modification, O- β -glycosylation. This single glycosylation cycling is regulated by substrate supply from glucose that parallels, and often opposes, the kinase system. We have demonstrated that the hyperglycemic condition both in vitro and in vivo markedly increases O- β -glycosylation of oocytes and cumulus cells and is accompanied by a marked decrease in post-fertilisation developmental capacity. We have determined that HSP90 glycosylation is a key target, suggesting disrupted chaperone function. Our work is now following new leads that O- β -glycosylation is a regulator of epigenetic mechanisms, including histone function.

Free Communication 13 - Nursing Counselling, Ethics, Psychology

13:30 - 15:00

Meeting Room P1

Chairs: Michael Condon (Australia)
 Sheryl De Lacey (Australia)

- 13:30 **FC075**
CROSS-BORDER REPRODUCTIVE CARE – ONE CLINIC'S NATIONAL AND INTERNATIONAL EXPERIENCE
C. Carrigan, L. Wilson, A. Arnold, J. Irving, A. Yazdani, K. Harrison (Australia)
- 13:45 **FC076**
OLDER, HEAVIER AND SINGLE: HOW THE FEMALE IVF PATIENT HAS CHANGED OVER THE PAST FIFTEEN YEARS.
L. Fien, S. Lax, J. Logan, J. Esler, J. Osborn (Australia)
- 14:00 **FC077**
THE EFFECT OF COMPULSORY IDENTIFICATION OF SPERM DONORS ON SPERM DONOR PRACTICE
K. Barber, S. Cooke, P. Illingworth (Australia)
- 14:15 **FC078**
YOU HAVE MILLIONS WE ONLY WANT ONE! A SMALL CLINICS EXPERIENCE IN ESTABLISHING A SPERM DONOR RECRUITMENT PROGRAM.
D. Donati, D. Price, A. Torres, R. Frouws, N. Beutel (Australia)
- 14:30 **FC079**
UNIQUE HRT REGIMEN PROVES MOST EFFICIENT FOR FET CYCLES
L. Clifton, M. Kleidon, J. Conceicao, J.L. Yovich (Australia)
- 14:45 **FC080**
ISLAMIC PERSPECTIVE ON CONTRACEPTION
N.M.N. Ismail, M.A. Masri (Malaysia)

Free Communication 14 - ART, Clinical

13:30 - 15:00

Meeting Room P2

Chairs: Kanthi Bansal (India)
 Pratap Kumar (India)

- 13:30 **FC081**
BEYOND THE GOOD PROGNOSIS PATIENT: EXTENDED CULTURE INCREASES LIVE BIRTH RATES FOR PATIENTS, REGARDLESS OF QUALITY AND QUANTITY OF EMBRYOS ON DAY 3
A. Bensch, L. Rombauts, T. Osianlis (Australia)
- 13:45 **FC082**
CONTINUOUS IMPROVEMENT IN NATIONAL ART STANDARDS BY THE RTAC ACCREDITATION SYSTEM IN AUSTRALIA AND NEW ZEALAND
K. Harrison, J. Peek, M. Bowman, M. Chapman (Australia)
- 14:00 **FC083**
RISK OF PLACENTA PREVIA IN ART BIRTHS LINKED TO ENDOMETRIAL THICKNESS
L.J. Rombauts, C. Motteram (Australia)
- 14:15 **FC084**
THE IMPACT OF MATERNAL BODY MASS INDEX ON IMPLANTATION RATES FROM EUPLOID FROZEN EMBRYO TRANSFERS
K. Gebhardt, D. Zander-Fox, K. Tremellen, M. Lane (Australia)

- 14:30 **FC085**
A PROSPECTIVE RANDOMISED CONTROLLED STUDY COMPARING THE COST EFFECTIVENESS OF IVF-ICSI TREATMENT: CLEAVAGE STAGE (DAY 3) EMBRYO TRANSFER VERSUS EXTENDED CULTURE(DAY 5/6 BLASTOCYST) TRANSFER.

R. Singh, M. Singh, A. Jindal, P.C. Jindal (India)

- 14:45 **FC086**
INFLUENCE OF RE-VITRIFIED HUMAN BLASTOCYST ON LABORATORY DATA, CLINICAL OUTCOMES AND RESULTING BABIES

A. Koike, S. Mizuno, H. Matsumoto, A. Fukuda, Y. Morimoto (Japan)

Free Communication 15 - Genetics

13:30 - 15:00

Meeting Room M2

Chairs: Julia Loginova (Russia)
 Richard Reindollar (USA)

- 13:30 **FC087**
THREE YEARS EXPERIENCE IN A REGIONAL CLINIC OF PREIMPLANTATION GENETIC DIAGNOSIS OF ANEUPLOIDY (PGD-A) USING EMBRYO TRANSPORT AND ARRAY-COMPARATIVE GENOMIC HYBRIDISATION (ACGH)

J. Wolyncevic, J. Irving, L. Whitman, J. Esler, J. Osborn, L. Wilton (Australia)

- 13:45 **FC088**
PARTIAL GENOME SEQUENCING - AN ALTERNATIVE TO MICROARRAYS FOR EMBRYO ANEUPLOID SCREENING

P. Barahona, D. Leigh, W. Ritchie, S.J. McArthur (Australia)

- 14:00 **FC089**
A CHANGE IN STRATEGY FOR PREIMPLANTATION GENETIC DIAGNOSIS (PGD) PATIENTS – COMBINING DETECTION OF SINGLE GENE DEFECTS WITH COMPREHENSIVE CHROMOSOME SCREENING IN ELECTIVE FREEZE ALL CYCLES

M. Traversa, D. Leigh (Australia)

- 14:15 **FC090**
IDENTIFICATION OF AN INCREASED INCIDENCE OF CFTR MUTATIONS IN MALE ART PATIENTS

P. Field, N. Martin (Australia)

- 14:30 **FC091**
GENOME-WIDE COPY NUMBER VARIATIONS SCAN IN CHINESE PATIENTS WITH PRIMARY OVARIAN INSUFFICIENCY

X. Zhen, J. Qiao (China)

- 14:45 **FC092**
CHROMOSOME SEGREGATION ANALYSIS AND ESTIMATION OF INTERCHROMOSOMAL EFFECT IN HUMAN EMBRYOS FROM CARRIERS OF CHROMOSOMAL REARRANGEMENT. MULTICENTRAL STUDY.

J.A. Loginova, O.G. Chiryaeva, O.E. Vasilyeva, Y.R. Pastukhova, E.L. Katsanda, S.A. Yakovenko, A.A. Gusareva, A.A. Feoktistov, N.L. Shamugia, V.S. Korsak (Russia)

"How to ... Session 11" - How to Lower Costs in an IVF Programme

13:30 - 14:15

Meeting Room M1

Chair: Prof Alan Trounson (USA)

13:30 Speaker: Molloy David (Australia)

IVF is an expensive process. Treatment requires a complex team approach. Improved success rates have engendered higher laboratory costs. Cost and complexity are increased by the sophistication of added services including egg and sperm donation, freezing and fertility preservation, quality assurance and audit. Ovarian hyperstimulation generates significant pharmaceutical costs. Limited international trends have led to some simplification. These include minimal ovarian stimulation and moving oocyte retrieval from an inpatient sedation setting to an outpatient, non-sedation setting. Simple repetitive models of IVF exist. The Queensland Fertility Group has introduced a new model of IVF treatment which improves access and affordability. Gentle ovarian stimulation aims for no more than four oocytes per cycle. Oocyte retrieval is an outpatient procedure. Strict protocolisation lowers cost. Fewer embryos are frozen. The pregnancy rates are slightly lower but represent a substantial pregnancy opportunity for patients who otherwise could not afford IVF in Australia.

14:00 **Discussion**

"How to ... Session 12" - How to do Double Stimulation and Egg Recovery for Poor Responders

14:15 - 15:00

Meeting Room M1

Chair: John McBain (Australia)

14:15 Speaker: Yanping Kuang (China)

Classical IVF procedure starts ovarian stimulation in the early follicular phase and retrieves oocytes once when follicles mature. Our previous data confirmed the luteal-phase ovarian stimulation is feasible to for producing competent oocytes in IVF, with optimal pregnancy outcomes in frozen-thawed embryo transfer cycles. Given that the difficulty of egg recovery for poor ovarian responders (POR), we performed a new protocol of performing double stimulations in a menstrual cycle in patients with POR. Mild ovarian stimulation was initiated in the follicular phase, after the first ovum pick-up, ovarian stimulation continued and second oocyte retrieval performed when dominant follicles matured. Double ovarian stimulations in a same menstrual cycle provided more opportunities to retrieve oocytes in POR. The stimulation can start at the luteal phase and get more oocytes in a short period of time, which will be a new hope for newly diagnosed cancer patients needing fertility preservation.

14:45 **Discussion**

Coffee Break and Exhibition Viewing

15:00 - 15:30

Great Hall Door 1

Special Symposium 2 - Access and iCSI Symposium - The Future of Patient Centred ART

15:30 - 17:00

Great Hall Door 6

Chairs: Linda Giudice (USA)
 Sandra Dill (Australia)

15:30 Patient-Centred ART - A Japanese Clinic Perspective
 Speaker: Yoshiharu Morimoto (Japan)

The mission for ART center is to recognize patients' hope and make their dreams come true. It is most important for staff in infertility centers to take notice patients' words carefully and comprehend what they want. Human body is controlled not only by organs and cells but by total functions such as nervous, endocrine and immune systems that maintain integrity and homeostasis. Therefore, it is not possible to get actual healing without a broad territorial approach, which combines modern scientific technologies including mental and nutritional field with physical exercise and the eastern medicine. We have developed the field of "Integrated Medicine in Reproduction" for a decade. The division of Integrated Medicine that was established in 2012 in our centers are effectively working for the patients with recurrent failure, poor quality embryos as well as aging. Integrated Medicine in Reproduction is a new trend in the field of reproductive medicine and would be a future vision.

15:45 Patient-Centred ART - A Japanese Consumer Perspective
 Speaker: Mikae Ueda (Japan)

The growing number of infertility patients has seen the number of fertility clinics in Japan increased to 586 in 2011. The information clinics provide is not regulated by government or other organisations. The Fertility Information Network conducted an online survey from October 2011 to July 2012, to explore how infertility patients seek information when they begin treatment or wish to change clinics and to explore their real feelings about their clinics while undergoing treatment. Five hundred sixty men and women aged 30 to over 46 responded. The presentation will examine the focus of patients when choosing a clinic and the reasons why they may wish to change clinics. The presentation will also introduce a clinic accreditation program, which includes patient representatives, established in 2005 by the Japan Institution for Standardizing Assisted Reproductive Technology, in pursuit of quality reproductive treatment and patient-centred care.

16:00 Patient-Centred ART - A Male Consumer Perspective
 Speaker: David Rawlings (Australia)

One valuable perspective in the ART process is sometimes hidden – the view of the male half of the couple participating in treatment. Men can be silent partners in reproductive treatment and this silence can often be perceived as something it is not. For many men, the acceptance of reproductive treatment brings with it a whole raft of logistical and management issues. Infertility poses emotional, mental, physical and sometimes spiritual threats for which they are not prepared. Some men can be marginalised in the ART process – by their partner or sometimes by the clinic assisting with their infertility. Understanding their infertility experience can improve treatment for both partners.

16:15 Patient Centred ART in Resource-Constrained Settings
 Speaker: Alan Trounson (USA)

The psychological, social, and economical consequences of involuntary childlessness are severe, particularly for women in resource-poor settings where childlessness is stigmatized. WHO considers infertility a global health problem and argue that assisted reproductive technologies (ART) should be an integral part of countries' sexual and reproductive health care agenda. In most low-income countries the cost of treatment is prohibitive for most people. Initiatives to make ART available to a broader range of people are emerging. Strategies to alleviate the suffering of infertility in low-income countries should include:

- Education to prevent infertility
- Advocacy to increase awareness of the problem of childlessness

- Training of local health professionals by reproductive medicine experts from high-income countries
- Use of simplified ART protocols that have been tested in high-income settings and shown to deliver acceptable success rates
- Establishment of private-public partnerships to establish high-quality infertility care in public hospitals.

16:30 **Panel Discussion**

Special Symposium 3 - ISAR Symposium - Endoscopy: Ultrasound at its best in Fertility Enhancement

15:30 - 17:00

Great Hall Door 8

Chairs: Manish Banker (India)
 Narendra Malhotra (India)

15:30 **Laparoscopy**

Speaker: Prakash Trivedi (India)

15:45 **Hysteroscopy**

Speaker: Sunita Tendulwadkar (India)

16:00 **“Which Patient? Which Surgery? For Maximising the Reproductive Outcome”**

Moderator: Jaideep Malhotra (India)

Panelist: Rekha Kurien (India)
 Panelist: Kaberi Bannerji (India)
 Panelist: Kuldeep Jain (India)
 Panelist: Sonal Panchal (India)
 Panelist: Sudha Prasad (India)
 Panelist: Kanthi Bansal (India)

Free Communication 16 - ART Basic, Embryology and Laboratory

15:30 - 17:00

Meeting Room P1

Chairs: Carolyn Hills (Australia)
 Keith Harrison (Australia)

15:30 **FC093**

THE PREVALENCE AND MORPHOKINETICS OF HUMAN EMBRYOS DEMONSTRATING REVERSE CLEAVAGE WHEN VIEWED USING THE EMBRYOSCOPE™ TIME LAPSE VIDEO SYSTEM.

Y. Liu, V. Chapple, P. Roberts, P. Matson (Australia)

15:45 **FC094**

STAGE SPECIFIC CYTOPLASMIC TEXTURE MEASUREMENTS ARE INDICATIVE OF THE PLOIDY STATUS OF PREIMPLANTATION HUMAN EMBRYOS

N. Symons, E. Zuvela, R. Drew, M. Walls, R. Hart, J.P. Ryan (Australia)

16:00 **FC095**

MEIOTIC SPINDLE LOCATION IN COMBINATION WITH SPINDLE NORMALITY MAY IMPROVE PREGNANCY PREDICTION.

S. Kilani, M.G. Chapman (Australia)

- 16:15 **FC096**
TECHNOLOGY FOR RESCUE FERTILITY OF AZOOSPERMIC PATIENTS BY TRANSPLANTATION OF SPERMATOGONIAL STEM CELLS
Y. Wu, F. Luo, S. Wu, Y. Zhang, H. Su, Q. Kong, S. Fu (China)
- 16:30 **FC097**
MORPHOKINETIC EVALUATION OF EMBRYOS: VARIABLES LINKED TO EMBRYO DEVELOPMENT AND IMPLANTATION
M. Montag, B.M. Petersen, M. Lægdsmand, T.Q. Kajhøj (Denmark)
- 16:45 **FC098**
DISCRIMINATION OF MATERNALLY OR PATERNALLY DERIVED PRONUCLEI BY EPIGENETIC DIVERGENCE IN HUMAN ZYGOTES FERTILIZED ABNORMALLY
Y. KAI, K. Iwata, K. Yumoto, M. Sugishima, C. Mizoguchi, S. Furuyama, Y. Matoba, Y. Iba, Y. Mio (Japan)

Free Communication 17 - ART, Clinical

15:30 - 17:00

Meeting Room P2

Chairs: Bhola Rijal (Nepal)
 Prashant Nadkarni (Malaysia)

- 15:30 **FC099**
TRANSCRIPTOME ANALYSIS REVEALS PLACENTA SUBJECTED TO ASSISTED REPRODUCTIVE TECHNOLOGY TREATMENTS MAY HAVE WIDESPREAD GENOMIC EFFECTS
Z. Liang, P. Jie.qiao, P. Rong.Li, P. Liying Yan, P. Yang YU (China)
- 15:45 **FC100**
TOWARDS AN OBJECTIVE DEFINITION OF POOR RESPONDERS IN ASSISTED REPRODUCTION
H.N. Sallam, N.H. Sallam, F. Ezzeldin, A. Farrag, A.F. Agameya (Egypt)
- 16:00 **FC0101**
ENDOMETRIAL BIOPSY PRIOR TO ARTIFICIAL HORMONE REPLACEMENT FROZEN EMBRYO TRANSFER IMPROVES PREGNANCY AND LIVE BIRTH RATE: A RETROSPECTIVE ANALYSIS
S.L. Yu (Singapore)
- 16:15 **FC0102**
IS FRESH BEST?
I. Rose, K. Sorby, P. Lutjen, T. Osianlis (Australia)
- 16:30 **FC0103**
DAY 4 AND DAY 5 BIOPSY: A COMPARISON OF ANEUPLOIDY AND PREGNANCY RATES
D. Zander-Fox, M. Lane, K. Gebhardt (Australia)
- 16:45 **FC0104**
DOES THE AMH CONCENTRATION PREDICT THE OUTCOME OF IVF IN WOMEN OVER 40?
P. Illingworth, S. Cooke, D. Garrett (Australia)

Free Communication 18 - Female Infertility, Fertility Preservation

15:30 - 17:00

Meeting Room M2

Chairs: Haroon Latif Khan (Pakistan)
Graeme Thompson (Australia)

- 15:30 **FC0105**
ENDOMETRIAL SECRETIONS; PREDICTING RECEPTIVITY AHEAD OF EMBRYO TRANSFER
T.A. Edgell, L. Rombauts, B. Vollenhoven, L.A. Salmonsens (Australia)
- 15:45 **FC0106**
ONE TO NINE YEARS FOLLOW UP OF BABIES BORN AFTER TREATMENT BY AROMATASE INHIBITOR AN OFF LEVEL OVULATION INDUCING AGENT.
R. Begum (Bangladesh)
- 16:00 **FC0107**
PREVALENCE, CAUSES AND RISK FACTORS OF INFERTILITY AMONG NEWLY MARRIED COUPLES IN SHANXI PROVINCE, CHINA - A COHORT STUDY
Q. Meng, A. Ren, L. Zhang, J. Liu, L. Jin, Z. Li, Y. Yang, R. Li, L. Ma (China)
- 16:15 **FC0108**
OUTCOME OF LIVE BIRTH IN MASSIVE ADENOMYOSIS AFTER RESECTION
N. Shafira, Z.R. Wismayanti, C.G. Puspita, D. Yusuf, K. Mutia, B. Wiweko (Indonesia)
- 16:30 **FC0109**
EFFICACY OF LOW MOLECULAR WEIGHTED HEPARIN (LMWH) TREATMENT IN UNEXPLAINED RECURRENT SPONTANEOUS ABORTION (RSA) PATIENTS WITH DECREASED UTERINE BLOOD FLOW
H. KOO, J.Y. Bae, S.H. Cha, H.O. Kim, M.K. Koong, H.J. Yi, Y.J. KIM, K.M. Yang (Korea)
- 16:45 **FC0110**
STUDY OF STRUCTURE AND ULTRASTRUCTURE, APOPTOTIC INCIDENCE AND FOLLICLE MATURATIONAL GENES EXPRESSION OF VITRIFIED-WARMED WHOLE RAT OVARY AFTER AUTOTRANSPLANTATION WITH UNILATERAL AND BILATERAL GONADECTOMY
R. Fathi, M.R. Valojerdi, M. Salehnia (Iran)

"How to ... Session 13" - How to do Ovulation Induction in Low Weight Individuals

15:30 - 16:15

Meeting Room M1

Chair: Pratap Kumar (India)

15:30 Speaker: Vuong Thi Ngoc Lan (Vietnam)

Low body weight is common in anovulatory Asian, particularly, East Asian population. Ovulation induction in low weight individuals requires different strategies to overweight patients. Clomiphene citrate (CC) is the first choice for ovulation induction in anovulatory patients due to the low risks of ovarian hyperstimulation (OHSS) and multiple pregnancies. For patients who do not respond to CC, an FSH low-dose step-up protocol is usually considered as the next option. A study using a low-dose step-up recFSH protocol for ovulation induction in Vietnamese anovulatory patients with low body mass index (BMI) showed that patients required lower starting dose of FSH (25IU/day), lower total dose of FSH used (484IU), but still achieved a comparable ongoing pregnancy rate (33.9%) compared to studies conducted in Western populations. No OHSS and low multiple pregnancy rate were reported. Such a regimen may have applications in other Asian patient populations with low BMI and similar endocrine profile.

16:00 **Discussion**

"How to ... Session 14" - How to Maximise Pregnancy Rates from an Embryo Transfer

16:15 - 17:00

Meeting Room M1

Chair: Kelton Tremellen (Australia)

16:15 Speaker: Teraporn Vutyavanich (Thailand)

Even in the age of evidence-based medicine, there are many controversies regarding the proper techniques of embryo transfer (ET). There is good evidence of benefit in the use of soft rather than rigid catheter, the performance of ultrasound-guided embryos transfers to ensure placement of embryo(s) 1-2 cm below the fundus, slow expulsion of embryo(s), and the use of hyaluronic acid in transfer media. The time that the embryo(s) remain in the catheter should not exceed 60 seconds. There is no evidence of benefit of having a full bladder, removal of cervical mucus, flushing of the endocervical canal, or the use prophylactic antibiotics or antispasmodic agents at the time of transfer. Delayed removal of transfer catheter, bed rest and mechanical closure of the cervical canal after the transfer are of no benefit. The "ease" of transfer, trial transfer, and the level of physician experience may also determine the outcome of ET.

16:45 **Discussion**

ASPIRE Annual General Meeting

17:00 - 18:00

Great Hall Door 6 & 7

Social Function: ASPIRE Gala Dinner

19:00 - 22:30

Brisbane City Hall

SCIENTIFIC PROGRAMME

Sunday, 6 April 2014





PATIENT-CENTRED ART: USING BIOMARKERS TO OPTIMISE OVARIAN STIMULATION

Saturday 5 April 2014 • 11:00am – 12:30pm
Great Hall Door 8
Brisbane Convention & Exhibition Centre
Brisbane, Australia

Co-chairs:

Robert Norman (Australia)
Pak Chung Ho (Hong Kong)

AGENDA

AMH: The start of the beginning

Richard Fleming (UK)

Biomarkers, evolution of use in ovarian stimulation – the Asia-Pacific experience

Vuong Thi Ngoc Lan (Vietnam)

Biomarkers, my use and your use to individualise ovarian stimulation

Richard Paulson (USA)

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Sunday, April 06, 2014

Concurrent Session 11 - The Future Of PCOS

08:30 - 10:00

Great Hall Door 6

Chairs: Benjamin Kroon (Australia)
Helena Teede (Australia)

08:30 PCOS in East Asians

Speaker: Jie Qiao (China)

Aim: To understand and evaluate the current PCOS status in Asian patients, including the prevalence, clinical and metabolic features, and PCOS-related risks.

Method: Systematic review of articles published in English before July 2013 and identified using the PubMed search engine.

Results: There is considerable ethnic variation in the expression of PCOS in Asian patients, including the prevalence and severity of obesity, insulin resistance, metabolic disturbances, and their correlates. Overall, East Asian women with PCOS have a lower BMI and a milder hyperandrogenic phenotype, but with the highest prevalence of metabolic syndrome. South Asians in particular have a high prevalence of insulin resistance and metabolic syndrome, and are at risk for type 2 diabetes, with central obesity more than BMI reflecting their metabolic risk. South East Asians with PCOS have a higher mean BMI than South Asian patients, but have comparable prevalence of metabolic syndrome. Acanthosis nigricans is a common clinical indicator of greater metabolic risk in South Asian and South East Asian women, although it is not included as a clinical marker in diagnosing PCOS. Genetic differences, different environments, cultures and lifestyles, and diet-induced genetic modifications might explain such wide phenotypic variation within these groups.

Conclusion: Ethnic difference of PCOS in Asian patients appears to be linked to the variety of hyperandrogenism, obesity, insulin resistance, and metabolic problems. Further assessment of the variations of PCOS among different ethnic groups in Asia is required in a standard definition, and ethnicity-specific guidelines are needed for identifying anthropometric thresholds for appropriate screening and diagnosis in high-risk ethnic groups.

09:00 Health Implication of PCOS in Australia

Speaker: Roger Hart (Australia)

The polycystic ovary syndrome (PCOS) is the commonest endocrine disorder of women in the reproductive age group. Apart from the reproductive difficulties associated with infrequent ovulation and the associated inconvenience of irregular menstrual cycles, it is believed that the condition is associated with increased metabolic risk and potentially endometrial cancer for women with the condition. Furthermore many reports suggest that after conceiving the patient with PCOS is at an increased risk of pregnancy related complications. In this talk the literature pertaining to women with PCOS will be reviewed and data from the Western Australian Health department derived by data linkage will be reviewed.

09:30 PCOS in South Asians

Speaker: Abha Majumdar (India)

PCOS is the most common endocrinological problem affecting women in their reproductive age. Its manifestations start at puberty and adolescence, with noticeable increase in prevalence of menstrual irregularities and symptoms of androgen excess, leading to fertility issues owing to anovulatory reproductive difficulties later in life. Various studies report ethnic variation in the expression of reproductive as well as non-reproductive symptoms of PCOS due to genetic and non genetic nuances in their makeup.

In PCOS women from south Asia, one third appear to have metabolic syndrome where altered waist hip ratio and hyper-androgenism appears to be a major determinant, apart from obesity being described as the most important risk factor. A screening policy of all PCOS especially with central obesity and androgen excess, for metabolic syndrome with its antecedent risk of cardiovascular disease and type 2 diabetes mellitus, in a low resource setting, is highly recommended.

Concurrent Session 12 - The Future For Preconception Planning: What The Patient Should Know About Preconception Care

08:30 - 10:00

Great Hall Door 8

Chairs: Louise Johnson (Australia)
 Karin Hammarberg (Australia)

08:30 Preconception Health – What The Clinician Should Talk about with Patients

Speaker: Tony Chung (Hong Kong, China)

Reproductive medicine has revolutionised the way infertility is managed in the last 30 years. There have been a succession of breakthroughs which in historical terms are revolutionary. However, there are many issues that have arisen with this success. Whilst some of these issues are being addressed, such as multiple pregnancies, many others remain largely neglected. This complex medical, economic, sociological, religious and political development requires attention from the people who practice reproductive medicine.

08:50 Preconception Health – Research and Action at a Population Level

Speaker: Karin Hammarberg (Australia)

Current evidence about the adverse effects of increasing age, obesity, and other lifestyle factors on fertility and pregnancy health will be presented. To help people make informed childbearing decisions, knowledge about the importance of optimising preconception health to improve fertility, reduce the risk of obstetric and perinatal complications, and give the baby a healthy start to life is crucial. *Your Fertility* is a government funded initiative to promote preconception health. Through its website www.yourfertility.org.au evidence-based information and other resources about factors that affect fertility and pregnancy health are available for the general public and health professionals. The potential barriers for uptake of preconception health promotion messages and *Your Fertility's* strategies to overcome these will be discussed.

09:10 Preconception Care in Developing Countries

Speaker: Kamini Rao (India)

Each day, approximately a thousand women die due to various causes attributable to pregnancy and childbirth; mostly in developing countries and majority are due to preventable reasons. Hence a "Continuum of care" model starting preconceptionally through pregnancy and childbirth needs to be formulated to avert this. "Preconception care" is the provision of biomedical behavior and social health interventions to women and couples prior to conception, aimed at improving their health status and reducing behavior and individual and environmental factors that contribute to poor maternal and child health outcomes; as per the 'WHO'. This would effectively translate into improved short term and long term maternal and child health outcomes. Implementing appropriate 'Preconception Care interventions' like treatment of nutritional deficiencies, modifying environmental risks, screening and treatment of sexually transmitted diseases and counseling for genetic disorders can make a difference in reducing the burden of maternal and childhood morbidity and mortality in developing countries.

09:30 Panel Discussion

Panelists: Cailin Jordan (Australia)
 Antony Lighten (Australia)
 Tony Chung (Hong Kong, China)
 Karin Hammarberg (Australia)
 Kamini Rao (India)

09:50 Q&A

Free Communication 19 - ART, Clinical

08:30 - 10:00

Meeting Room M1

Chairs: Sabina Shrestha (Nepal)
Tongtis Tongyai (Thailand)

- 08:30 **FC111**
COMPARISON OF CLINICAL RESULTS FOLLOWING SPLIT CYCLES OF IN VITRO FERTILIZATION (IVF) AND INTRACYTOPLASMIC SPERM INJECTION (ICSI)
K. Kyono, Y. Nakamura, T. Ikeno, Y. Sato, T. Kyoya, H. Hattori, Y. Nakajo (Japan)
- 08:45 **FC112**
BIRTH WEIGHT FOLLOWING VITRIFIED-WARMED EMBRYO TRANSFER WAS HIGHER THAN THOSE IN NON-ART AND FRESH EMBRYO TRANSFER.
M. Shibata, K. Ito, E. Tsukuda, M. Satoh, Y. Akamatsu, S. Hashimoto, T. Himeno, T. Inoue, Y. Nakaoka, Y. Morimoto (Japan)
- 09:00 **FC113**
A PROSPECTIVE RANDOMIZED CONTROLLED TRIAL STUDYING THE NEED FOR HOSPITALISATION OF SURROGATES FOR A BETTER PREGNANCY OUTCOME
M. Singh, P.C. Jindal, R. Singh, A. Jindal (India)
- 09:15 **FC114**
CC-HMG combined with Growth Hormone in Ministimulation Protocol may Improve Clinical Outcome in Poor Ovarian Responders
A. Polim, I. Sini, I. Anwar, C. Hutomo, A. Pradana, A. Kurnia (Indonesia)
- 09:30 **FC115**
ENDOSCOPIC EMBRYO TRANSFER AND IMPLANTATION(HEED AND SEED) - IMPROVING SUCCESS AND DECREASING RISKS
M. Kamrava, M. Yin (USA)
- 09:45 **FC116**
DUAL EMBRYO TRANSFER: BENEFITS AND RISKS.
A. Smirnova, M. Anshina, N. Shamugia, D. Zhordanidze, K. Ilyin, S. Sergeev, I. Kalinina (Russia)

Coffee Break and Exhibition Viewing

10:00 - 10:30

Great Hall Door 1

Concurrent Session 13 - The Future Treatment Of Endometriosis

10:30 - 12:00

Great Hall Door 6

Chairs: David Molloy (Australia)
 Zi-Jiang Chen (China)

10:30 Advances in Clinical Management
 Speaker: Luk Rombauts (Australia)

The optimal management of endometriosis often relies on index surgery performed under ideal circumstances followed by individually-tailored medical treatment to manage residual symptoms and the risk of disease recurrence. New developments in imaging techniques have dramatically changed the preoperative planning of the patient with severe endometriosis. These patients require and deserve a streamlined multidisciplinary approach. Mapping the deep infiltrating endometriosis prior to surgery has significantly reduced the risk of re-operation. Nevertheless, current management often falls short in delivering acceptable outcomes for patients. Given the prevalence, the physical and psychological impact and the health-economic burden of endometriosis, it is vital that new medical treatments are developed which are better tolerated and more effective. An overview of the pharmacological drugs in development will highlight new agents in the pipeline, including new GnRH-antagonists, immune-modulators, angiogenesis inhibitors, selective progesterone receptor modulators, anti-oxidants and new classes of pain-killers.

11:00 Biomarkers of Endometriosis
 Speaker: Chii-Ruey Tzeng (Taiwan)

Endometriosis occurs when shed endometrium from the female reproductive tract grows outside the uterus, which might cause infertility and dysmenorrhea. The causes and mechanisms of endometriosis remain unclear so far. Moreover, the current diagnosis still needs invasiveness laparoscopy. Our current studies based on cDNA microarray, we have discovered several genes including; integrins/cell adhesion molecules, metalloproteinases (MMP 14), cathepsins, estrogen receptors, VEGFs, Tensin-1, Osteopontin, and cytokines. By the proteomic approach, we have indentified Alpha-1 antitrypsin (a1-AT) as a potential marker which can be down-regulated by long-acting GnRH agonist (GnRHa) treatment. Hence, we aimed to combine functional genomics, microRNA, proteomic analysis and clinical validation to study the gene expression pattern, mechanism and signal transduction of endometriosis and evaluate the efficacy of employing long-acting GnRHa in patients with endometriosis before IUI or IVF treatment. Our final goal is to develop the diagnostic kit or chip for early diagnosis and follow-up the disease.

11:30 Genetics of Endometriosis
 Speaker: Jenny Fung (Australia)

Endometriosis is a common disease that affects 6-10% of women during their reproductive years. Genome-wide association studies (GWAS) have revealed associations with endometriosis at seven genomic regions. A critical step to enable translation of our results is to identify the genes and pathways contributing to endometriosis risk from these regions and to characterize their functional effects. GWAS studies show markers associated with disease risk are generally located in regulatory regions of the genome and likely to affect expression of specific genes. We are investigating gene expression in both blood from 862 individuals and endometrial tissue from 157 women. By integrating with the genotype data, our results show "cis" regulatory effects on expression of 40% of genes and some "trans" effects on the expression of genes on the other chromosomes. Understanding the role of GWAS "hits" on gene expression in endometriosis is the first step in further translation of our results.

Concurrent Session 14 - The Future Of Andrology Symposium

10:30 - 12:00

Great Hall Door 8

Chairs: Derek Lok (Australia)
Craig Niederberger (USA)

10:30 **Assessing the Male**

Speaker: Craig Niederberger (USA)

In this age of precision medicine, the evaluation of the infertile male includes novel and specific means of assessing male reproductive potential, which include endocrine assays and analysis of semen. However, understanding the new is only half of the puzzle; our view of traditional tests such as the bulk semen analysis has also evolved markedly. This talk will dive into the modern evaluation of male reproductive function and consider not only what is currently available, but interpretation in a patient specific context with the features and limitations of each assay discussed. Knowing what to order is the first step; knowing what not to order is the second; and understanding the results of what was ordered is the largest step forward in evaluating male reproductive function and dysfunction.

11:00 **Sperm Extraction in ART**

Speaker: Ju Tae Seo (Korea)

Approximately 20% of men visiting for infertility are azoospermia. Of these patients, about 40% have obstructive azoospermia (OA). OA may result from previous vasectomy, epididymal, vasal or ejaculatory duct abnormalities like congenital bilateral absence of vas deferens (CBAVD). In OA patient, surgically correction may try to restore for fertility. However, TESE/ICSI should be needed in case of surgically uncorrectable cases or CBAVD. With advent of ICSI in conjunction with sperm retrieval via testicular sperm extraction (TESE), many of non-obstructive azoospermic patients are able to father own babies. Also TESE/ICSI is successful in intervention in Klinefelter syndrome. However, 20-50% of NOA patients are not able to have sperm retrieved for ART. Microsurgical TESE is an advanced type of TESE that applies microsurgical techniques. Microsurgical TESE is effective sperm retrieval from men with NOA for ICSI. Microsurgical TESE is more effective in men with NOA than conventional TESE.

11:30 **Hormonal Approaches to Male Fertility Regulation**

Speaker: Robert McLachlan (Australia)

Hormonal approaches are used to promote (male infertility) or impede (contraception) spermatogenesis. Hypogonadotropic hypogonadism is uncommon but provides the opportunity to restore natural fertility using gonadotrophin therapy in ~70% and >90% of prenatal- and post pubertal onset cases, respectively. Most infertility arises from poor spermatogenesis of unknown cause and specific treatment is unavailable; empirical or unproven hormonal approaches need proper evaluation.

New safe, effective and reversible male contraceptives are required. Male hormonal contraception relies on gonadotrophin suppression by exogenous androgens, often combined with progestin. Over several months sperm densities fall to <1 million/ml with contraceptive efficacy similar to female methods. Inadequate suppression in ~5% of men probably results from residual intratesticular androgen action. Despite many encouraging human studies, including a recent multinational trial sponsored by the WHO and CONRAD, development has stalled with the withdrawal of industry support due to uncertainty about the risk: benefit ratio.

Free Communication 20 - ART Basic, Embryology and Laboratory

10:30 - 12:00

Meeting Room M1

Chairs: Aisaku Fukuda (Japan)

Deirdre Zander-Fox (Australia)

10:30 **FC117**

QUALITY OF SIBLING CLEAVAGE-STAGE EMBRYOS GENERATED IN SYNTHETIC PROTEIN-FREE AND CONVENTIONAL PROTEIN-CONTAINING COMMERCIAL EMBRYO CULTURE MEDIA

J. Ali, N. Razali, M. Arasu, H.A. Tarmizi, S. Mahfudzah, A. Arjunan, G.A. Ata'Allah, M. Ramalingam, M.M. Rashid, S. Hanafiah, H. Mohammad, S.Z. Omar, N.A. Mat Adenan (Malaysia)

10:45 **FC118**

INCIDENCE OF MOSAICISM IN CHROMOSOMALLY ABNORMAL HUMAN EMBRYOS DETECTED BY ARRAY-CGH.

R. Kobayashi, A. Ohgaki, H. Matsumoto, S. Mizuno, A. Fukuda, Y. Morimoto (Japan)

11:00 **FC119**

SINGLE VITRIFIED EMBRYO TRANSFER OF BLASTOCYSTS CULTURED IN DEFINED MEDIA WITH RECOMBINANT HUMAN ALBUMIN: A RANDOMIZED TRIAL

M. Murakami, A. Egashira, T. Kuramoto (Japan)

11:15 **FC120**

INFLUENCE OF BLASTOCYST PRE-FREEZE MORPHOLOGY & RE-EXPANSION POST WARMING ON PREGNANCY RATES IN VITRIFICATION CYCLES.

S. Chopra, G. Majumdar, M. Kochhar, A. Majumdar (India)

11:30 **FC121**

OOCYTE AND GRANULOSA-CUMULUS CELLS INTERACTIONS: LH RECEPTOR MRNA EXPRESSION ON THE OOCYTE QUALITY

A. Bowolaksono, B. Wiweko, K. Mutia, P. Ameilia Iffanolida, F. Alwahida, A. Hestiantoro (Indonesia)

11:45 **FC122**

BENEFICIAL EFFECTS OF RESVERATROL ON BOVINE OOCYTE MATURATION AND SUBSEQUENT EMBRYONIC DEVELOPMENT AFTER IVF

F. Wang, X.Z. Tian, L. Zhang, C.J. He, P.Y. Ji, Y. Li, G.S. Liu (China)

Lunch and Poster Session

12:00 - 13:00

Great Hall Door 1

Late-Breaking Lectures

13:00 - 14:00

Great Hall Door 6 & 7

13:00 **What is the Most Cost-Effective Gonadotropin Treatment for Poor Responders?**

Speaker: Virochana Kaul (Australia)

13:25 **Genome and Transcriptome Analyses of Single Human Oocytes and Preimplantation Embryos**

Speaker: Jie Qiao (CHINA)

Special Symposium 4 - ASPIRE in 2014 onwards

14:00 - 14:30

Great Hall Door 6 & 7

Chairs: Bruno Lunenfeld (Israel)

PC Wong (Singapore)

14:00 Speaker: Jaideep Malhotra (India)

ASPIRE is a Young Vibrant fast growing organization with its horizons reaching from highly developed to the most underdeveloped countries in the region. With Infertility on the rise and a lot of awareness and holistic approach is required towards helping our patients achieve access to good quality infertility care. I feel ASPIRE being the largest body in the region , has a responsibility towards understanding the related issues and bringing these countries together, by providing an academic platform for the exchange of innovative ideas and also creating opportunities for the young to learn the newest techniques in the field of ART. ASPIRE should educate and help upgrade and also start a process of accreditation, which will standardise the practice in the region. I am looking forward to consolidating the efforts put in by my predecessors and have plans to take steps towards achieving and encouraging the goals of evidence based practice which will make an impact ,not only in the region but on the whole world.

Closing Ceremony

14:30 - 15:00

Great Hall Door 6 & 7

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FREE COMMUNICATION ABSTRACTS

FC001

ANALYSIS OF BOTH MORPHOKINETICS AND BLASTOCYST METABOLISM TO DEVELOP A COMBINED QUANTITATIVE PREDICTOR OF VIABILITY

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Previous research has shown that morphokinetics and metabolism of the preimplantation embryo can be used independently as markers of developmental competence and subsequent viability. The aim of this study was to determine the inter-relationship of cleavage stage embryo kinetics and blastocyst quality, as determined by cell number and metabolism of carbohydrates and amino acids. Morphokinetics of *in vitro* fertilised C57BL/6xCBA (F1) mouse zygotes were analysed using a Sanyo time-lapse incubator with continuous imaging capability and embryos were sorted into quartiles with respect to cleavage time to 2-cell. The embryos observed with faster cleavage times (first quartile, designated 'fast') were found on average to be 2.7h ahead of the slower embryos (fourth quartile, designated 'slow', 11.5 ± 0.1 h vs. 14.2 ± 0.1 h, $p < 0.0001$). Subsequently on day 5, blastocysts developed from 'fast' embryos showed a higher ratio of inner cell mass to trophectoderm cells (0.2 ± 0.1 vs. 0.1 ± 0.02 , $p < 0.05$) and a significantly lower glycolytic rate compared to 'slow' embryos ($54.4 \pm 3.1\%$ vs. $67.3 \pm 5.1\%$, $p < 0.05$). Further non-invasive metabolomic analysis revealed that 'fast' blastocysts consumed more aspartate than 'slow' blastocysts (2.8 ± 0.1 pmol/embryo/h vs. 2.2 ± 0.2 pmol/embryo/h, $p < 0.01$) and consumed rather than produce glutamate (0.2 ± 0.1 pmol/embryo/h vs. 0.1 ± 0.1 pmol/embryo/h, $p < 0.05$). These findings suggest that kinetically different embryos develop into blastocysts with different metabolic profiles. Ongoing work is now using these viability markers in combination to increase embryo selection efficacy, in order to further improve implantation and pregnancy rates.

FC002

DNA DAMAGE AND VACUOLES: WHEN DOES SPERM VACUOLATION BECOME IMPORTANT?

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Aim:

Sperm head vacuoles and their size have been associated with sperm DNA damage⁽¹⁾, and decreased pregnancy rate⁽²⁾. Data on the significance of the varying vacuole volumes have been limited to a small number of patients and sperm.

The aim of this study was to assess the relationship between the DNA Fragmentation Index of a sperm sample and the vacuole volume within the sperm head.

Method:

312 semen samples had 400 motile sperm assessed for the presence of vacuoles at x7026 magnification using IMSI Strict (Hamilton Thorne). Sperm were grouped into 3 categories for vacuole size – 0-4%, 5-10% and >10%.

The DFI of each sample was assessed using SCSA. DFI ranges⁽³⁾ were - Low= $\leq 15\%$, Mid= $15-29\%$, High= $\geq 30\%$.

Results:

The % size of vacuoles increase as the DFI increases. There is a significant difference between the Low and Mid range groups ($p < 0.01$) and the Low and High range groups ($p < 0.01$).

DFI range	Vacuole size (%)			X ²
	n	0-4%	5-10%	>10%
Low	218	55067/87200 (63.2%)	26004/87200 (29.8%)	6148/87200 (7.0%) (a) (b)
Mid	75	16908/30000 (56.4%)	9607/30000 (32.0%)	3485/30000 (11.6%) (a) (a) p=<0.01
High	19	3883/7600 (51.1%)	2172/7600 (28.6%)	1545/7600 (20.3%) (b) (b) p=<0.01

Conclusion:

(a) Sperm head vacuoles significantly increase with increasing DNA damage in sperm,

(b) Whilst WHO 5th edition suggests vacuoles >20% should be recorded, we propose that 10% vacuolation of sperm heads is significant .

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FC003

USE OF HIGH CALCIUM MEDIUM ON PATIENTS FOLLOWING POOR OR FAILURE TO FERTILIZE ICSI CYCLES.

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Aim

The aim of this study was to evaluate the impact of high calcium medium on fertilization and subsequent embryo development for patients that had prior ICSI cycles with failed or poor fertilization.

Method

Retrospective analysis of 14 patients between 2009 – 2013 that received high calcium supplementation in an ICSI cycle following previous poor or failure to fertilize cycles. The high calcium (Fertibooster) suite of Genea media consists of 3 solutions: Sperm holding medium; ICSI sperm medium and post-ICSI cleavage medium for culture of injected oocytes. Fertilization rates and quality of cleavage and blastocyst embryos were compared between the poor or failure to fertilize cycles and the subsequent cycles treated with Fertibooster.

Results

Fertilization rates between the two groups were found to be significantly different (p=0.01) with previous poor or failure to fertilize cycles rate of 27% (32/118) compared with the Fertibooster cycles of 44% (45/103). The percentage of good quality cleavage and blastocyst stage embryos increased with the use of the Fertibooster media, this was not significant.

Conclusion

Treatment with high calcium significantly improves ICSI fertilization rates. There is a trend towards better quality embryos at the cleavage and blastocyst stage with high calcium treatment; however this was not statistically significant. The results suggest that the Fertibooster suite may be a useful tool in improving ICSI outcomes for patients that would otherwise have a minimal chance of achieving a pregnancy. The numbers presented are small and would benefit from confirmation of these findings with a larger study.

FC004

HIGH FERTILIZATION AND LOW ZYGOTE ARREST RATES ARE INDICATORS OF GOOD LABORATORY PRACTICE IN ASSISTED REPRODUCTION TECHNOLOGY

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Objective: The objective of this study is to identify indicators of laboratory quality that could enable workers aim for high clinical pregnancy rates (CPRs) per oocyte retrieval (OR) for day 2 embryos.

Materials and methods:

Retrospective analysis of laboratory data during 2 periods when CPR was good (48.2% & 46.9%) or during two periods of very poor CPR (16%&17.2%) respectively was undertaken. Parameters investigated were: fertilization rate (FR), zygote arrest rate (ZAR), mean blastomere number (MBN), mean embryo grade (MEG), proportion at or above normal blastomere number (%≥NBN), proportion of embryos at or above a score of "good" quality (%≥GEQ).

Results: Fertilization (FR) and zygote arrest rates (ZAR) appear to impact CPR more consistently than other parameters. Fertilization rates appears to have a direct effect; higher the FR, higher the CPR (FRs of 80.4, 75.5, 53.3, 56.4% showed CPRs of 48.2, 46.9, 17.2, 16.0% respectively) whereas the ZAR has an inverse effect such that the lower the arrest rate, the higher the CPR (ZARs of 3.4, 7.7, 10.3, 17.7% gave CPRs of 48.2, 46.9, 17.2, 16.0% respectively). These parameters were significantly different between groups that gave higher CPRs (48.2%, 46.9%), compared to the group with a poor CPR (16%&17.2%).

Discussion and conclusion: High FR and low ZAR are very sensitive indicators of quality of in vitro culture conditions impacting CPR. FR and ZAR can be utilized to monitor and ensure culture conditions are maintained at highest standards of quality. High FR and low ZAR are indicators of good laboratory practice.

FC005

GETTING TO ELECTIVE SINGLE EMBRYO TRANSFER WITH BLASTOCYST CULTURE AND PREIMPLANTATION GENETIC SCREENING FOR PATIENTS OF ALL AGES

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Preimplantation genetic screening (PGS) can allow wider application of eSET, since patients can transfer a single euploid embryo to the uterus and expect high implantation rates. In the current study, we performed PGS on day 5 and 6 blastocysts following biopsy. We report outcomes for patients transferring only chromosomally normal embryos.

Embryos were cultured in sequential medium (G1 and G2; Vitrolife) and blastocysts were biopsied and then vitrified on day 5 or 6 of development. Trophectoderm cells were removed and sent for chromosome analysis (Natera), and the embryos were collapsed and vitrified shortly after biopsy. When patients returned for transfer they were strongly encouraged to transfer one embryo regardless of their age. The embryos were transferred within 1 hour of warming to the uterus on day 6 of progesterone in a natural or controlled cycle as appropriate.

From 95 warming cycles, an overall clinical pregnancy rate of 67% and implantation rate of 57% was achieved, and most patients warmed and transferred 1 embryo. No differences were seen in the proportion of embryos that were abnormal on day 5 or 6, and the number of retrievals with biopsy decreased with patient age as expected, as did the number of euploid embryos.

Good pregnancy/implantation rates were achieved across all age groups. Older patients were less likely to have any embryos biopsied or to have normal embryos available after PGS, but this allowed them to avoid unnecessary transfers. The application of PGS facilitated the widespread use of eSET, even in older patients.

FC006

TIME LAPSE IMAGING AND GENE EXPRESSION PROFILING IN CUMULUS CELLS AS PREDICTORS OF BLASTOCYST QUALITY

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Non-invasive markers of embryo quality have the potential to improve IVF success. Promising non-invasive markers of embryo quality include time-lapse imaging analysis of early cleavage events and gene expression profiling in cumulus cells.

While these markers have been studied individually, our study aimed to combine time-lapse imaging analysis with gene expression profiling in cumulus cells and correlate this with subsequent blastocyst quality. Embryos underwent time-lapse imaging analysis for early cleavage events using the Primo Vision monitoring system. For 80 of these embryos, individual cumulus cell masses had the expression levels of 29 genes that are important indicators of follicular microenvironment evaluated. These genes included those involved in cumulus cell expansion, signalling and energy metabolism. Time-lapse imaging analysis and gene expression profiles were analysed in blastocysts graded as high or low grade, or embryos that had arrested on day 5. Our findings show that time-lapse imaging analysis of early cleavage uncover events which are predictive of blastocyst quality on day 5, and that these patterns of development in the embryo reflect the gene expression profile of the cumulus cells. This study provides further evidence that the developmental potential of embryos is highly reflective of the follicular environment that the oocyte completed development. Additionally, it is possible that combining time-lapse analysis of early cleavage events with gene expression profiling in cumulus cells may provide a more reliable marker of embryo quality.

FC007

MOTIVATIONS AND EXPERIENCES OF PATIENTS SEEKING CROSS BORDER REPRODUCTIVE CARE: THE AUSTRALIAN AND NEW ZEALAND CONTEXT

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Over the past years increased attention has been paid to cross border reproductive care (CBRC) as practitioners and consumer groups report growing use of fertility treatment outside of consumers' home countries. Concern has been expressed regarding the quality of care received, access to implications counselling, the ability to engage in informed decision-making, and the consequences of accessing treatment in jurisdictions with different legal frameworks from Australia and New Zealand.

This study sought to investigate the motivations and experiences of residents engaging in treatment outside Australia and New Zealand. Quantitative and qualitative data was collected via an online anonymous questionnaire, and analysed using descriptive statistics and thematic analysis.

Results from 139 participants suggest that motivations for engaging in CBRC include: treatment sought not being readily available in the home state (either for practical reasons, such as limited donor and surrogate availability) or legislative reasons (such as prohibition of gender selection); difficulty in meeting access criteria, and perceived length of time to access treatment. Whilst donor availability is a motivating factor, the majority were not seeking anonymous donors. Experiences of care received were generally rated positively in terms of costs, safety and information provided. However, less than half received professional counselling, and most relied on non-clinic sources for support. Participants indicated that access to post-treatment counselling, particularly with regards to their emotional wellbeing, legal implications, and donor-conceived children, would be useful.

The implications of findings for policy and the provision of best-practice psychosocial support for patients considering future CBRC are discussed.

FC008

AN EXAMINATION OF RECIPIENTS' EXPERIENCE OF INTRA-FAMILIAL SPERM DONATION

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Background

Heterosexual couples requiring sperm donation often turn to a father or brother for assistance. Very little is known about the outcomes for such families.

Aims

This qualitative study explores the decision-making surrounding father to son and brother to brother sperm donation, how their relationships are impacted once a child is born and how disclosure to others and to the child is viewed and managed. The aim was to learn more about psychosocial outcomes in order to inform counselling for couples contemplating this option.

Method

Relevant couples who had been through successful treatment at Geneva in the past 10 years were approached for recruitment. Out of 23 couples approached, 9 consented to in-depth guided interviews.

Results

The importance of a genetic relationship was the primary reason couples opted for intra-familial donation. After initial concerns about how relationships would develop, recipients were satisfied with the donors' level of involvement, as couples assumed their normal roles of grandparents or uncle and aunt. Disclosure was the most difficult issue, with views ranging from complete openness to total secrecy. Typically, recipients whose brother was the donor were more comfortable with telling than those whose father was the donor. This included disclosure to other close family members, notably parents and siblings.

Conclusion

Relationships in both inter-generational and intra-generational donation were experienced as developing normally, with boundaries being maintained. Disclosure was an individual decision and an on-going challenge for all couples. Further research into longer term outcomes would be useful.

FC009

HEALTH NUMERACY AND RISK AVERSION IN INFERTILE PATIENTS

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Introduction:

Low numeracy affects the way patients compute health-related information and can lead to bias in judging health risks. Infertile couples often balance the risks and benefits of IVF interventions, such as when they decide to transfer one or two embryos. The aim of this study was to assess risk perception and risk aversion in infertile and fertile couples and to correlate this with health numeracy.

Methods:

642 subjects were surveyed. Objective and subjective numeracy was assessed with validated scales and correlated with Likert scale responses to questions assessing risk perception and aversion. Spearman's rank-order test was used to identify significant correlations.

Results:

Numeracy was only weakly correlated with risk perception or bias. Fertile and infertile women were more likely than men to worry about pregnancy risks and this was correlated with their perception that they were at higher than average risk (inferiority bias). Infertile men and women felt they needed to take greater risks because they had been trying for a long time, a sentiment that was strongly correlated with their view that having a baby is more important than the potential risks. In infertile couples concern for the risks of amniocentesis was highly correlated with their attitude towards increased risks of cerebral palsy in multiple pregnancies.

Conclusions:

Infertile couples were more worried about pregnancy risks than fertile couples and yet they felt they needed to take more risks. The short-term goal of a pregnancy appeared to outweigh any risks associated with the longer-term goal of a healthy baby.

FC010

ASSISTED CONCEPTION, MATERNAL AGE AND BREASTFEEDING: AN AUSTRALIAN COHORT STUDY

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Aim: To establish the relationships among age, mode of conception and breastfeeding.

Method: Consecutive cohorts of nulliparous women >25 weeks pregnant who had conceived through ART (ARTC) or spontaneously (SC) in three age-groups ≤30, 31-36 and ≥37 years were recruited. Data were obtained via telephone interviews and postal questionnaires in late pregnancy and four months postpartum. Sociodemographic characteristics, reproductive health, birth and breastfeeding experiences were assessed by study-specific questions. Self-rated general health and symptoms of depression and anxiety were assessed with standardized psychometric instruments. Main outcomes were exclusive breastfeeding at discharge from maternity hospital and four months postpartum.

Results: Of 1179 eligible women, 791 (67%) participated; 549 (93%) had singleton infants, provided complete data and were included in analyses. Overall 37.2% of participants aged ≤30; 33% aged 31–36 and 55.1% aged ≥37 years experienced caesarean births. Regardless of age, compared with the SC group, ARTC women had twice the rate of caesareans prior to labour. Controlling for other factors, exclusive breastfeeding rates at hospital discharge and four months postpartum were lowest among ARTC women who experienced caesarean prior to labour ($p < .001$).

Conclusion: Independent of age, assisted conception increases the risk conferred by caesarean birth, to breastfeeding initiation and maintenance.

FC011

INFERTILITY INFORMATION: ACCESS , ACCURACY AND LIABILITY IN INDONESIA, NOVEMBER 2013

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Infertility is one of reproduction problem that has a great effect and burden for the infertile couple. In recent years, the development of information technology and easier access to all kind of digital information, making the infertility information has a greater demand. However the information for infertility couple in Bahasa is very limited and contain of this information is various and questionable. The aim of this study to explore about the digital information related to infertility in Bahasa from the Google search engine and to know the accuracy and liability of the information. The accuracy and the liability of the information match with the guideline like NICE guideline, ASRM, ACOG and RCOG. From the Google search engine with the keyword of "infertilitas" results 1020 articles, keyword for infertility etiology "penyebab infertilitas" has 392000 results, and with the infertility clinic keyword has 133000 results. The information contain from 30 infertility clinics website all over Indonesia, 73.3 % explain about the definition of infertility, only 20% explain about the pathophysiology of infertility problem, 66.7% explain about the etiology of infertility, 46.7% explain about the symptom of infertility and 50% explain about the diagnosis of infertility, 53.3% explain about the treatment of infertility and 13.3% explain about the psychosocial effect of infertility. These results show us the picture of information that digital information for infertility couple in Bahasa still very limited in number and poor in the accuracy of the information.

Keywords : Infertility Information, Accuracy

FC012

DISTRIBUTION OF LEVEL OF STRESS AMONG INFERTILITY PATIENTS

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Background: Infertile couples often suffer from chronic stress each month if fertilization does not occur. The relationship between stress and infertility make a cycle that will mutually reinforce impact. In the process, the more complex infertility therapies will increase stress and affect the outcome of therapy.

Methods: Sixty-three infertile patients who came to Yasmin IVF Clinic, Dr. Cipto Mangunkusumo General Hospital Jakarta, were given self-assessment questionnaire (self-reporting questionnaire) to know the presence of stress encountered.

Results: Of 63 infertile patients incorporated into this study, 14 (22.3%) were experiencing stress and 49 (77.7%) were not showing symptoms of stress. Of the 20 symptoms listed in the questionnaire, feeling fatigue was the most complain (38.1%). Duration of infertility showed a significant correlation with the level of stress experienced by patients ($p < 0.05$).

Conclusion: Twenty-two percent of infertile patients in the Yasmin IVF Clinic experienced stress, mainly associated with the duration of infertility. The symptoms occurred were physical symptoms and interfered with daily activities. Holistic treatment including psychosocial approach is important in the management of infertility.

Keywords: stress, infertility.

FC013

THE IMPACT OF ORAL CONTRACEPTIVES ON COGNITION

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BACKGROUND: Oral Contraceptives(OCs) containing estrogen and progesterone analogues are the most commonly prescribed medications in young women, but their cognitive impact is unknown. With increasing evidence for neurological effects of reproductive hormones, there is clear rationale for investigation. We examined the impact of OCs on cognition by comparing cognitive performance between pill phases ('active' hormonal vs. placebo 'sugar'), and between progestin classes(old vs. new generation).

METHODS: We recruited healthy volunteers taking OCs to undertake cognitive testing using CogState software at two menstrual cycle points, coinciding with 'active' and 'sugar' pills; order of testing counterbalanced. Data was analysed using SPSS software to perform repeated-measures ANOVAs assessing effects of pill phase and progestin class.

RESULTS: 35 women completed testing. Analysis by pill phase revealed significant improvement in verbal memory during active pills($p=0.03$). Analysis by progestin class showed a main effect between groups in visual memory($p<0.01$) and social-emotional cognition($p=0.01$), with improved performance older generation (androgenic)progestin users in both. An interaction between pill phase and progestin class was seen in verbal learning, whereby new progestin OC users improved verbal learning during active phase, with impairment in old($p=0.02$).

CONCLUSION: These results support previous findings that OC use improves verbal memory, and expand upon the suggestion that different progestins have different impacts according to androgenicity. In this study, older(androgenic) OCs impaired verbal memory(a female-favouring task) and enhanced visual memory(a male-favouring task), with opposite effects seen in users of new (anti-androgenic) progestins. Establishing the cognitive impact of OCs allows women to make better-informed contraceptive choices.

FC014

CLOMIPHENE CITRATE, METFORMIN OR THE COMBINATION OF BOTH, AS FIRST LINE OVULATION INDUCTION DRUG IN POLYCYSTIC OVARIAN SYNDROME ; A RANDOMISED CONTROLLED TRIAL.

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Objective :- To compare Clomiphene Citrate (CC) , metformin or the combination of CC and metformin as first line ovulation induction drug in women with polycystic ovarian syndrome (PCOS).

Design :- Randomised controlled trial.

Setting :- Private Practice

Material and methods

One hundred and twenty infertile women newly diagnosed as PCOS,(ESHRE/ASRM criteria) and treatment naïve, were included in the study.They were randomised into any of the three groups, Group I (Metformin 1500 mg per day) 38 patients , Group II (Clomiphene Citrate in incremental dose of 50 to 150 mg) 42 patients and Group III (CC + Metformin) 40 patients . All patients underwent transvaginal follicular monitoring and timed intercourse. Study period was six months. Ninety nine women completed the study. Primary outcome measures were; rates of ovulation, pregnancy and first trimester loss.

Results:

There was no statistically significant difference between the groups with respect to age ($P = 0.42$) BMI (0.06) and duration of infertility ($P = 0.86$). Ovulation rate was 35.7%, in metformin group; 62.5 % in CC group and 63.6 % in the combination group. Combination group had highest pregnancy rate (54.5%) followed by metformin (35.7 %) and CC (31.25%).

Between the three groups there was no statistically significant difference in both ovulation & pregnancy rates. The P value and odds ratio and 95% CI for ovulation and pregnancy rates comparing the groups are shown in table 1.

Conclusion: Combination of CC and metformin gives best outcome in terms of ovulation and pregnancy in infertile PCOS women.

FC015

EFFECT OF SECOND GENERATION COCS ON SERUM LIPID PROFILES, FASTING BLOOD SUGAR, BLOOD PRESSURE AND BMI IN CHILD BEARING AGE WOMEN IN KHYBERPUKHTUNKHWA PROVINCE-PAISTAN.

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Background: Combined Oral Contraceptives Pills (COCs) are effective and widely used method for contraception. There is a positive relationship between COCs and lipid and carbohydrate metabolism in previous studies. We have seen the effect of duration of COCs (0.3mg norgestrel and 0.03mg ethinyl estradiol) used in tertiary care hospitals of Peshawar Khyber Pukhtunkhwa Pakistan on the lipid and carbohydrate metabolism.

Study Design: This cross sectional analytical study included 100 participants women of child bearing age 14-49yrs using COCs divided in three groups according to the duration of use group A at least 6 month COCs users, group B were 1 year COCs users , group C more than 1 year COCs users. Serum Total cholesterol (TC) , triglyceride (TG), high density lipoprotein cholesterol (HDL-C), low density lipoprotein cholesterol (LDL-C), very low density lipoprotein cholesterol (VLDL-C), fasting blood sugar(FBS) were determined by using standard colorimetric techniques BMI and BP were also measured in all subjects. Their levels were found gradually increasing from 6months to those who are using it for 1 year and more than 1year.

Results:

To estimate the effect of duration of use of combined oral contraceptives on the levels of different biochemical parameters, the results showed significant elevation of cholesterol (p-0.0003), HDL-C (p-0.0229), LDL-C (p-0.0271),VLDL-C (p- 0.0004),Triglycerides (p- 0.0006) levels in the group of more than 1 year users females when compared with 6 months users.

Conclusion: The levels of cholesterol, HDL, LDL, VLDL and Triglyceride levels were found to be increased with the duration of use.

FC016

HYPOTHALAMIC GHRH AND PITUITARY GH GENES EXPRESSION LEVELS IN NEONATAL INHIBIN-IMMUNONEUTRALIZED FEMALE RATS

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The present study has been designed to evaluate the potent role of prepubertal passive immunization against inhibin alpha subunit on mRNA expression level of hypothalamic GHRH and pituitary GH genes. The present study has been conducted by induction of inhibin immunoneutralization during rats female life early as 15-20 days. Forty eight neonate *Wistar* female rats (weighted 24.5±1.32 g., aged 15 days) were randomly assigned to two equal groups; treated and control, injected (ip) with inhibin- α antiserum (1 μ g dissolved in 100 μ l of normal saline) and normal saline (100 μ l), respectively, in the 15th, 16th, 17th, and 20th days of age. Eight females from each group were sacrificed in the 23rd, 30th and 45th days. Hypothalamus and Pituitary tissue samples were obtained for evaluation of mRNA expression level of GHRH and GH genes using qRT-PCR. In all of the experimental periods (23d, 30d and 45d), female rats of treated group showed up-regulation of hypothalamic *GHRH* and pituitary *GH* genes. In conclusion, passive immunization against endogenous circulating inhibin during neonatal age of female rats can perform an important role in sexual maturity, pituitary function and gonadal activity after puberty.

Key words: Inhibin, passive immunization, pituitary, hypothalamus

FC017

SERUM SAMPLE PREPARATION FOR AMH MEASUREMENT WITHIN AN EXTERNAL QUALITY ASSURANCE (EQA) SCHEME: SAMPLE STABILITY UNDER DIFFERENT STORAGE CONDITIONS.

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The measurement of AMH is now an established part of fertility assessment. However, some doubt has been cast recently on the stability of AMH in stored blood, and the potential instability of samples would be of great concern for an AMH EQA scheme designed to monitor within- and between-laboratory variability. Eleven laboratories participated, all using the Beckman Coulter Gen II assay. Two blood samples were collected, centrifuged immediately and the serum removed and allowed to clot. Following removal of the clot, half the serum was stored in a refrigerator (4°C) prior to packaging and posting the next day with the other half being left in storage at 4°C for use in a subsequent distribution. Laboratories were asked to analyse the samples under routine conditions and return results within 4 weeks. No significant difference was seen in AMH concentration between the initial result and that after storage in the fridge for 3 months (7.8 ± 1.7 vs 7.7 ± 1.4 pmol/L) or 17 months (23.2 ± 4.3 vs 20.9 ± 3.9 pmol/L). Similarly, a blood sample collected 3 weeks before distribution but processed immediately gave similar results when the serum was stored at either 4°C (13.2 ± 1.9 pmol/L) or frozen at -20°C (13.3 ± 2.3 pmol/L) for those 3 weeks. Delaying the centrifugation of the blood by 4 hrs compared to immediate centrifugation had no effect (16.7 ± 5.3 vs 17.4 ± 5.6 pmol/L). In summary, blood separated within 4 hrs can be stored in a fridge for up to 17 months without any deterioration prior to distribution within an EQA scheme for the analysis of AMH.

FC018

IS PREMIXING OF SERUM SAMPLES NECESSARY TO OBTAIN REPRODUCIBLE AMH RESULTS USING THE ANSHLAB ASSAY

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Introduction

The widely used Beckman Gen II AMH assay has recently been modified by introducing a sample premixing step, possibly to reduce interference from complement causing inconsistent results, particularly after sample storage. In previous studies we have found that results obtained from pre-mixed samples are significantly higher than those using the original method. A second commercial company, Anshlab, has produced new AMH assay with a different primary antibody. We investigated the stability and reproducibility of AMH measurement using this method and also investigated whether pre-mixing has a similar effect as seen with the Beckman Gen II assay.

Method

Fresh and stored samples from a group of pre-menopausal women were assayed using both assays with and without premixing with assay buffer prior to plating.

Results

Measured AMH levels were on average 2-fold higher using the Anshlab method compared with the Beckman assay, and similar to those using the premix Beckman assay. The Anshlab assay produced consistent AMH measurement when samples were stored up to 48 hours at room temperature or at -20°C but inconsistent results when samples were stored for longer periods. Premixing samples with assay buffer before assay using Anshlab kit also produced on average 40% higher but consistent AMH values regardless of the storage conditions.

Conclusion

Measured AMH levels seemed to be more consistent using the Anshlab kit at least for samples stored up to 48 hours. However, a similar premixing effect was also observed with Anshlab kit which suggests the likely presence of serum interference in the assay.

FC019

HEALTH NUMERACY IN FERTILE AND INFERTILE POPULATIONS

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Introduction:

IVF patients are often faced with complex medical decisions trading off success, risks and costs. This requires an ability to attach meaning to numbers. Low numeracy has been linked to ineffective patient choice and risk communication, reduced treatment compliance and adverse medical outcomes. The aim was to compare the numeracy of infertile females (IF) and males (IM) with those of fertile females (FF) and males (FM).

Methods:

In total, 642 subjects were surveyed. Two validated tools were used to measure health numeracy: the objective numeracy scale (ONS) and the subjective numeracy scale (SNS). Demographic data were used as predictors of numeracy in an ordinal logit regression analysis.

Results:

The correlation between the ONS and SNS was high ($r=0.40$; $p<0.001$). The ONS was significantly higher in IM than in IF and FM and both of these were significantly higher than in FF. The SNS was significantly higher in IM and FM than in IF and this was significantly higher than in FF. Differences in demographic background were important. Higher educational status ($aOR=2.57$), Caucasian descent ($aOR=1.59$), and male gender ($aOR=2.39$) all increased the ONS score, but age had no influence. The SNS scores were increased by higher educational status ($aOR=3.25$) and male gender ($aOR=2.96$), but age and ethnicity had no influence.

Conclusions:

These findings confirm earlier studies in different patient populations showing that gender and education are key determinants of health numeracy. These results may provide guidance on how the medical decision-making process is tailored to the needs of infertile couples.

FC020

GAMETE DONORS' EXPECTATIONS AND EXPERIENCES OF CONTACT WITH THEIR DONOR OFFSPRING

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Background: Little is known about the continuing influence of donor conception on gamete donors' lives. An opportunity to increase knowledge of donors' expectations and experiences of contact with donor offspring recently arose in Victoria, Australia, when donors' views were sought on a parliamentary recommendation to introduce mandatory identification of donors on request from donor offspring, with retrospective effect.

Aim: To investigate donors' expectations and experiences of contact with donor offspring.

Method: Qualitative, semi-structured interviews including open questions.

Results: The potential legislation prompted participation by donors who would not normally identify themselves to researchers or government inquiries. Participants were 36 men and 6 women who donated 1970-1997 believing that identifying information would never be released or their permission would be sought before release to donor offspring. Thematic analysis revealed that most donors did not characterise their intrinsic link to their donor offspring as parental. Some donors acknowledged that offspring may need to know their donor's identity. Donors' expectations ranged from wanting or expecting no contact of any kind to a strong desire for a close relationship. Most had not had contact with their donor offspring; some had experienced anything from an inquiry through a register to a close personal relationship.

Conclusion: Donors' needs and desires are not homogeneous; policy and practice must be sensitive and responsive to a wide range of circumstances and preferences. Decisions made to restrict or facilitate contact or the exchange of information have ramifications for donors as well as for donor-conceived people.

FC021

GENETICS, DUPED DADS AND DISCARDED CHILDREN

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The new genetics can now include DNA fingerprinting for paternity testing. This paper will examine the extent to which the courts should rely on genetic contribution to grant an application to remove legal parentage for fathers in law and the ethical rationale for doing so. It will also consider the impact these actions may have on the child.

Application of the new genetics for paternity testing has created new opportunities for men who can offer evidence that the child they had raised as their own has been proven not to be their genetic offspring. Coexistent with these opportunities, this evolution has also provided new challenges for the law as individuals have used it to avoid future child support payments and even to claim reimbursement for support payments made during the time before a particular family relationship had broken down. Their premise has been to use this evidence to argue for legal recognition of the importance of genetic connection in the construction of family.

CONCLUSION

Information provided by the new genetics carries with it the responsibility for the courts to use it judiciously. It should not be used for punitive retribution to allow fathers to 'discard children' when they discover they are not genetically related to them after they have raised them from birth.

FC022

OLDER MATERNAL AGE, ASSISTED CONCEPTION AND OBSERVED PARENTING QUALITY IN MOTHERS OF TODDLERS.

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Background

Despite the well-established trend to older maternal age at first birth there is little empirical data regarding the parenting quality of older mothers (Pennings, 2013), and most evidence to date is limited to self-report measures, subject to social desirability bias. This study aimed to explore maternal age and mode of conception influences on the quality of mother-toddler interaction.

Method

Participants were 133 primiparous women (81 Spontaneous conception SC; 52 Assisted conception AC; Mean age 33.8 years, SD 4.66) recruited in the third trimester of pregnancy and subsequently observed during a 15-minute free play episode recorded in their home two years later (Mean child age 20.2 months, SD 2.85). Trained coders blind to maternal age and mode of conception assessed emotional availability (EA: Biringen, 2008, 4th Edition).

Results

Multivariate analysis of variance (MANOVA) for maternal EA (sensitivity, structuring, non-hostility, non-intrusiveness), controlling for maternal education indicated significant effects for assisted conception, $F(4,117) = 2.67$, and maternal age, $F(4,117) = 3.59$, $ps < .05$. Univariate tests indicated AC mothers were rated more sensitive (Mean AC= 23.60; Mean SC= 21.30) and non-intrusive, (Mean AC= 24.17; Mean SC= 22.58), $ps < .05$. Older maternal age was associated with more optimal structuring, (Mean AC 24.6; Mean SC = 22.7) $p = .03$.

Discussion

Results indicate assisted conception and older maternal age are independently associated with more optimal maternal emotional availability. Findings are discussed in the context of motivation for parenthood and psychological maturity associated with older maternal age.

FC023

IS THE BEST INTERESTS OF THE CHILD A USEFUL PRINCIPLE? EMERGING ISSUES IN ART PRACTICE.

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Legislation and ethical guidelines that regulate the provision of Assisted Reproductive Technologies (ARTs) in Australia require that consideration of the child's best interests is paramount whilst in New Zealand it is an important consideration. Yet frequently in this situation a woman is not yet a parent and a child doesn't exist. This raises a question of what the welfare principle means in practice, how it is interpreted and how it is applied.

To investigate this question we conducted six focus groups with ART counsellors: three in Australia and three in New Zealand. Focus group size ranged from 3 – 12 participants. Within the groups vignettes were presented of typical cases or situations that could be expected to invoke an application of the welfare principle in ART provision. The group discussions centred on policy requirements and approaches to practice, emerging ethical issues, and tensions in counselling practice. The digital recordings of the focus groups were de-identified and professionally transcribed then subjected to thematic analysis.

Several themes emerged relating to the 'presence' of the child, institutional support for child protection, grounds for refusing treatment, strategies for managing concerning cases, and tensions between moral duties of patient support and child protection. In this paper similarities and differences within these themes will be presented.

There are different beliefs about the role of child protection in ART counselling and inconsistency between state requirements and counselling opportunities. An existing regulatory framework is both helpful yet ineffective.

FC024

"HOW MUCH DO THEY KNOW WHAT THEY DO?" A LOCAL PERCEPTION STUDY TARGETED ON PATIENTS OF ASSISTED REPRODUCTIVE TECHNIQUE (ART) IN HK

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Assisted reproductive technology (ART) has been increasingly used in many countries. There is no local study in Hong Kong (HK) on potential ART patients in assessing their knowledge, perception and expected outcome on the treatment. The present study evaluated the 232 subjects on their knowledge and attitude towards ART. These subjects were recruited during a fertility talk and these subjects have the intention to seek ART treatment.

The study showed that about 40% of subjects overestimated IVF success rate. Their basic knowledge (process, risks, legal aspects) towards ART is generally satisfactory. Most of them consider they do not understand ART treatment, but only 65% would searched for information from different sources. In addition, our results showed that donation of oocytes have not yet accepted by the majority of subjects, regardless of the religious beliefs or age. Further analysis showed the knowledge on the following areas are less satisfactory: number of embryos transferred permitted, possibility of multiple pregnancies, side effects after ART treatments, and the fate of unused embryos. The study showed no significant correlation between knowledge on IVF success rate and respondents' demographic particulars, including gender, age, household income, education level and religious beliefs.

Knowledge about ART is less than satisfactory in a group of subjects anticipating fertility treatment. Subjects tend to overestimate IVF success rate. Education and counseling are important before embarking on ART treatment.

FC025

ANDROGEN-INDUCED MOUSE MODELS FOR POLYCYSTIC OVARY SYNDROME - WHICH ONE IS BEST?

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Polycystic ovary syndrome (PCOS) affects 5-10% of women of reproductive age, causing anovulation, infertility, hyperandrogenism, obesity, hyperinsulinism and increased risk of type 2 diabetes and cardiovascular disease. However, the etiology of PCOS is unclear and ethical and logistic constraints limit definitive experimentation to determine mechanisms involved. We aimed to develop a PCOS mouse model by treating 21 day old mice for 90 days with a sub-dermal implant of 1) DHT, a potent non-aromatizable androgen; 2) DHEA, which is elevated in PCOS patients; and 3) Letrozole, a compound

which raises endogenous androgens by blocking aromatization to estrogens. All DHT mice were acyclic. Their ovaries were polycystic and exhibited a significantly higher percentage of unhealthy large antral follicles compared to controls. Ovaries had no corpora lutea present, indicating anovulation. DHT mice exhibited a significant increase in body and fat pad weights compared to controls. Blood cholesterol levels were also significantly increased in DHT mice, indicating an increased cardiovascular risk. DHEA mice were cyclic and their ovaries were of normal appearance. DHEA mice lost weight but had unchanged fat pad weights and blood cholesterol levels. Letrozole mice exhibited absent or irregular cycles. Their ovaries were polycystic and contained fewer corpora lutea, however, hemorrhagic cysts which are not a true PCOS phenotype were present. Body and fat pad weights as well as blood cholesterol levels remained normal in letrozole mice. Our results support postnatal treatment of mice with DHT as an effective approach to induce a mouse model to study pathogenic mechanisms in PCOS.

FC026

THE EFFECTIVENESS OF HERBAL MEDICINE PLUS A LIFESTYLE INTERVENTION FOR OLIGO/AMENORRHOEA IN WOMEN WITH PCOS; A RANDOMISED CONTROLLED TRIAL.

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Aim: Many women with PCOS use complementary medicine including herbal medicine for the management of PCOS and to maintain their general wellbeing. However the evidence base of effectiveness and safety is lacking. The aim of this study was to undertake a randomised controlled trial comparing herbal medicine plus lifestyle intervention versus lifestyle alone for women with PCOS for menstrual regularity.

Methods: Overweight women with PCOS were randomly assigned to one of two groups, lifestyle intervention alone or lifestyle plus herbal medicine tablets. Participants were provided with dietary advice and a personalised exercise program. Women assigned to the herbal group were administered tablets containing four herbal medicines and an additional herbal tablet on days 5-14 of the menstrual cycle. The primary endpoint was length of the menstrual cycle at 3 months. Secondary outcomes included anthropometry, hormone concentrations, ovulation, pregnancy and quality of life at 3 months

Results: Recruitment to the trial is complete, 121 women have been randomised, seven women have dropped out of the trial, and there have been two adverse events.

Conclusion: This trial will report on the effect of the intervention on reproductive and anthropometric outcomes for 121 women.

FC027

A CROSS SECTIONAL STUDY OF POLYCYSTIC OVARIAN SYNDROME (PCOS) AMONG ADOLESCENT AND YOUNG GIRLS IN MUMBAI, INDIA

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Adolescent obesity and PCOS individually and together have emerged as important public health issues in India. A cross sectional community based study was undertaken in a sampled census block of Mumbai to assess the prevalence of PCOS among 778 adolescents and young girls aged 15-24 years. Among them, 600 completed all clinical, USG and biochemical investigations. The prevalence of PCOS among them was 22.5% by Rotterdam and 10.7% by AES criteria. Non-obese comprised 71.8% of diagnosed PCOS. Mild PCOS (oligomenorrhea and PCO on USG) was the most common phenotype (52.6%). Hyperinsulinemia ($>15 \mu\text{IU/ml}$) was present among 19.2% of diagnosed PCOS cases. PCOS cases were on the higher scale of the normal range for biochemical parameters. Obese girls with PCOS were more hirsute, hypertensive and had significantly higher mean insulin and two hours post 75 gm glucose levels compared to non-obese PCOS suggesting pertinent danger to develop metabolic syndrome with increasing age as is already known.

To our knowledge, this is the first study diagnosing PCOS and phenotypes among adolescent and young girls in urban community settings in India. This study demonstrates that PCOS is an emerging disorder during adolescence and screening could provide opportunity to promote healthy lifestyles and dietary changes along with exercises for weight

management, coupled with medical management to treat clinical symptoms and hyperinsulinemia and early interventions to prevent future morbidities. A retention rate of 77.2% in the study highlights the need to create awareness among general population about this disorder and its sequelae

FC028

SEX HORMONE BINDING GLOBULIN (SHBG) AS A PREDICTOR OF RESPONSE TO METFORMIN THERAPY IN POLYCYSTIC OVARIAN SYNDROME (PCOS)

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OBJECTIVE: Current consensus advocates individualized approach for metformin use in PCOS. SHBG is emerging as a biochemical predictor of response to metformin therapy. The objective was to assess whether SHBG levels could be used to predict response to Metformin in PCOS by evaluating the change in clinical, biochemical and ultrasonic parameters after six months of metformin therapy and by correlating the pre-treatment SHBG levels with the change in the objective criteria of response.

METHOD : Prospective follow up study on ninety PCOS women fulfilling Rotterdam's criteria. Pre-treatment serum SHBG levels were determined by ELISA. These women were evaluated clinically every month, ultrasonically every three months and biochemically after completion of six months of metformin therapy.

RESULTS: Out of 90 women, 84 completed the study. 61% had biochemical hyperandrogenism and 92% had ultrasonic morphology of PCO. 60% showed positive response to metformin i.e. significant improvement in menstrual cyclicity, anthropometric parameters, biochemical hyperandrogenism, metabolic profile and ultrasonic PCO morphology. The responders had a significantly lower levels of SHBG (mean 70.31nmol/l) as compared to the non-responders (154.84nmol/l)($p<0.01$). For every unit increase in Serum SHBG level, the odds of a patient having a positive outcome to metformin treatment fell significantly by a factor of 1.038 ($p<0.001$). Serum SHBG level of ≤ 130 nmol/L was found to be an excellent predictor to response.

CONCLUSION: SHBG may be used as an accurate, reliable and highly sensitive predictor of response to Metformin therapy in PCOS women. SHBG thus may be included in the routine battery of tests for PCOS.

FC029

DETERMINATION OF OVARIAN FOLLICLE LOCALIZATION IN PATIENTS WITH PRIMARY OVARIAN INSUFFICIENCY (POI)

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Ovarian tissue cryopreservation by vitrification is a promising method for fertility preservation in women undergoing gonadotoxic treatments for malignant disease. This procedure could also be useful for patients suffering from progressive ovarian dysfunction, including POI. For successful vitrification, the step of cryoprotectant equilibration is important. The failure of vitrification can be minimized by preparing thin layers of ovarian tissues. Although a few papers showed that early follicles were detected within 750 μ m from the surface of ovarian cortices, exact data on their localization were limited. Here, we investigated the localization of follicles in the ovaries obtained from patients with POI. We obtained informed consent from patients and approval from local Human Subject Committees. After fixing and embedding in paraffin, ovarian samples were serially sectioned, and stained with hematoxylin and eosin. The depth of each follicles from the surface of ovarian cortices were histologically measured. In POI patients ($n=7$; mean 34.3y), primordial and primary follicles were located in depth of $425\pm 16\mu$ m. In some cases ($n=9$; mean 34.5y), we obtained normal samples from patients received autopsy. The follicles in women at 20s, 30s and 40s were located in depth of $502\pm 17\mu$ m, $463\pm 9\mu$ m and $493\pm 15\mu$ m respectively. However, female infants ($n=3$; mean 1.7y) showed shorter depth: $271\pm 4\mu$ m. In conclusion, early follicles were located within 1mm from the surface of ovarian cortices. It was not different in POI patients who exhibited shrunken ovaries. Because some secondary follicles were located 1-2mm in depth, the ovarian tissue could be dissected in 1-2mm thickness to involve those follicles for cryopreservation.

FC030

HOW DOES FOLLICULAR GROWTH RATE AT DIFFERENT TYPE AND DOSES OF OVARIAN STIMULATION DIFFER? A RETROSPECTIVE STUDY

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Introduction Ovulation induction is one of the effective methods for fertility treatment. It is traditionally used for unifollicular ovulation induction in women with oligoovulation or as part of IUI treatment. It is now more commonly indicated for hyperovulation in IVF treatment. With different stimulation drug regimes, the natural growth rate of developing follicles may be altered. The need of close monitoring is important to correctly predict ovulation. This study aims to identify the growth rate of follicle from different stimulation protocols. The primary outcome of this study is growth rate per day. **Methods** Retrospective analysis was done examining patient's record who underwent IUI in University Malaya Medical Centre. Documented size of leading follicles was plotted against day of stimulation. Statistical analysis done using SPSS. **Results** Total of 223 women was included (154 Malays, 38 Indians, 21 Chinese and 10 others). The mean age was 32.81 (21-46, SD 3.906) with majority within the age group of 31 to 35 (n=100). More than half women had cycle length from 25 to 30 days (n=120). Almost one third of the patients were polycystic ovarian syndrome (PCOS). The fastest growth was patient stimulated with FSH (1.07cm/day), Clomiphene Citrate (0.97cm/day) and natural cycle was the slowest at 0.6cm/day. In our sub-analysis we found that the follicular growth after the 10th day of stimulation is faster when women artificially stimulated with FSH and Clomiphene Citrate compared to women in natural cycle, which showed slower growth. **Conclusion** Artificially stimulated cycles showed different follicular growth patterns compared with spontaneous cycles and faster rate advance stage of stimulation

FC031

REACTIVE OXYGEN SPECIES (ROS) LEVELS IN SEMEN FROM INFERTILE COUPLES

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Introduction

High levels of ROS in semen can damage DNA, proteins, lipids and impair sperm function such as motility, viability, morphology, capacitation and acrosome reaction.

Objective

This is a descriptive study on level of ROS in semen samples from Vietnamese infertile couples.

Methods

Semen samples with sperm count $\geq 2 \times 10^6/\text{ml}$ were recruited. ROS levels were measured by luminol-dependent chemiluminescence assay and values were expressed as $\times 10^4$ counted photon per minute (cpm)/ $\times 20^6$ spermatozoa. Correlations between ROS and different semen parameters were also assessed.

Results

Six hundred infertile couples at a fertility clinic from April 2013 to June 2013 were recruited.

ROS values, presented as median and interquartile values (25th; 75th percentiles), were 0.0586 (0.0154; 0.2236) ($\times 10^4 \text{cpm}/20 \times 10^6$ spermatozoa).

The levels of ROS in neat semen had significantly negative correlations with sperm concentration ($r = -0.147$, $P < 0.001$), motility ($r = -0.189$, $P < 0.001$), and normal morphology ($r = -0.096$, $P < 0.018$).

Conclusion

Negative correlations of semen ROS levels with sperm concentration, motility, and normal morphology were confirmed in large sample Vietnamese infertile couples. ROS levels in semen can be a potential marker to assess male infertility.

FC032

THE ANALYSIS OF APOPTOSIS OF FRESH VERSUS THAWED-VITRIFIED ISOLATED PRE-ANTRAL FOLLICLES

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Background

The cryopreservation of ovarian tissue has several potential advantages over other preservation techniques, including the presence of a large number of follicles in the cortex. The risk of micrometastasis as well as re-perfusion injury short after ovarian transplantation is an important issue on ovarian tissue vitrification. Pre-antral follicle vitrification is considered as an option based on its number on ovarian tissue and also its resistance to cryoprotectant agent during vitrification

Objective

To evaluate the efficacy of vitrication of pre-antral follicle as a method of fertility preservation

Design

Experimental study

Setting

Department of Obstetrics and Gynecology Dr. Cipto Mangunkusumo General Hospital Jakarta

Method:

Pre-antral follicles isolation were performed by enzymatic digestion technique (librase, collagenase, DNA-se) from 6 ovaries of women who underwent oophorectomy due to cervical cancer. One hundred sixty seven pre-antral follicles were divided into 2 groups, one was fresh and the other was thawed-vitrified. Apoptosis was analysed based on morphology and expression of FasLigand, caspase-3, BAX and Bcl-2 on follicles by using immunohistochemistry and RT-PCR

Results

The expression of FasLigand, caspase-3, BAX and Bcl-2 were not significantly different between fresh versus thawed-vitrified pre-antral follicles. However the expression of apoptosis gene in ovarian stroma was higher in thawed-vitrified groups

Conclusion

Vitrification of pre-antral follicles can be considered to be used as method of fertility preservation

Keywords: pre-antral follicles; apoptosis; isolation; enzymatic digestion

FC033

EFFECTS OF THREE DIFFERENT TYPES OF ANTIFREEZE PROTEINS ON MOUSE OVARIAN TISSUE CRYOPRESERVATION

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Objective: Ovarian tissue cryopreservation may result in follicular depletion. Many studies demonstrated that different types of antifreeze proteins (AFPs) have beneficial effects on organ preservation. Therefore, we investigated the cryo-protective effects of various types of AFPs on whole ovarian tissue cryopreservation using mouse models.

Methods: The B6D2F1 mice were randomly assigned to 3 groups: fresh control, sham control and AFPs supplemented groups. The AFPs supplemented groups were sub-divided into 9 groups by types (FtIBP, LeIBP and Type III AFP) and dose (0.1 mg/mL, 1 mg/mL and 10 mg/mL) of AFPs. Ovaries were collected from mice and cryopreserved by two-step vitrification method. Ovarian follicle morphology and apoptosis were assessed by H&E stain, TUNEL assay and immunohistochemistry for γ H2AX, respectively.

Results: High concentration of LeIBP group (10 mg/mL) showed significantly higher intact follicle ratio than control group. In addition, significantly higher intact primordial follicle ratios were observed in all three 10mg/mL AFPs supplemented groups compared to control group. LeIBP supplemented groups (1 and 10 mg/mL) showed significantly lower apoptotic follicle ratio than control groups in TUNEL assay. Moreover, γ H2AX positive follicle ratios were significantly decreased in all three 10 mg/mL AFPs supplemented groups compared to control group.

Conclusion: Our results suggest that three different types of AFPs showed beneficial effects on mouse ovarian tissue cryopreservation. Among these, 10 mg/mL of LeBP supplementation in vitrification and warming media has the most protective effects during mouse whole ovarian tissue cryopreservation.

FC034

GENETIC VARIANTS IN THE ETV5 GENE IN FERTILE AND INFERTILE MEN WITH NON-OBSTRUCTIVE AZOOSPERMIA ASSOCIATED WITH SERTOLI CELL ONLY SYNDROME

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Recent advances in animal model production have facilitated the identification of many novel fertility genes. To date, over 400 genes have been shown to be critical for male and/or female fertility. However, only a small number has taken the step of associating such gene dysfunction with human male infertility.

Within this study we have shown that ETV5 is critical for both human and mouse male fertility and loss of function results in Sertoli cell only (SCO) syndrome. Homozygous male mice carrying a knockout or nonsense (stop codon) mutation in the *Etv5* gene are sterile due to a progressive loss of germ cells, which ultimately led to the SCO phenotype.

For the human study we utilized a high throughput-sequencing platform to screen the entire protein-coding region of the *ETV5* gene in gDNA samples from control fertile and infertile men with SCO and non-obstructive azoospermia (NOA). We found a total of 12 genetic variants of which 6 variants had not been reported previously. Importantly, our data suggest that the homozygous TT allele of the +48845 G>T variant confers a higher risk for male infertility associated with SCO and NOA in the Australian men.

In summary, our study illustrates the utility of the use of mouse models to define the causes of human infertility.

FC035

ALCOHOL CONSUMPTION AND SEMEN QUALITY: A RETROSPECTIVE, CROSS-SECTIONAL STUDY OF MEN PRESENTING FOR FERTILITY ANALYSIS.

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Introduction:

It has been established that lifestyle factors contribute to a large proportion of cases of infertility. Importantly, improvements in fertility have been demonstrated through addressing these factors. The effects of human male alcohol consumption on fertility in prior research is equivocal. This research determined if male alcohol consumption within the NHMRC's recommendations is likely to impact fertility, specifically in men presenting to fertility clinics.

Aims:

To determine whether alcohol consumption has a negative impact on semen quality in males presenting to fertility clinics for semen analysis.

Methods:

Semen quality analysis results and survey responses completed at the time of sample collection at a private fertility clinic (N=645 male patients) were analysed retrospectively using t-test and ANOVA. Group comparisons were made between non-drinkers, alcohol consumers drinking within the NHMRC's recommended intake and those consuming more than the recommendations (>14 standard drinks/week).

Results:

Drinkers and non-drinkers did not differ in sperm concentration, motility or morphology. Semen volume was higher in drinkers vs non-drinkers; 3.5ml vs 3.1ml (p=0.03), as was total sperm count; 182×10^6 vs 141×10^6 (p=0.02). Drinkers consuming within the recommended limit had a higher semen volume compared to both non-drinkers and those consuming more than the recommended intake. There were no statistically significant differences between the three groups in all other semen quality parameters.

Conclusion:

The failure to establish an association between alcohol consumption and semen quality makes it unlikely that male patients presenting to fertility clinics will improve their fertility by consuming less than Australia's current national guidelines.

FC036

SPERM ANTIBODY INCIDENCE AND CYCLE OUTCOMES IN 1086 SAMPLES FROM COUPLES PRESENTING WITH INFERTILITY

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Background:

Antisperm antibodies (ASAs) are estimated to occur in 5-15% of infertile men. Pregnancy rates (PR) of approximately 35% have been reported when ICSI is used in those with considerable ASA binding (Zini *et al.*, 2011). The present study aimed to identify the incidence of ASAs in male partners of couples presenting with infertility, and to determine cycle outcomes of those with significant binding who went on to receive treatment.

Methods:

Data from 1806 semen samples obtained between 1997 and 2012, and the outcomes of any subsequent fertility treatment, were analysed. Significant binding was defined as $\geq 50\%$, with other normal parameters also as defined by WHO (2010).

Results:

Significant IgG and IgA binding was identified in 4.6% and 2.8% of samples, respectively. As IgG binding increased above 50%, progressive motility decreased ($p < 0.05$). As IgA binding increased above 50%, sperm concentration and progressive motility decreased ($p < 0.05$). Of those with significant ASA, 60 patients and their partners underwent 108 oocyte pick-ups, the majority of which (93/108) used ICSI. The PR from 98 fresh embryo transfers was 35%, and from 52 frozen embryo transfers 29%, with 58% of couples thus achieving a pregnancy.

Conclusions:

ASAs may be associated with decreased sperm concentration and progressive motility. In those with significant binding, ICSI may overcome the potentially deleterious effects of ASAs.

References:

World Health Organisation, 2010. WHO laboratory manual for the examination and processing of human semen, 5th edition.

Zini *et al.*, 2011. *Hum. Reprod.* 26: 1288-95.

FC037

OBESITY HAS LIMITED PSYCHOLOGICAL IMPACT IN PATIENTS UNDERGOING FERTILITY TREATMENT

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Abstract

Aim

Patients who are overweight or obese are known to present with clinically significant health concerns impacting negatively on reproductive functioning, psychological wellbeing and overall quality of life. This study investigated the level of distress experienced by infertile women with elevated body mass index (BMI).

Methodology

Infertility patients (N = 409) attending three ART clinics in Perth, Western Australia were assessed on standardized psychological factors. Patients were stratified according to WHO (2000) BMI categories (normal, overweight, obese) and compared on psychological outcome measures of self esteem, depression anxiety, general and fertility related stress, mood, perfectionism and eating attitudes and behaviours.

Results

Results of this study did not confirm that infertile women with elevated BMI were more psychologically vulnerable than infertile women with normal BMI levels in regards to perceived stress, positive or negative affect, levels of perfectionism or depression, anxiety and specific fertility related problems (all $p > 0.05$). Participant self-esteem, body shape concerns and binge eating with loss of control however were found to vary between BMI groups ($p < 0.05$) highlighting vulnerability in a subgroup of obese women characterised by disordered eating patterns.

Conclusions

Whilst infertile women with adiposity are at greater risk for poorer reproductive outcomes, these reproductive risk messages do not automatically translate into additive deleterious effects on psychological wellbeing, specific fertility related stress and emotional concerns. Given the emotional status quo, lifestyle modification programmes aimed at reducing weight loss may require different motivational weight management strategies.

FC038

THE NIPT EXPERIENCE: REVIEW OF OVER 250 CASES FROM A SINGLE PROVIDER

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Background: Non-invasive prenatal testing (NIPT) is now widely available in Australia as part of the screening process for fetal aneuploidy. To date there have been no reports on the Australian experience of NIPT.

Aim: To report a single providers experience of NIPT screening since December 2012

Methods: We review over 250 cases of NIPT that have occurred in a single centre since the introduction of NIPT in December 2012. We provide information regarding failure rates, turn-around times, false positives, and outcome information where available. We review factors such as first trimester serum screen results, maternal BMI, gestational age and the impact they have on the success of NIPT. We present qualitative data regarding reasons for NIPT testing.

Results: In over 250 cases of NIPT performed since December 2012 at a single provider, the failure rate and false positive rate is consistent with the published data. Patients from low risk and high risk have been satisfied with NIPT as a screening method.

Discussion: NIPT is being rapidly incorporated in prenatal screening. Our experience represents a significant cohort within a single practice, and it is important to discuss implementation strategies and effectiveness of service. There are many instances in which NIPT can help guide the provider in determining the recommendation for invasive testing procedures. Our experience supports that NIPT must be looked at in conjunction with ultrasound to determine the need for further testing.

FC039

THE PERCEPTIONS AND MOTIVATIONS OF POTENTIAL ANONYMOUS SPERM DONORS IN QUEENSLAND

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Background

2007 NHMRC guidelines require sperm donors when beginning donations to consent to the release of their identifying information once the donor-conceived child reaches the age of 18. Since this change there has been a decrease in Australian sperm donors. Is this due to misconceptions of future identification?

Aim

To explore the psychosocial attitudes of men towards sperm donation, which contribute to their motivations to enquire with our clinic. We also wish to determine the understanding of future identification by sperm donors compared to the general population and investigate any discrepancy.

Method

Men who have both enquired about sperm donation and who have been accepted as donors were sent a survey through email for completion. The general population were surveyed through social media random population sampling.

Preliminary Results

Initial results indicate a poor response rate from men who made enquiries about sperm donation. The current donor response rate was fair and shows primarily altruistic motivations for donation in addition to other motivating factors, and an accurate understanding of future identification requirements. Initial general population responses demonstrate that 25% of respondents believe donor conceived children have legal rights to meet their donor at the age of 18, and 5% of respondents believe donor contact details will be given out.

Discussion

From the data so far collected we can surmise that our current donor pool has been educated appropriately regarding their current responsibilities, and general population education could result in an increase in donor numbers once current misconceptions have been corrected.

FC040

WHAT DO PATIENTS UNDERSTAND ABOUT THEIR PARTICIPATION IN THE IVF ACUPUNCTURE MULTICENTRE RANDOMISED CONTROLLED TRIAL?

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Background: It is important that study participants understand what is involved with any research study. Researchers and ethics committees attempt to ensure that patient information is pitched at an appropriate level, but few studies have examined whether patients that participate in clinical trials have been given adequate information to make an informed decision. This study aimed to examine study participants' views on the information they received about the study, and whether they were given adequate opportunity to ask questions about the study before agreeing to participate in the IVF acupuncture randomised controlled trial.

Methods: Ethics approval has been obtained from 10 separate Human Ethics Research Committees in Australia and New Zealand for the randomised controlled trial. Women were randomised to acupuncture or sham acupuncture, three interventions treatments were provided, with the last treatment session occurring on the day of embryo transfer. Participants are also asked to complete questionnaires relating to trial primary and secondary outcomes on three occasions.

Trial participants were recruited from nine participating IVF sites during 2013 and were invited to participate in this study. An anonymous self completion questionnaire was designed for this study with questions examining about their reasons for participating in the trials, the adequacy of the information provided on the participant information and consent forms, the time available to ask questions and discuss the trial and participant's understanding of the information presented.

Results: During 2013 183 women were invited to participate in the trial. Outcomes will be presented at the conference.

FC041

SEXUAL FUNCTION AND HEALTH-RELATED QUALITY OF LIFE IN WOMEN WITH CLASSICAL BLADDER EXSTROPHY

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Objective

Classical bladder exstrophy is a congenital malformation affecting the pelvic organs. Medical and surgical advances have improved survival rates. The aim of this study was to investigate sexual function and quality of life in adolescent and adult women with bladder exstrophy.

Study Design

This was a two-part observational study with a cross-sectional (questionnaire) arm and a retrospective case review arm.

Results

Forty-four women were identified from departmental databases, of whom 28 (64%) completed postal questionnaires. Sexual function and health-related quality of life scores were significantly poorer compared to normative data.

Conclusions

Bladder exstrophy has a detrimental psychological impact on women. In future, methodical multi-disciplinary pediatric follow-up research will help to identify predictors for better and worse adolescent and adult outcomes. Development and evaluation of cost-effective psychological interventions to target specific problems is also warranted.

FC042

MATERNAL ANXIETY, ART OUTCOME AND EARLY CHILDHOOD TEMPERAMENT: A THREE-YEAR FOLLOW-UP STUDY FROM CHINA

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Objective: Investigate whether maternal anxiety during IVF/ICSI treatment would be related to ART outcomes and difficult early childhood temperament.

Design: The longitudinal cohort design followed 283 nulliparous women receiving IVF/ICSI over a 3-year period, from trans-vaginal oocyte retrieval (TVOR), embryo transfer (ET) in IVF/ICSI, to the third trimester of pregnancy and at 1-2 years after birth.

Participants/Materials and Methods: 283 nulliparous women who received IVF or ICSI were recruited in our department. Participants completed state, trait and preoperative anxiety measures in the morning of one day before TVOR (time 1) and in the morning of the day of ET (time 2). For pregnant women, pregnancy-specific anxiety and depression were measured during the third trimester of pregnancy (time 3). For the child born after IVF/ICSI, toddler temperament was measured at 1-2 years after birth (time 4). At time 1, relevant maternal socio-demographic, biomedical variables were obtained. At time 3, pregnancy information was recorded. At time 4, childbirth and post-natal information were obtained.

Results: After strictly matching all socio-demographic and biomedical variables, women conceiving through ART reported lower state and preoperative anxiety at time1 and 2 than their unpregnant counterparts (All P s<0.05). Hierarchical regression analyses including all anxiety and depression variables, social-demographic variables, pregnancy variables and childbirth variables indicated that maternal trait anxiety predicted more difficult early childhood temperament (P <0.01).

Conclusion: Less state and preoperative anxiety during IVF/ICSI could help improve ART outcomes, and trait anxiety could predict more difficult early childhood temperament.

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FC043

ANEUPLOIDY PATTERNS IN BLASTOCYSTS.

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Aim:

The vast majority of data regarding the distribution of specific aneuploidies in preimplantation embryos is based on cleavage stage samples. Consequently, our aim was to characterize the breakdown of aneuploidies in blastocyst biopsy samples.

Method:

Retrospective analysis of 342 blastocysts undergoing preimplantation genetic screening at Monash IVF.

Results:

A total of 56.1% of blastocysts contained only a single chromosomal error compared to the 19.7% of cleavage stage embryos reported by Rabinowitz et al (2012). Interestingly 84.9% of blastocysts contained ≤ 3 errors, more closely resembling the patterns seen in prenatal testing than it does the prevalence of complex abnormalities seen at the cleavage stage. This suggests that a significant portion of the shift seen between preimplantation and prenatal samples is actually occurring prior to blastocyst formation.

In our blastocyst population, the proportion of individual monosomies was 18.75% compared with 81.25% trisomy. This is markedly different from widely published cleavage stage data where there appears to be a relatively even prevalence of monosomies and trisomies. This difference is similarly pronounced in our blastocysts displaying only a single error, with 31.2% of these being a monosomy and 68.8% a trisomy.

When analyzing individual chromosomes blastocysts display a very early shift toward the patterns seen in prenatal samples, with chromosomes 13, 18, 21 and 22 noticeably prevalent compared with the more even spread seen at day 3.

Conclusion:

Blastocysts display a very different distribution of aneuploidies to cleavage stage embryos, with a distinct shift toward fewer complex aneuploidies and fewer monosomies.

FC044

PREDICTING SEGREGATION MODES FOR AUTOSOMAL RECIPROCAL TRANSLOCATIONS AT PREIMPLANTATION GENETIC DIAGNOSIS

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Introduction:

Reciprocal translocation carriers often experience reproductive difficulties due to the production of chromosomally unbalanced gametes. Preimplantation Genetic Diagnosis (PGD) provides the opportunity to distinguish between "normal/balanced" embryos (potential for successful pregnancy) and unbalanced embryos (potential for implantation failure/miscarriage/abnormalities at birth). During PGD counselling, couples want to know the chance of obtaining a normal/balanced embryo for transfer. Accurate reproductive risk assessment is vitally important in helping couples decide if PGD is their best option. Currently, determination of reproductive risks is based on pre-/postnatal data. There is no resource that can be applied to translocation carriers that is specific for PGD.

Aim:

To analyse autosomal reciprocal translocation segregation modes in IVF-PGD embryos and use the observed segregation modes to generate a predictive model that applies specifically to PGD.

Method:

A retrospective analysis was performed on 1641 D3 embryos (139 translocations) from 144 couples who had undergone PGD at Monash IVF between 2002 and 2013.

Results:

Segregation analysis showed that 22.4% of embryos were "normal" or "balanced" (alternate segregation) and 77.6% were unbalanced for the translocation chromosomes. Of the unbalanced embryos, 18.8% showed adjacent-1 segregation, 9.5% adjacent-2 segregation, 17.8% 3:1 segregation, 1.9% 4:0 segregation and 29.6% unascertained/mosaic mode of segregation.

Conclusion:

This data represents the largest analysis yet of meiotic segregation for reciprocal translocation carriers in D3 embryos and provides the information required to generate a predictive model for PGD. This model would ensure that PGD patients with reciprocal translocations have realistic expectations regarding their chance of obtaining a normal/balanced embryo for transfer.

FC045

DOES FEMALE AGE, AMH, CYCLE TYPE, AND FSH DOSING IMPACT ON THE NUMBER OF DAY 5/6 BLASTOCYSTS SUITABLE FOR BIOPSY AND SUBSEQUENT ANEUPLOIDY RATES?

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Aim:

To determine whether female age, AMH, cycle type, and rFSH dosing impacts on the number of day 5/6 blastocysts suitable for biopsy and the proportion of euploid embryos.

Methods:

All patients who had undergone a stimulation cycle resulting in at least one day5/6 blastocyst biopsy for CGH at Genea, Sydney Australia between August 2009 and September 2013 were considered eligible. This resulted in 611 stimulation cycles for analysis. Explanatory variables included female age, AMH, cycle type, starting rFSH dose, days of rFSH, total rFSH dose, and number of oocytes retrieved. Outcome data included number of blastocysts suitable for biopsy and the proportion of euploid embryos.

Results:

Female age, starting rFSH dose, and total rFSH dose was inversely correlated with the number of blastocysts suitable for biopsy and the proportion of euploid embryos. AMH and the number of oocytes retrieved correlated with the number of blastocysts suitable for biopsy but not with the proportion of euploid embryos. Cycle type and the number of days of rFSH did not correlate with either outcome variable. Female age and total rFSH dose were included in the final logistic regression model for the number of blastocysts suitable for biopsy and the proportion of euploid embryos after non-confounders were removed.

Conclusion:

Female age and total rFSH dose were the only explanatory variables that independently correlated with the number of blastocysts suitable for biopsy and the proportion of euploid embryos.

FC046

NEXT GENERATION SEQUENCING-BASED ANEUPLOIDY DETECTION

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Background: Next generation sequencing (NGS) technologies have evolved from the methods developed in the Human Genome Project, allowing rapid and cost-effective analysis of genomic DNA. The possibility of using NGS technologies to analyse the genetic profile of a single cell has recently arisen, creating the potential for concurrent detection of numerical, structural and molecular genetic abnormalities. Methods: Whole genome amplification products from embryos with known diagnoses on array CGH were sequenced on the Illumina MiSeq. Libraries were constructed according to standard protocols and samples barcoded to allow multiple analyses per chip. High quality reads were aligned to the human genome (hg19) and depth of coverage analysed in a low pass whole genome aneuploidy analysis workflow. Computer modelling allowed determination of the minimum information required to achieve reliable detection of aneuploidy. Results: Using the independent reads which aligned to the human genome from each sample, reliable aneuploidy diagnoses can be reached with a depth of coverage analysis. Results from the NGS workflow were compared with array CGH results performed on the same WGA products. Computer modelling allowed determination of the maximum number of embryos to be analysed per experiment and the simplest available workflow for clinical application. Quantification of mitochondrial DNA was also possible using a next generation sequencing approach, suggesting possible mechanistic explanations for the incidence of aneuploidy in human embryos. Conclusion: Next generation sequencing technologies have the potential to significantly impact on the practice of reproductive medicine, allowing the rapid detection of aneuploidy on multiple samples concurrently and significantly improving the cost and efficiency of current preimplantation genetic screening programs. Future directions of this research will be focused on an investigation of the underlying causes of aneuploidy in human embryos.

FC047

A HIGH-THROUGHPUT AND ROBUST BAC-ON BEADS BASED COMPREHENSIVE CHROMOSOME SCREENING (KARYOLITE) TO IMPROVE THE SUCCESS OF PREIMPLANTATION GENETIC SCREENING

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Interrogate the DNA at blastomere or blastocyst stages to ensure euploidy represent a useful strategy to improve the success of IVF. Several aCGH and SNP array platforms have been showing comprehensive chromosome screening was highly predictive of clinical outcome. However the current methods were expensive and not able to achieve a high throughput to meet the demands of a busy reproductive unit. Here we validated a high throughput BAC-on Beads based screening (KaryoLite) method with resolution down to arm-specific chromosome aberrations for preimplantation genetic screening (PGS). The aim of this study is to reduce the cost and increase the throughput of PGS, to make this technique suitable for PGS of Day-3 and Day 5 embryos. Twelve single amniocyte from six aneuploid prenatal samples and 96 blastomeres DNA from Day-3 or 44 from Day 5 embryos preserved after PGS by aCGH were analyzed by KaryoLite. In 100% of the amniocytes analysed, the characteristic aneuploidies of each samples were correctly identified by KaryoLite. The euploidy results of the blastomeres and blastocysts were consistent between KaryoLite and aCGH results. Twelve arm specific chromosomal changes events were also appropriately identified by KaryoLite. KaryoLite was able to perform up to 90 DNA samples per run. Our data is the first proof of principle study validation a high throughput, low cost approach that can be applied to Day-3 and Day 5 aneuploidy analysis and perhaps helping to improve implantation rate after PGS.

FC048

SELECTING A FERTILITY SPECIALIST: WHOSE OPINION MATTERS?

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Background: Patients seeking a consultation with a Fertility Specialist in Australia must obtain a referral from their General Practitioner (GP) in order to be eligible for Medicare rebates. There have been no reports to date regarding how patients select a Fertility Specialist, and the role the GP plays in this process.

Aim: To investigate how patients choose a Fertility Specialist.

Methods: Patient population consisted of 2385 patients presenting to a single fertility provider in 2012 and 2013. Patients were contacted via email and asked to complete a confidential online questionnaire.

Results: We received 540 responses representing a response rate of 23%. In 2012, 52% of patients consulted their GP for information regarding fertility and 42% relied on their doctors recommendation of a Fertility Specialist. In 2013, 45% consulted their GP for information and 36% relied on GP recommendation. Patients seek information from a variety of sources that include online research as well as discussions with friends and family members.

Discussion: The majority of patients do not rely on their family doctor to choose a Fertility Specialist. Reliance on a family doctor for specialist recommendation may be declining despite patients still having to visit their GP for a referral.

FC049

CHANGES IN THE SURFACE AREA OF HUMAN OOCYTES FOLLOWING ICSI: OBSERVATIONS MADE USING THE EMBRYOSCOPE™ TIME-LAPSE SYSTEM.

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Oocytes after ovulation are not static structures but undergo a series of dynamic changes which affect their size, including the self-regulation of cytoplasmic volume and the extrusion of polar bodies. Previous studies have indicated that human oocytes are no exception, but the availability of the new time-lapse video system (Embryoscope™) has enabled a more detailed investigation. The present study aimed to (a) determine the time course of second polar body extrusion in fertilized human oocytes, (b) compare surface area of both the first and second polar bodies, (c) compare the surface area over time of fertilized and unfertilized oocytes, and (d) determine the impact of second polar body extrusion upon oocyte size. In total

192 oocytes from 35 ICSI treatment cycles were included for assessment using the Embryoscope™ time-lapse video system, with surface area measured retrospectively at 1 hour intervals for 9 hours post insemination. The second polar body was extruded at 2.9 ± 0.1 hours (range 0.97-7.5 hours) relative to insemination, and had a smaller surface area than the first polar body ($p < 0.005$). Overall, oocytes reduced in size following ICSI ($p < 0.05$) with shrinkage ceasing after 2 hours in the unfertilized oocytes and at second polar body extrusion in the fertilized oocytes. In conclusion, all human oocytes reduce in size following ICSI, but the extrusion of the second polar body in fertilized oocytes is a pivotal event in terminating shrinkage of the vitellus.

FC050

MEIOTIC SPINDLE NORMALITY PREDICTS LIVE BIRTH IN PATIENTS WITH RECURRENT IVF FAILURE

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Objective: To investigate the impact of meiotic spindle normality on live birth rates in women with recurrent IVF failure.

Design: A retrospective comparative study.

Setting: IVF Australia, Sydney, Australia.

Patients: 505 ICSI/ET cycles of patients who have fulfilled the criteria for Recurrent IVF Failure (RIF) - > 3 previous fresh or frozen embryo transfers with no ongoing pregnancy.

Interventions: Polarised Light Microscopy (PLM) was used at the time of ICSI.

Main Outcome Measure(s): Clinical pregnancy rates (CPR) and live birth rates (LB) were compared depending on the morphology of the meiotic spindle of the oocyte(s) from which the embryo(s) transferred, were derived.

Results: Women receiving embryos where at least one was derived from a normally spindled oocyte had significantly increased CPR and LBR when compared to those who had only embryos derived from abnormally spindled oocytes (CPR: 31% vs. 7%) (OR=6.45 with 95% CI (3.65 – 11.40) and (LBR: 24% vs. 4%) (OR=7.24 with 95% CI (3.62 – 14.49). Comparison between the abnormal spindle subgroups showed significantly higher CPR and LBR from the group of patients receiving embryos where at least one was derived from an oocyte with no visible spindle compared to the group receiving embryos from dysmorphic spindles only (CPR: 9% vs. 6%) (OR=0.58 with 95% CI (0.22 to 1.57) and (LBR: 8% vs. 1%) (OR= 0.16 with 95% (0.03 to 0.77).

Conclusions: Normally spindled oocytes as determined by polarised light microscopy, are associated with significantly higher clinical pregnancy rates in patients with Recurrent IVF Failure.

FC051

EXPANDED BLASTOCYST MORPHOLOGY ASSESSMENT AND THE DECISION TO TRANSFER: RESULTS FROM AN EXTERNAL QUALITY ASSURANCE PROGRAMME.

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The morphological examination of an embryo is a basic assessment made by embryologists to gauge its quality and ultimately whether it is suitable for use (transfer or cryopreservation). This process is applied to all embryos from the early cleavage stage to the expanded blastocyst, and requires that all embryologists are consistent when assessing an embryo. External quality assurance (EQA) programmes are important tools to monitor variability in measurements or observations, and EQASRM has operated a blastocyst assessment programme since 2007 whereby videos of expanded blastocysts are sent to participating laboratories and questions asked about the condition of the inner cell mass (ICM) and whether the laboratory would transfer the embryo or not. From 2007 to 2012, between 22 and 26 laboratories enrolled each year and all received 4 (in 2007) or 8 (in 2008-2012) videos per annum over quarterly distributions. Of the 1088 embryo videos distributed to the laboratories over this period, 990 (91.0%) responses were received. The condition of the inner cell mass seemed an important criterion when deciding to transfer, with laboratories overall agreeing to transfer the embryo on only 11/92 (12.0%) of occasions when the ICM was absent, but the decision to transfer rose progressively when the ICM was said to be poor (193/283; 65.9%) or average/good (587/615; 95.5%). In summary, the EQA programme has proved valuable in monitoring the assessment of expanded human blastocysts, quantifying the decision-making process and hopefully providing feedback to laboratories on their performance.

FC052

EFFECT OF GONADOTROPINS ON EPIGENETICS AND GENOMIC IMPRINTING IN OOCYTES: ART SAFETY IN VITRO MODEL

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Gonadotropins (Gns) have been played key roles during fertility treatment and widely used in human assisted reproduction and animal science over the past four decades. However, the specific effect of Gonadotropins on epigenetic alteration (DNA Methylation) and genomic imprinting changes (*H19* and *IGF2*) in oocytes during physiological and high dose stimulations is poorly understood. In order to further our understanding of the role that Gns during these events, we chose bovine oocytes as animal model to study the effect of Gns in a dose-response manner. Bovine cumulus-oocyte complexes (COCs) were purchase from commercial company and matured *in vitro* in media supplemented with varying doses of Bravelle or Gonal-F (B/F), B/F + Menopur (B/F+M) and B/F + Repronex (B/F+R) for 24 hours. Then after, we evaluated oocytes' epigenetic and imprint alterations. The results showed that the maturation rate of bovine oocyte is highest in 75 and 750 mIU / ml Gns dose groups. H19 methylation level on CpG island is positively correlated to Gns doses, while IGF2R gene expression were similarly correlated with Gns dose, but significantly decreased in overdose group.

Conclusion(s): global DNA methylation status were not altered in a dose-response manner, however H19 methylation and IGF2R gene expression level is positively correlated to Gns doses, but IGF2 significantly declined in overdose group. These results may provide guide for clinical stimulation protocols, helping reduce the risks associated with ART treatment.

FC053

THE EFFECTIVE OF PLATELET LAYSET (PL) ON PROGESTERONE (P4) SECRETION FROM MOUSE PRE-ANTRAL FOLLICLES AND CONSEQUENCE OVULATION

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Introduction

Progesterone (P4) is one of intra follicular steroid mediator which is required for ovulation and fertilization. Platelet Layset with high percentage of growth factors and micro-element can influence P4 secretion and consequence oocytes ovulation. This study has been made to evaluate whether PL has any effect on P4 level in culture medium.

Material and method

In this project we obtained pre-antral follicle from 12 days female NMRI mouse and cultured them in culture medium with different supplement for 13 days. Our culture medium was αMEM, first and second mediums were enriched by 5% and 10% FBS as control groups and our experimental groups were enriched by 5% and 10 %PL . We refreshed our medium every other day and Conditioned medium were frozen for progesterone detection at day 9 and 13.

Result

There was significant increase in progesterone level in medium with PL ($P \leq 0.05$). The level of P4 in medium with 5 and 10% PL was 22.43 ± 1.401 and 27.03 ± 1.001 Pgr/ml at day 9 and this level reached to 35.76 ± 3.1 and 50.06 ± 2.33 Pgr/ml at day 13 before conduction of ovulation compare to our best control group which this amount was 1.7 ± 0.65 at day 9 and reached to 19.8 ± 2.02 Pgr/mlm at day 13.

Discussion

PL with high percentage of growth factors and micro element seem to be effective on ovulation and can be used in ovulation medium.

Keyword

PL (platelet Layset), pre-antral follicle, progesterone P4, Fetal Bovine Serum

FC054

CORRELATION OF OXIDATIVE STRESS BIOMARKERS IN HUMAN FOLLICULAR FLUID WITH OUTCOME IN ASSISTED REPRODUCTION CYCLES

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To investigate whether oxidative stress markers in human follicular fluid (FF) surrounding oocytes are related to embryo development, we examined the relationship between oxidative stress markers and the in vitro fertilization (IVF) outcomes. Seventy-eight infertile women were included in the study. FF was obtained from mature follicles at the time of oocyte retrieval. The total antioxidant capacity (TAC), glutathione, vitamin C, and 8-2'-deoxyguanosine (8-OHdG) concentrations were measured. There was a significant negative correlation between 8-OHdG and vitamin C levels ($r = -0.295$, $p < 0.01$). Total GSH and vitamin C levels were found to be lower in the case of low fertility. In addition, 8-OHdG levels were found to be higher in the case of low fertility and low development competence. Total GSH activity was found to be lower in endometriosis patients, as opposed to male factors. The results of the present study suggest that measuring individual activities of antioxidants in FF will become an important marker for fertilizing in ART. We also found that FF may be an optimal source of non-invasive biochemical markers for the diagnosis of the fertilization failure in IVF. The study also demonstrated that endometriosis patients had lower antioxidant levels compared with patients with male factor infertility. ROS appear to have negative roles in oxidative stress in relation to female reproduction.

FC055

ADMINISTRATION OF SAI-REI-TOU AND CABERGOLINE IS EFFECTIVE TO PREVENT SEVERE OHSS

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Objective

The prophylactic cabergoline administration has been shown to effectively avoid severe OHSS following in controlled ovarian stimulation (COS). Besides this, a herbal medicine Sai-rei-tou (TJ-114) is known to induce the fluid shift from the third space to intravascular space due to its steroid-like effect. We conducted this study to investigate whether administration of TJ-114 and cabergoline reduces the incidence of OHSS in agonist long protocol.

Design

Retrospective clinical study.

Materials and methods

One hundred fifty-six patients who had COS with agonist long protocol showed their serum E2 exceeding 3500pg/ml. We divided them in following three groups. Group A: no medication, B: cabergoline 0.5mg/day, C: TJ-114 7.5g/day and cabergoline 0.5mg/day. Seven days after OPU, we performed ultrasonography to measure ascites volume and ovarian size and ran a blood test to evaluate hemoconcentration. We employed the classification of the Japan Society of Obstetrics and Gynecology to evaluate OHSS. Statistical analysis of this data was performed using t-test and $p < 0.05$ was considered significant.

Results

The mean age of the patients and maximum serum E2 level were comparable in three groups. The volume of both ovaries after OPU was significantly smaller in Group C than A ($p < 0.05$). The average grade of OHSS was significantly lower in Group C than A ($p < 0.05$). However there was no difference in hematological value and ascites volume in three groups.

Conclusion

Our study suggested that the administration of TJ-114 and cabergoline was effective to prevent severe OHSS in agonist long protocol.

FC056

ENDOMETRIAL RECEPTIVITY OF ENDOMETRIOSIS PATIENTS MOLECULAR BIOLOGY STUDY ON POLYMORPHISM OF MUC-1 AND COX-2

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ENDOMETRIAL RECEPTIVITY OF ENDOMETRIOSIS PATIENTS

Molecular Biology Study on Polymorphism of MUC-1 and COX-2

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Abstract

Background: The implantation defects is considered as a major cause of infertility in endometriosis. Comprehensive understanding of the molecular mechanisms regulating embryo implantation hopefully increase the success of implantation in endometriosis therapies.

Objective: To investigate endometrium receptivity disorders on endometriosis by evaluating the difference of MUC-1 and COX-2 genes polymorphism frequency relating to the endometrium receptivity defects.

Methods: Case-control study. Blood samples from 35 endometriosis patient and 32 normal ones were taken during secretory phase of cycle. MUC-1 polymorphism was examined by Amplification Refractory Mutation System (ARMS) and Gene COX-2 Polymorphism by Polymerase Chain Reaction - Restriction Fragment Length Polymorphism (PCR-RFLP). Frequency distribution of gene polymorphism between two groups were compared by bivariate analysis.

Result: There are 7 variants of genotype combination between MUC-1 and COX-2: AAGC; AAGG; GACC; GAGC; GAGG; GGGC; GGGG. Genotype combination test of AAGC was significant with OR=6.43; (95% CI: 1.09 – 37.62) and p = 0,01. Genotypes combination of MUC-1 and COX-2 (AAGC) cause receptivity defect in endometriosis.

Conclusions: The combination of MUC-1 and COX-2 genotypes (AAGC) cause defect of endometrial receptivity in endometriosis. There are differentiations in frequency distribution of genotype combination which AAGC is higher on endometriosis than control.

Key words: MUC-1, COX-2, polymorphism, endometrium, receptivity, endometriosis.

FC057

CORRELATION BETWEEN LEVELS OF HIGH-DENSITY LIPOPROTEIN LEVEL IN SERUM AND FOLLICULAR FLUID WITH OOCYTE QUALITY IN PCOS PATIENTS UNDERGOING IVF

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Background: Approximately 60-70% of embryos in IVF patients fail to achieve implantation. Low oocyte quality might attribute to this failure. HDL is the only lipoprotein found in the follicular fluid during folliculogenesis. High-density lipoprotein (HDL) has been suggested as parameter correlated with embryo quality and folliculogenesis, but data on this matter is limited.

Objective: To assess the correlation between levels of high-density lipoprotein (HDL) in the serum and follicular fluid with oocyte quality in PCOS patients undergoing IVF.

Design: This is a cross-sectional study conducted in the Department of Obstetrics and Gynecology, Dr Cipto Mangunkusumo General Hospital and Sammarie Basuki Rachmat Hospital, Jakarta, from June 2011 to December 2011.

Result: We obtained 48 subjects, consisting of 29 PCOS patients and 19 controls. We found the medial level of HDL, ApoAI, and PON1 was significantly higher in the serum compared to the follicular fluid. We found significant correlation between levels of HDL and ApoAI in the serum and follicular fluid with oocyte quality.

Conclusion: HDL level in the serum and follicular fluid correlates with oocyte quality in PCOS patients undergoing IVF.

Keywords: PCOS, high-density lipoprotein, follicular fluid

FC058

INFLUENCE OF THE STATUS OF FALLOPIAN TUBE ON OVARIAN RESERVE AND PREGNANCY RATE OF IVF-ET

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(Objectives)

Pelvic inflammatory disease including salpingo-oophoritis is known to cause not only tubal lesions, but impair ovarian reserve. The present study was conducted to investigate the relationship between tubal status and ovarian reserve, and also clinical outcomes of IVF-ET.

(Materials and Methods)

One thousand two hundred ninety five patients with retained bilateral tubal patency (group A) and 642 patients with unilateral or bilateral occlusion (group B) were applied for analysis. Average antral follicle count (AFC) and anti-mullerian hormone (AMH) levels (ng/ml) in different age groups were compared between the two groups. Pregnancy rate (PR) under 39 years old of eSET of cleavage-stage embryo and blastocyst in fresh (ET) and frozen-thawed (FET) transfers were also compared.

(Results)

AFCs of group A and B were as follows. Age<30: 6.6±2.7 and 6.3±2.6, Age 30-34: 6.1±2.4 and 5.3±2.2 ($p<0.01$), Age 35-39: 4.8±2.6 and 4.2±1.9 ($p<0.05$), Age>39: 3.0±1.5 and 2.7±1.5, respectively. AMH values of group A and B were as follows. Age<30: 6.43±4.18 and 4.66±2.38 ($p<0.05$), Age 30-34: 5.43±6.08 and 4.81±3.74, Age 35-39: 3.55±2.85 and 2.72±1.94 ($p<0.01$), Age >39: 1.63±1.29 and 1.55±1.83, respectively. PRs of cleavage-stage embryo in group A and B were as follows. ET: 34.1 and 19.7 ($p<0.05$). FET: 27.6 and 21.1. Total: 31.7 and 21.4 ($p<0.05$). PRs of blastocyst in group A and B were as follows. ET: 44.9 and 44.8. FET: 52.8 and 47.3. Total: 50.2 and 46.4.

(Conclusions)

The present study suggests tubal status relates to the ovarian reserve and even implantation of cleavage-stage embryo transfer.

FC059

AN INDIRECT MECHANISM TO EXPLAIN THE BENEFICIAL EFFECT OF GROWTH HORMONE ON HUMAN OOCYTE QUALITY IN POOR RESPONDER PATIENTS.

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Aim: To determine the mechanism, which could explain the clinical benefit from growth hormone (GH) used in women with poor ovarian response (POR)¹.

Methods: Granulosa cells (GCs) were collected through trans-vaginal oocyte recovery from 137 gonadotrophin-stimulated patients of varying ages with or without GH supplementation. GCs were purified, immuno-labelled with fluorescent antibodies to receptors (R) of GH, FSH, LH and BMPR1B then analyzed by flow cytometry for quantification.

Results: In untreated patients, the levels of GHR on GCs were significantly ($p<0.0004$) reduced in cells from older (≥ 40 yrs) patients than from younger (< 40 yrs). In addition, GHR was significantly ($p<0.0001$) elevated in patients treated with exogenous GH, suggesting an auto-upregulation mechanism. More interestingly, the expression of FSHR and LHR was significantly ($p<0.0001$) increased on GCs from GH-treated patients compared with age-matched (≥ 40 yrs) untreated patients. Also, the level of BMPR1B was significantly ($p<0.0001$) increased in cells from GH-treated patients compared with untreated.

Conclusions: This study demonstrates for the first time an indirect mode of action of GH, restoring the responsiveness of GCs of maturing follicles of POR women to gonadotrophin stimulation hence indirectly improving developmental competence of the oocyte. Together with recent observation², it provides a further scientific mechanism for the clinical benefit from GH in such patients.

¹Yovich & Stanger 2010 Reprod BioMed Online 21:37-49. ²Weall et al 2014 ASPIRE.

FC060

A DIRECT MECHANISM FOR THE PUTATIVE ROLE OF GROWTH HORMONE IMPROVING OOCYTE QUALITY FROM POOR RESPONDER PATIENTS.

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Aim: To determine the mechanism by which growth hormone (GH) could improve oocyte developmental competence, in support of a recent clinical finding.

Methods: Oocytes were recovered from women of varying ages who were classified as either normal or poor ovarian responders (NOR or POR). Some of the POR women were also treated with GH. The oocytes were analysed for GH receptor (GHR) expression by immune-labeling, confocal microscopy and computer-based quantification. Also, mitochondrial structural and functional integrity was assessed following MitoTracker and cytochrome oxidase labeling.

Results: GHR was observed on the surface of human oocytes. Oocytes recovered from younger (<35yrs) NOR women exhibited a significantly ($p<0.004$) higher level of GHR compared to those recovered from older (≥ 35 yrs) NOR. Similarly, oocytes recovered from younger NOR women had a significantly ($p<0.002$) more functional mitochondria than older NOR. More interestingly, oocytes recovered from the older POR women, treated with GH, also had a significantly ($p<0.005$) higher level of functional mitochondria, compared to their age-matched untreated women. Although increased functional mitochondria in treated POR patients was lower than in untreated young patients, this was not significant. The total mitochondrial membrane was comparable between the patients irrespective of age or treatment.

Conclusions: This study demonstrates for the first time the presence of GHR on human oocytes, enabling a direct mode of action in improving the oocyte quality in POR patients, probably through the promotion of mitochondrial functional integrity. Together with recent observation, these results provide further scientific support for GH use in such patients.

FC061

EMBRYO MORPHOKINETIC DEVELOPMENT IS THE SAME FOR PATIENTS WITH AND WITHOUT POLYCYSTIC OVARIES AND IS NOT IMPAIRED BY IVM TREATMENT.

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AIM: To compare embryo morphokinetic parameters between ICSI and In-Vitro Maturation (IVM) treatments and between patients with and without Polycystic Ovaries (PCO).

Method: A prospective case control study comparing embryos from three groups of patients; patients with and without PCO undergoing ICSI treatment and patients with PCO undergoing IVM treatment, defined as Non-PCO ICSI, PCO-ICSI and IVM respectively. Participants were recruited between November 2012 and October 2013 and all embryos were cultured in an Embryoscope incubator. Morphokinetic annotations were performed retrospectively. Only embryos with annotations completed to time tB (blastocyst) were included in the analysis.

Results: There were 49 embryos from 14 Non-PCO ICSI patients, 68 embryos from 12 PCO-ICSI patients and 52 embryos from 15 IVM patients. There were no significant differences between the three groups in the mean times for pronuclei fading, cell cleavage timings from 2 through to 9 cells, time to compaction, start of blastulation or to form a complete blastocyst. Furthermore there was no difference in time of the second or third cell cycles (t3-t2, t5-t3) or the time of synchrony in the second cell cycle (t4-t3).

Conclusions: This study indicates that embryo morphokinetic development does not differ between patients undergoing ICSI treatment with or without PCO, and that for PCO patients there is no difference in embryo morphokinetic development between embryos derived from IVM and traditional ICSI treatment. This research is further evidence that IVM can be used as a successful treatment option whilst lowering the risks associated with FSH stimulation in PCO patients.

FC062

THE ROLE OF SERUM ANTI-MULLERIAN HORMONE (AMH) IN PREDICTING POLYCYSTIC OVARIAN SYNDROME (PCOS)

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Introduction

Serum AMH has been widely used for predicting ovarian response. However, there is limited data about AMH in Vietnamese PCOS patients.

Objectives

To investigate the utility of AMH as a predictor of PCOS and to identify the cut-off for PCOS in subfertile Vietnamese women.

Methods

This was a cross-sectional study conducted on subfertile women from January to June 2013. Patients were < 35 years old and had AMH measurement. PCOS was diagnosed following the Rotterdam criteria. AMH was analyzed to investigate its correlation with symptoms of PCOS. The AMH cut-off value was identified with the optimal sensitivity and specificity.

Results

In 1001 patients enrolled in the study, the prevalence of PCOS was 16%. Mean AMH level of PCOS patients was significantly higher than that of non-PCOS patients (9.86 ± 4.49 vs 4.94 ± 3.13 , $p < 0.0001$). ROC curve of AMH in predicting PCOS had an AUC of 0.83. The AMH cut-off value of 6.6 ng/ml predicted PCOS with a sensitivity of 73% and specificity of 74%. AMH level correlated well with menstrual disorders and PCO images but not with hyperandrogenism.

Conclusions

Serum AMH is a predictive marker of PCOS in Vietnamese patients. AMH could increase the reliability of the Rotterdam criteria for diagnosing PCOS.

FC063

HAVE SPERM CHARACTERISTICS CHANGED OVER TIME IN A POPULATION OF QUEENSLAND MEN? AN ANALYSIS OF 1799 SAMPLES FROM 2003 TO 2012.

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Background:

There are conflicting reports in the literature as to whether sperm characteristics and population fecundity have declined over time (te Velde *et al.*, 2010). The present study aimed to determine if a change in semen parameters occurred over time and with respect to patient age in a population of Queensland men.

Methods:

Using WHO standards (2010), 1799 semen samples obtained between 2003 and 2012 were analysed. Statistical analysis was performed on these results to identify potential temporal relationships between routine semen parameters.

Results:

Excluding all other factors, the average sample volume and progressive motility decreased as patient age increased ($p < 0.05$). The percentage of IgG and IgA binding increased with patient age ($p < 0.05$).

When controlled for patient age, the average sample volume, progressive motility and percent normal morphology was found to decrease over time ($p < 0.05$). Conversely, the average sample count increased over time when controlled for age, as did sperm concentration ($p < 0.05$). No effect was identified of time on percentage IgG or IgA binding ($p > 0.05$).

Conclusions:

The effect of age on sperm characteristics amongst the current study population is consistent with published literature. While other semen characteristics decreased over time, the total count and concentration increased. Future studies to examine the effects of this trend on cycle outcomes would be helpful.

References:

te Velde *et al.*, 2010. *Hum Reprod.* 25: 1348-53.

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FC064

DYNAMIC EXPRESSION OF SEPT12 AFFECTS THE INTEGRATION OF NUCLEAR ENVELOPE DURING HUMAN SPERMIOGENESIS

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The septin gene belongs to a highly conserved family of polymerizing GTP-binding cytoskeletal proteins. SEPTs perform cytoskeletal remodeling, cell polarity, mitosis, and vesicle trafficking by interacting with various cytoskeletons. Our previous studies have indicated that *SEPTIN12*^{+/-/+/-} chimeras with a *SEPTIN12* mutant allele were infertile. Spermatozoa from the vas deferens of chimeric mice indicated an abnormal sperm-head and -tail morphology, decreased sperm count, and immotile sperm. Mutations and genetic variants of *SEPTIN12* in infertility cases also caused oligozoospermia and teratozoospermia. However, the biological roles of SEPT12 during spermiogenesis are still unclear. First, we identified the SEPT12-interacted proteins via yeast-two-hybrids. One of twenty SEPT12-interacted proteins is SPAG4, sperm-associated antigen 4. SPAG4 belongs to SUN family and is a nuclear envelope protein. Second, SEPT12 are co-localized and interacted with SPAG4 and LAMIN during spermiogenesis and male germ cell cell line. Third, the structure of nuclear envelope is specifically dis-regulated by ectopic expression of SEPT12, excluding altered expression of SEPT1, SEPT6, SEPT7 or SEPT11. In this study, we identified a novel role of SEPT12 for regulated the structure of nuclear envelope during human spermiogenesis.

FC065

COMPARISON OF CYCLE NUMBER TO SUCCESSFUL IVF OUTCOME AFTER ICSI FROM EJACULATE SPERMATOZOA, PERCUTANEOUS EPIDIDYMAL SPERM ASPIRATION AND TESTICULAR SPERM EXTRACTION IN SEVERE OLIGOZOOSPERMIA PATIENTS

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Objective

The aim of this study was to investigate the IVF cycle number required for embryo transfer, gestational sac (GS) observation and live birth after ICSI from ejaculate spermatozoa, PESA and TESE in severe oligozoospermia patients.

Materials and Methods

This study was retrospectively analyzed in 208 cases with severe oligozoospermia (sperm density: $<1.0 \times 10^6/\text{ml}$) during January 2008 until December 2011. The oocytes were retrieved by mild stimulation or natural cycles and inseminated by ICSI from ejaculate spermatozoa (Oligo: n=66), PESA (n=83) or TESE (n=69). Fresh or Frozen-thawed single embryo transfer (SET) was performed and the number of cycles required for first time SET, GS observation and live birth was compared between Oligo, PESA and TESE. Mann-Whitney U test was applied for statistical analysis.

Results

The mean age of patients was 39.1 ± 4.8 in male and 34.8 ± 4.5 in female and there was no significant differences between each case groups. The number of cycles required for first time SET, GS observation and live birth was significantly fewer in PESA cases compared with Oligo and TESE cases (PESA vs Oligo and TESE ; SET: 1.35 ± 0.65^a vs 1.88 ± 1.76^a and 1.96 ± 1.08^b , GS: 2.78 ± 2.14^a vs 4.68 ± 3.92^b and 3.72 ± 2.89 , Live birth: 3.19 ± 2.37^a vs 4.68 ± 3.47^b and 4.29 ± 2.96^b , $P < 0.05$, difference between a and b).

Conclusion

The number of cycles required for SET, GS observation and live birth was significantly fewer in PESA cases suggesting that the duration to yield successful pregnancy outcome was shorter after ICSI from PESA than ICSI from ejaculate spermatozoa and TESE in severe oligozoospermia patients.

FC066

MESA: DOES IT CONTINUE TO IMPACT ICSI RESULTS?

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Introduction: Testicular sperm extraction (TESE) is a widely accepted sperm retrieval method due to its technical simplicity. Obstructive azoospermic patients involving the irreparable congenital absence of seminal tract or failed microscopic seminal reconstruction, are suitable candidates for microscopic epididymal sperm aspiration (MESA). We assessed whether MESA has an impact to ICSI results.

Materials and Methods: We underwent sperm retrieval for 182 azoospermic patients between 2011 and 2012. Of these, thirty-five patients underwent MESA. MESA was employed for unilateral epididymis except in instances characterized by only immotile sperm retrieval. Conventional TESE were conducted in the presence of immotile sperm exclusively in the MESA specimen.

Results: Motile sperm recovery was realized in 30 patients as a result of MESA; 4 patients subsequently underwent conventional TESE; however, the remaining one subject exhibited only immotile sperm. Collected sperm was cryopreserved and supplied for ICSI. Thirty-one couples received ICSI involving frozen thawed sperm. Pregnancy was achieved in 23 (74.2%) and healthy delivery was observed in 20. One patient underwent vasoepididymostomy following MESA.

Conclusion: MESA needs no special requirements, e.g., mincing and disposition, prior to cryopreservation, unlike TESE; moreover, easily apply to ICSI. These data demonstrated that MESA has still impact on ICSI results significantly.

FC067

A GLOBAL PROTEOMIC APPROACH FOR ELUCIDATING THE MECHANISM OF THE ACTION OF AIRE IN GERM CELLS

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Autoimmune Regulator (Aire) is a gene that is usually associated with a rare autosomal recessive autoimmune disease, Autoimmune polyendocrinopathy-candidiasis-ectodermal dystrophy (APECED). The gene product, AIRE protein, a known transcriptional factor has been extensively studied in the thymic epithelial cells where it has been shown to play an important role in maintaining self tolerance. But its role in testis, a tissue which also shows heavy expression of AIRE is not very well understood. Previous studies have pointed towards a possible role of AIRE in regulating germ cell apoptosis suggesting that AIRE may be important during normal spermatogenesis. Homozygous Aire deficient mice were shown to reproduce only occasionally indicating that AIRE might also impact fertilization and embryo development. In this report we have evaluated how AIRE alters the cellular proteome of GC1 cell line, a germ cell derived cell line. High efficiency capillary liquid chromatography-tandem mass spectrometry (LC-MS/MS) was used to analyze proteins extracted from AIRE transfected and untransfected GC1 cells. Peptide identification was performed using PLGS (Protein Lynx Global Server) software. Functional analysis of the proteins was carried out using DAVID software. We observed that AIRE transfected cells showed increased levels of several cytoskeletal proteins as compared to non transfected cells. Our lab as well as others have previously shown that AIRE co localizes with the cytoskeletal filaments. Taken together we postulate that AIRE might mediate its action in GC1 cell line through regulation of the cytoskeletal elements.

FC068

IS MICRO-DISSECTION TESTICULAR SPERM EXTRACTION THE MINIMUM STANDARD OF CARE FOR NON-OBSTRUCTIVE AZOOSPERMIC MEN?

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Introduction: About 10% of male factor infertility is due to obstructive (OA) or non-obstructive azoospermia (NOA). The latter involves a choice of surgical techniques for testicular sperm extraction (TESE). This study evaluated the sperm retrieval rate

(SRR) of our unit's practice and compared these results with published retrieval rates from micro-dissection TESE (micro-TESE) which has been suggested as the 'gold standard' approach.

Methods: A retrospective analysis of SRRs was conducted of consecutive NOA men over a four-year period in a large multicenter and multi-doctor practice. On the day of egg collection, the protocol called for an initial fine needle aspiration (FNA) from one or both testes followed by, if needed, random open biopsies (one-three sites per testis) of one or both testes.

Results: In 216 FNAs, sperm was found in 147 cases (SSR 68%). Of the 94 open biopsies conducted, either as the primary intervention (n=68) or following an unsuccessful FNA (n=26), sperm was retrieved in 49 cases (52%). Overall successful sperm isolation was achieved in 63% of men.

Conclusions: FNA is frequently successful in NOA, requires less surgical skill and resources, and has a low morbidity and cost. That subgroup proceeding to open TESE achieved SRR similar rate to that reported with microTESE. The overall SRR rate is similar to that of experienced programs exclusively using microTESE. We find the sequential FNA/open TESE approach to be effective while reserving microTESE for selected cases with a poor prognosis cases (e.g. prior TESE failure) or Klinefelter's syndrome.

FC069

PAIN PATHOGENESIS IN ENDOMETRIOSIS

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Endometriosis-associated chronic pain symptoms are dynamic and do not correlate with anatomical disease severity. How pain persists in endometriosis may be related to the sensory innervation of lesions and eutopic endometrium and nociceptive sensitisation.

Assessment of peripheral and central nociceptive system changes in women with endometriosis was done with Quantitative Sensory Testing (QST). QST measured how nociceptive stimuli were perceived in women with persistent endometriosis-associated pain, compared to pain-free controls. QST activated the nociceptive system by the application of defined stimuli to cutaneous sites, in a controlled laboratory setting.

Nineteen confirmed-endometriosis and 20 control-women rated the intensity of a single pinprick stimulus and of a series of 10 pinprick stimuli, at one second intervals; scored as 0-no pain to 100-worst possible pain. Application sites included the dorsum hand (C7), abdomen (T11) and back (L4). Pain intensity scores were analysed.

Preliminary findings suggest women with endometriosis experience changes in pain sensitivity in regions close to the confirmed pathology: greater back sensitivity to stimuli correlated with greater abdominal sensitivity ($r=0.58$, $p=0.01$). Additionally, greater abdominal sensitivity correlated with greater hand sensitivity ($r=0.54$, $p=0.01$), suggesting changes in pain processing at sites distant to the confirmed pathology.

Detailing the peripheral and central changes in the processing of nociceptive information in women with endometriosis suggests widespread changes in pain sensitivity. Sensitisation may be due to altered neuronal activity, enhanced response to sensory inputs or diminished nociceptive inhibition. Sensitisation is a step on the pathway to chronic pain.

FC070

IMPLEMENTATION OF ROBOTIC IN REPRODUCTIVE SURGERY IN INDONESIA

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Robotic Surgery has been a remarkable breakthrough in minimal invasive surgery. Indonesia has started to acquire the technology in 2012 and has gained great interest in many surgical specialties. 50 cases have been performed in the first 12 months with mainly in gynecology reproductive field.

The clear benefit of minimal invasive over open surgery has been shown. The data to support the use of robotic in reproductive field remains unclear. Robotic hysterectomy and myomectomy are the main indications that have been shown to have clinical benefit. In endometriosis, to most experienced surgeons laparoscopy may still provide an excellent outcome. Unfortunately not many surgeons are competent and skillful enough to manage more advanced disease. In 40% of our case series were in advanced stage of endometriosis and 26% were for myomectomy. Clinical outcome were significant in amount of bleeding with 85% less than 100cc and pain score of no greater than 4 in 24 hours and 40% of only 1.

Successful spontaneous pregnancies have resulted from myomectomies group. In advanced endometriosis with fertility indication, AMH post surgery reduced to only 18 % compared to known 33-61% reduction after conventional laparoscopy. With 3D HD facility improves the surgical maneuverability and in author's experience this technology implementation has also increase the level of conventional laparoscopy competencies. Robotic surgery may increase the expense but with greater surgical excision of pathology in which debates will continue over the overall outcome.

FC071

ADIPONECTIN SERUM LEVELS IN ENDOMETRIOSIS

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Endometriosis is a disease that is suspected estrogen-dependent. Increased estrogen levels have an impact on increasing the activity of endometriosis. Adiponectin has pleiotropic effect, i.e. the effects of anti-inflammatory and anti-angiogenic. Low levels of adinopectin resulted in increased levels of free estrogen and inability of angiogenesis and inhibition of inflammatory processes that lead the development of increasingly severe endometriosis. This study aimed to determine differences in serum adinopectin levels in patients with endometriotic cyst compared with non-cystic endometriosis and its relationship with the stage of endometriosis. The design of this study was the comparative analytic cross cut method in women with endometriotic cyst (n=25) and women with non-endometriotic cyst (n=25), which is done either laparoscopic surgery or laparotomy in Dr. Hasan Sadikin Hospital. Results showed no significant difference ($P>0.05$) on the characteristics of the study subjects in term of age and body mass index. The results showed a significant difference ($P<0.001$) between the mean levels of adinopectin group of endometriosis patients (3.91 mg/ml) than in patient with non-endometriois group (8.59 mg/mL). The results also showed no significant difference ($p>0.005$) between endometriosis stage III adiponectin levels (4.24 mg/mL) with stage IV endometriosis (3.54 mg/mL). It can be concluded that serum adinopectin levels in patient with endometriosis is lower than the non-endometriotic cyst. Adiponectin levels are not associated with the stage of endometriosis.

Key words: adiponectin, endometriotic cyst, endometriosis stage

FC072

THE EFFECT OF INCREASING DOSE OF CURCUMIN SUPPLEMETNATION ON EXPERIMENTAL ENDOMETRIOSIS PROGRESSIVITY IN MICE

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Background : To evaluate increasing dose of curcumin supplementation on experimental endometriosis progresivity in mice.

Study : Animal laboratory experimental study

Methods: 28 randomized mice (*mus musculus*) were given cyclophosphorin A and estrogen injection intramuscularly and human endometrium intraperitonealy on day 1 to induce endometriosis like lession. On day 14 mice were divided into 4 groups, group A had placebo and group B,C,D had curcumin 240 gr/kgbb, 500gr/kgbb and 1000gr/kgbb respectively. treatments were given for 7 days. On day 28 mice were sacrificed and evaluated area of endometriosis like implant, immunohystological staining of Prostaglandin E2 (PGE2) and Matrix metalloproteinase 9 (MMP9) in peritoneum.

Result: area of endometriosis like implant on peritoneum on group A,B,C,D were $348.43 \pm 185.1 \text{ mm}^2$, $140.14 \pm 75.59 \text{ mm}^2$, $79.43 \pm 26.98 \text{ mm}^2$, $31.71 \pm 7.02 \text{ mm}^2$ ($p = <0.001$). PGE2 on group A,B,C,D were 5.14 ± 1.95 , 4.28 ± 1.38 , 3.43 ± 1.62 , 2 ± 0.58 respectively ($p = 0.003$), MMP9 on group A,B,C,D were 3.71 ± 1.89 , 2.25 ± 1.97 , 1.43 ± 0.78 , 1.28 ± 0.49 respectively ($p = 0.034$). We found a significant positive correlation between PGE2 ($r = 0.735$) and MMP9 ($r = 0.561$) with area of endometriosis like implant.

Conclusion: On experimental endomentriosis in mice, endometriosis progresivity responded to increasing dose of curcumin in dose dependend manner. PGE2 had better correlation with endometriosis than MMP9. Further study is needed

FC073

THE EFFECTIVENESS OF DLBS 1442 IN ALLEVIATING ENDOMETRIOSIS- AND/OR ADENOMYOSIS-RELATED PAIN

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Background

The overexpression ER- β and the COX-2 enzyme coupled with the absence of expression of progesterone receptors (PR) is critical to the pathogenesis of endometriosis- and adenomyosis-associated pain. DLBS1442, a novel bioactive extract of *Phaleria macrocarpa*, exerts its action by down-regulating the overexpressed ER- β and COX-2 products and up-regulating PR gene expression.

Design

This case study was conducted to evaluate the effectiveness of DLBS1442 treatment in alleviating endometriosis- and /or adenomyosis-related pain.

Setting

Ten endometriosis and/or adenomyosis patients were recruited consecutively at Yasmin Clinic, Dr. Cipto, Mangunkusumo General Hospital between January and March 2013.

Method

Pain associated with menses, including pre-menstrual pain, dysmenorrhea, dyschezia and dysuria, was measured using the Visual Analog Scale (VAS). Patients reporting one or more pain symptoms with a VAS score ≥ 4 were given 100 mg of DLBS1442 three times daily for 12 weeks. The VAS score for each pain symptom was evaluated at each of the next three menstrual cycles.

Results

VAS score reduction was noted in the first post-treatment menstrual cycle (approximately 5.3 weeks after treatment initiation), and VAS scores continued to decline over the final two cycles. The average VAS score reduction per cycle for symptoms of pre-menstrual pain, dysmenorrhea, dyschezia and dysuria was 0.4, 1.6, 1.6 and 0.9, respectively

Conclusion

This case study demonstrated that DLBS1442 was effective in alleviating endometriosis- and / or adenomyosis-related pain, as demonstrated by early pain reduction as evaluated using the VAS. Pain continued to decline during each subsequent menstrual cycle.

FC074

FACTORS ASSOCIATED WITH SUCCESSFUL PREGNANCY AFTER CONSERVATIVE SURGERY FOR ADENOMYOSIS.

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Objective: To analyze the determinants of successful pregnancy following laparoscopic adenomyomectomy. **Study design:** Retrospective cohort study. **Population:** A total of 141 women who underwent laparoscopic adenomyomectomy during 5 years. **Method:** Surgical data was retrieved from the patients' database. Pregnancy outcomes were collected from questionnaires. We defined the 'clinical pregnancy' as the presence of fetal heartbeat at 12 weeks of gestation. Median observation period was 24 months. Step wise logistic regression analysis was employed to analyze determinants of clinical pregnancy. **Results:** Among the 141 women who underwent conservative surgery, 112 desired pregnancy. When the women were divided into under 39 y.o. and over 40 y.o. groups, clinical pregnancy rates were 41.3% and 3.7%, respectively. Factors associated with successful pregnancy were: history of preoperative IVF; posterior wall involvements of adenomyosis; and age at surgery, with the odds ratios of 7.83, 0.17, and 0.82. In the younger group, a total of 60.8% of women with the history of preoperative IVF failure showed successful pregnancies after the surgery. **Conclusions:** Posterior wall involvements of adenomyosis and age at surgery were the negative impact factors of successful pregnancy after adenomyomectomy. Surgical intervention may have a positive effect on postoperative IVF outcome in patients of repeated IVF failure.

FC075

CROSS-BORDER REPRODUCTIVE CARE – ONE CLINIC'S NATIONAL AND INTERNATIONAL EXPERIENCE

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Background

Cross-border reproductive care (CBRC) is the accession of ART in another country or jurisdiction to access better, more comprehensive, private, or cheaper care or treatments unavailable in the home country/state for regulatory or ethical reasons. The principal concern around CBRC is the potential for harm from services sought in countries where health and safety concerns exist over clinical practice standards. The incidence of CBRC has been estimated at 4% (USA) to 5% (Europe). Federated countries like Australia with varied state laws contribute to CBRC similar to international CBRC.

Aim

To assess CBRC nationally between the federated Australian states and in patients transporting their cryopreserved reproductive tissues internationally.

Method

Donor sperm patient records for 2008-2013 assessed for their state or country of residence, along with international exports of cryopreserved reproductive tissues.

Results

CBRC patients seeking donor semen comprised 14(2008), 34(2009), 57(2010), 65 (2011), 29 (2012). The demand peak followed introduction of Victorian pre-treatment police checks and a NSW donor register, both in 2010. Increased demand came mostly from these two states and largely from single women rather than same-sex and heterosexual couples. 18 patients came from overseas, predominantly New Zealand. In the same period patients shipped 23 lots of semen and 12 cohorts of embryos overseas, seeking donated oocytes, surrogacy and sex selection.

Discussion

CBRC is an inevitable consequence of differing laws, costs and service accessibility. Provided the potential for harm to the patients or any resulting baby is not increased, CBRC should be recognised, accepted, and facilitated.

FC076

OLDER, HEAVIER AND SINGLE: HOW THE FEMALE IVF PATIENT HAS CHANGED OVER THE PAST FIFTEEN YEARS.

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Aim

Improvements in IVF have led to significant increases in both treatment and success rates. However, the average age of women using IVF has also increased. This study examines this and other changes in the population of women undergoing IVF in our clinic.

Materials and Methods

1532 women undergoing 3136 cycles of oocyte recovery from 1998 to 2013 were included in the study. The mean age, mean weight, B.M.I. and aetiology of infertility were analysed for each year of the study period and compared using regression analysis.

Results

The mean age of women undergoing treatment increased significantly from 33 ± 4.6 years in 1998 to 34.4 ± 5.3 in 2013 (P=0.001). This resulted from a significant shift in the proportion of patients < 35 to those > 39 (P = 0.001). Mean female weight also increased significantly (65.9Kg to 76.8Kg; P = 0.0009) as did B.M.I. (24.9 to 27.9; P < 0.0001). There was little overall change in the distribution of aetiology from 1998 to 2013. However, when only female aetiology is analysed, there has been a marked increase in the past five years in the incidence of single women and same sex couples having treatment (2.3% in 2008 to 19.4% in 2013) and the use of AMH to define the cause of infertility.

Conclusion

Women attending our clinic in 2013 are significantly older and heavier with a higher B.M.I. than they were in 1998. The aetiology of infertility has also changed with more women choosing IVF for social reasons.

FC077

THE EFFECT OF COMPULSORY IDENTIFICATION OF SPERM DONORS ON SPERM DONOR PRACTICE

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Introduction. It is commonly asserted (Bay et al., Fertil Steril, epub 2013) that removal of anonymity will prohibitively inhibit sperm donor recruitment. This was investigated by studying characteristics of donor practice, before and after legislation in New South Wales prohibiting anonymity (2010).

Method. A retrospective data review was conducted at IVFAustralia comparing donor treatment cycles in 2003-2005 (992 cycles) and 2010-2012 (1018 cycles). The review studied: sperm donor characteristics (Anonymous, no consent to release of identifying information; Identifiable, identity recorded for later access by offspring; Known, identity of donor known to recipients); sperm donor recruitment; characteristics of recipients accessing treatment. Percentages are by treatment cycles.

Results: 1. *Treatment cycles.* 2003-2005, donor characteristics were 37% anonymous, 48% identifiable, 15% known donors. In 2010-2012, 2% anonymous, 77% identifiable, 23% known. 2. *Donor recruitment.* In 2003-2005, 5 identifiable donors (mean age 39.4) recruited in NSW. From 2010-2012, 62 donors recruited in NSW (mean age 36.8). 3. *Characteristics of recipients.* In 2003-2005, the characteristics (mean age 37.2yr) were; 56% heterosexual couples, 36% single women, 8% same sex couples. Treatment mode was; 51% intrauterine insemination (IUI), 36% IVF/ICSI, 13% frozen embryos (FET). In 2010-2012 the characteristics (mean age 37.8yr) were; 28% heterosexual couples, 54% single women and 18% same sex couples. Treatment mode was; 28% IUI, 55% ICSI, 17% FET.

Conclusions: This work supports previous findings (Shukla et al., HumReprod676:28;2013) that recruitment strategies for sperm donors remain effective despite compulsory identifiability. The pattern of women accessing donor sperm has changed in the past decade.

FC078

YOU HAVE MILLIONS WE ONLY WANT ONE! A SMALL CLINICS EXPERIENCE IN ESTABLISHING A SPERM DONOR RECRUITMENT PROGRAM.

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The number of sperm donors being recruited by Australian clinics has declined over the years, making it difficult for clinics to keep up with the demand for donor sperm. The reasons for this decline in sperm donors could partially be attributed to the complexities associated with donors having to agree to be contactable by offspring when they reach the age of 18 (if not earlier) and the fact that it is not 'legal' in Australia to buy and sell human tissue (Human Tissue Act 1982 S.38). Recruiting has become an arduous task to say the least and as a result, many clinics have chosen not to embark upon this option, choosing instead to either refer patients to other clinics who offer treatment with clinic recruited donor sperm or import donor sperm from overseas. The moral and ethical issues that importation presents are many and varied; however, local demand for this service is becoming ever more apparent. As a result, the team at Fertility Solutions Sunshine Coast decided to establish a local donor sperm recruitment program. The process taken to establish such a program will be outlined giving consideration to some of the legal, social, moral and ethical issues encountered along with the rationale for some of the decisions made. Has this approach made a difference?

FC079

UNIQUE HRT REGIMEN PROVES MOST EFFICIENT FOR FET CYCLES

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Aim:

Retrospective audit of pregnancy rates in FET program utilising LDS and HRT schedules.

Method:

PIVET has increasingly moved towards an HRT regimen for FET cycles to avoid issues arising from LDS and natural cycles, namely cancelled cycles, weekend transfers and multiple pregnancies.

From January 2011 to August 2013, PIVET applied a unique HRT regimen with Progynova tablets from day1 of the cycle then progressively introducing pessaries from day10; Estradiol progressing to Progesterone and an Estradiol/Progesterone combination pessary.

Day3 embryos (mostly SET) are transferred on day4 of progesterone pessaries or a single blastocyst on day6.

Mid-luteal serum Estradiol and Progesterone levels are then tested 9 days post-transfer and adjustments made to the regimen if not adequate.

Results:

Overall 260 transfers were performed on Day3 across all age groups with 63 pregnancies arising (24%), the results being slightly higher for HRT. In the <35yr group the pregnancy rates for HRT and LDS were the same at 39% and 38% respectively.

There were 581 blastocyst transfers with 268 pregnancies arising (46%) with HRT rates slightly higher. In the <35yr group the pregnancy rates for HRT (53%) were higher than non-HRT cycles (44%).

Conclusion:

Having demonstrated that the HRT schedule provides potentially higher pregnancy rates, PIVET's adoption of this method for FET's is justified. It enables flexibility for avoiding weekend transfers, efficient scheduling during the working week as well as minimisation of blood tests and ultrasounds. Furthermore it reduces cycle cancellation rates and provides a safety feature with no unexpected multiple pregnancies.

FC080

ISLAMIC PERSPECTIVE ON CONTRACEPTION

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It is estimated that 80% of pregnancies are unintended contributing to high maternal mortality worldwide. Family planning can reduce 20% of the maternal mortality.

Islam encourage family planning. There is NO TEXT in the Holy Quran prohibiting the prevention of pregnancy for the purpose of family planning. It was practiced during the time of Prophet Muhammad and he did not object. However, Islam does not allow termination of pregnancy unless with valid reasons. In Islam, family planning is practiced within marriage. The consent of both husband and wife is required to undertake a safe and legitimate method of family planning. Islam encourages family planning due to economic and health reasons.

All methods of contraception are permissible in Islam. The patch is limited in Islam as it has to be removed during the bath after sexual activity. It is then not going to stick again. The consent of the husband is mandatory.

The Muslim leaders have declared that family planning as permissible when the wife is too ill or weak, or the presence of hereditary disease or when the pregnancies are too frequent leading to social and economic difficulties. What more when Muslim women are encouraged to take care of their 'beauty', one of which is to be healthy.

Finally, Islam allows any contraceptive method with the aim to serve Allah. It should promote Muslim towards good deeds with honorable objectives. and to practice 'My Home is My Heaven'. After all Islam is a religion of ease, quality and moderation.

FC081

BEYOND THE GOOD PROGNOSIS PATIENT: EXTENDED CULTURE INCREASES LIVE BIRTH RATES FOR PATIENTS, REGARDLESS OF QUALITY AND QUANTITY OF EMBRYOS ON DAY 3

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Introduction:

There is Level 1a evidence confirming the value of extended culture (EC) for good prognosis IVF patients. However, there is still controversy regarding the benefits of EC for patients with few or poor quality embryos. Here we explore whether the live birth rate (LBR) following single embryo transfer (SET) on day 3 (D3) or day 5 (D5) varies with the number of good quality embryos available on D3.

Method:

Retrospective analysis of 6644 patients seeking treatment at Monash IVF between 2006-2012. Inclusion criteria: first IVF cycle, at least one embryo suitable for transfer on D3, and SET. Data analysis: binary logistic general linearised model (SPSS v20.0) to calculate adjusted odds ratios for LB, accounting for age, etiology, stimulation protocol, ICSI/IVF, endometrial thickness, BMI, and quality/quantity of D3 embryos.

Results:

The LBR is 40% higher following D5 SET compared to D3 SET (aOR 1.39, 95%CI: 1.21-1.59, $p < 0.0001$), regardless of the number of zygotes (≤ 4 , 5-8, ≥ 9) or the quality and number of D3 embryos. As per the inclusion criteria all patients electing a D3 SET had a transfer, whereas patients proceeding with EC had a 3.4%, 1.3% and 0.1% cancellation rate for cycles with ≤ 4 , 5-8, ≥ 9 zygotes respectively. Patients in the D3 group had a significantly higher number of frozen embryos.

Conclusions:

When at least one transferable D3 embryo is available, opting for extended culture is associated with higher LB rates even in women who had less than 4 zygotes available

FC082

CONTINUOUS IMPROVEMENT IN NATIONAL ART STANDARDS BY THE RTAC ACCREDITATION SYSTEM IN AUSTRALIA AND NEW ZEALAND

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Background

The Reproductive Technology Accreditation Committee (RTAC) accreditation system evolved in 1986 as an Australian requirement for patient access to government subsidised treatment. It used volunteer peer reviewers from within the profession. In 2008 it evolved into an annual audit by professional Certifying Bodies approved by JAS-ANZ for compliance with a Code of Practice (COP) containing both Critical Criteria (CC) and Good Practice Criteria (GPC), reviewed triennially. Additionally, clinics submit de-identified treatment data to a national database for analysis and comparison.

Aim

To assess the incidence of variances and findings identified by Certifying Bodies within the currency of the most recent COP version.

Methods

Results of findings by certifying bodies for 87 Australian and New Zealand clinics inspected in four six-month periods between July 2011 and June 2013 were analysed, divided according to CC and GPC.

Results

The mean number of CC variances per clinic fell from 1.53 to 0.14 and the number with variances falling from 77% to 14%. Within GPC, mean variances per clinic fell from 77% to 14% while the number with no findings was relatively constant around 50%.

Discussion

The RTAC accreditation system has contributed to steady improvement in standards achieved. Findings now are largely of negligible patient risk, predominantly documentation issues. The stable incidence of Good Practice Criteria findings reflects the true nature of quality management systems - continual improvement seeking maximal risk management and treatment outcomes. There is evidence that this scheme has led to improved ART success rates whilst progressively reducing adverse outcomes.

FC083

RISK OF PLACENTA PREVIA IN ART BIRTHS LINKED TO ENDOMETRIAL THICKNESS

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Introduction:

Compared to naturally conceived pregnancies, ART pregnancies are associated with a six-fold increased risk of placenta previa (PP), a serious obstetric complication. Endometriosis is independently associated with an increased risk of PP. Together these findings suggest that the risk of PP relates to changes in the endometrium. Here we investigate the relationship between endometrial combined thickness (ECT) measured prior to embryo transfer (ET) and the relative risk of PP.

Methods:

Observational study between 01/01/2006 and 30/06/2012 of 3306 IVF births following single ET in a fresh or frozen cycle. The dataset was analysed using a binary logistic General Linearised Model (SPSS v20.0) to calculate odds ratios for PP adjusted for known confounders (aOR).

Results:

A total of 145 cases of PP were observed. Endometriosis was associated with a two-fold increased risk of PP (aOR 2.00; 95%CI: 1.21-3.31). Compared with an ECT of <7mm the risk of PP was two- and four-fold higher respectively for cycles in which the ECT was 7-12mm (aOR 2.02; 95%CI: 1.12-3.65) and >12mm (aOR 3.84; 95%CI: 1.95-7.57). ECT measurements were not available for natural cycle FETs but in a separate analysis they were associated with a more than two-fold lower risk of PP compared to HRT-FET or fresh stimulated cycles (aOR 0.44; 95%CI: 0.28-0.71).

Conclusions:

Increased ECT prior to ET is associated with higher risk of PP, but the pathophysiology remains unclear. Further studies are planned to elucidate whether ECT is a surrogate marker of increased uterine contractility or altered endometrial receptivity.

FC084

THE IMPACT OF MATERNAL BODY MASS INDEX ON IMPLANTATION RATES FROM EUPLOID FROZEN EMBRYO TRANSFERS

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Background:

Maternal obesity is associated with sub-optimal reproductive outcomes and decreased pregnancy rates. However, the exact mechanisms by which BMI affects reproductive success remain unknown. It is thought that high maternal BMI may impact embryo implantation by altering the uterine environment. This study investigated the impact of maternal BMI on implantation following the transfer of a known euploid embryo(s) in a frozen embryo transfer cycle.

Methods:

Pregnancy rates were retrospectively analysed from frozen embryo transfer cycles where only known euploid embryo(s) were transferred (n=97). The BMI categorisations were as follows: normal - 18.5-24.9 kg/m²; overweight - 25-29.9 kg/m²; obese - 30-34.9 kg/m²; morbidly obese class I - 35-39.9 kg/m²; morbidly obese class II ≥40 kg/m². Embryo chromosome normality was determined using the 24sure array CGH preimplantation genetic screening platform.

Results:

Women with a normal, overweight or obese BMI had similar pregnancy rates following the transfer of a known euploid embryo (45.8%, 45.8%, and 46.2% respectively). Women with a BMI in the morbidly obese I and morbidly obese II did not

achieve pregnancy following the transfer of a known euploid embryo. Pregnancy rates in the normal, overweight and obese women were similar regardless of maternal age.

Conclusions:

Obesity ($\text{BMI} \geq 35 \text{ kg/m}^2$) in women of all ages reduces clinical pregnancy rates. The current data suggests that obesity impairs embryo implantation. Furthermore, the age related decline in pregnancy is less prominent following the transfer of a euploid embryo in women with a $\text{BMI} < 35 \text{ kg/m}^2$, further suggesting that maternal obesity impairs endometrial receptivity.

FC085

A PROSPECTIVE RANDOMISED CONTROLLED STUDY COMPARING THE COST EFFECTIVENESS OF IVF-ICSI TREATMENT: CLEAVAGE STAGE (DAY 3) EMBRYO TRANSFER VERSUS EXTENDED CULTURE(DAY 5/6 BLASTOCYST) TRANSFER.

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Objective: Worldwide, it has become necessary to perform the more economical Day 2/3 transfer in order to reduce the cost of extended Day 5/6 culture. This prospective study was aimed at comparing the outcome and cost of Day 3 transfer versus Day 5/6 (blastocyst) transfer.

Design: The present study is a prospective, randomized controlled trial carried out between Jan 2007 and June.2013, for first IVF-ICSI.

Materials and Methods: Couples were assigned to Day 3 or Day 5/6 transfer by a computer generated randomised list. If more than 6 oocytes were retrieved and minimum three top quality embryos were observed at day 3, couples were included in the study. Fresh embryo transfer was performed at day 3 for 258 couples and at day 5/6 for 196 couples. The primary outcome measured was the clinical pregnancy rate and cost of cycle.

Results: The clinical pregnancy rate was 28.9% for Day 3 transfer and 30.7% for Day 5/6, the difference between the two being not statistically significant.

Conclusions: At day 3 post oocyte-retrieval, with 3 top quality embryos, the pregnancy outcome is similar after embryo transfer at cleavage stage or blastocyst stage. Moreover, the day 3 embryo can be better assessed morphologically than the day 2 embryo. These results have shown that day 3 transfers do not compromise pregnancy outcome compared to day 5/6 transfers, with top quality embryos yielding the highest implantation. Cleavage stage (day 3) transfer can therefore be considered a better and more economical option to expensive day 5/6 blastocyst culture.

FC086

INFLUENCE OF RE-VITRIFIED HUMAN BLASTOCYST ON LABORATORY DATA, CLINICAL OUTCOMES AND RESULTING BABIES

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Introduction

Elective single embryo transfer of blastocyst has been increasing lately. Therefore, the opportunity of not only vitrified, but also re-vitrified blastocyst has also increased. The objective of the present study was to determine the influence of re-vitrified blastocyst on laboratory and clinical outcomes of frozen-thawed embryo transfer compared to one time vitrified blastocyst

Materials& Methods

Total of 1450 pregnancies between March 2006 and September 2012 from vitrified (n=1433) and re-vitrified (n=233) blastocyst transfers was used for analysis. Survival rate after thawing, pregnancy rate, miscarriage rate and the rate of congenital abnormality at birth were compared. There were 9 cases of stillbirth or abortion in the control group.

Results

Survival rates after thawing in re-vitrified and vitrified group were 94.0% (219/233) and 97.4% (1402/1433) ($p < 0.05$), respectively. Pregnancy rates were 39.5% (77/195) and 50.2% (628/1251) ($p < 0.05$). Miscarriage rates were 23.4% (18/77) and 24.4% (153/628) (n.s.). The rates of congenital abnormality were 0% (0/50) and 2.7% (10/368).

Conclusions

Survival rate after thawing and pregnancy rate of re-vitrified blastocysts were significantly lower than those of vitrified blastocysts. This might suggest that the process of repeated vitrification-warming may affect the quality of blastocysts or the quality of re-vitrified blastocyst was somewhat lower. Congenital abnormality did not increased by repeated vitrification so far. Re-vitrification of blastocyst decreased survival and pregnancy rates, but does not affect resulting newborn baby.

FC087

THREE YEARS EXPERIENCE IN A REGIONAL CLINIC OF PREIMPLANTATION GENETIC DIAGNOSIS OF ANEUPLOIDY (PGD-A) USING EMBRYO TRANSPORT AND ARRAY-COMPARATIVE GENOMIC HYBRIDISATION (ACGH)

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Aim

PGD-A using aCGH facilitates the selection of chromosomally normal embryos and an increased pregnancy rate. However, the cost of providing the full service is prohibitive to most regional clinics. This study reports the results obtained after transporting day 3 embryos to a capital city clinic for biopsy and aCGH and their subsequent return for transfer and cryopreservation.

Materials and Methods

After ovarian stimulation, oocytes were retrieved and fertilised by ICSI. Good quality embryos were transported on day 3 from Toowoomba to Brisbane for single blastomere biopsy. The blastomeres were air-freighted overnight to Melbourne IVF for whole genome amplification and arrayCGH using 24sure BAC microarrays (BlueGnome, UK). Biopsied embryos were returned to Toowoomba on day 4. Embryo quality was assessed on day 5 and normal embryos were transferred or vitrified on the same day.

Results

337 embryos from 65 patients were biopsied. 324 of 337 (96%) blastomeres yielded conclusive results, with 69 embryos (21.3%) diagnosed as euploid. 23.8% of the aneuploid embryos had a single aneuploidy, while 16.1% showed two aneuploidies and 37.1% had complex aneuploidy involving ≥ 3 chromosomes. 73.1% of biopsied euploid embryos formed blastocysts on day 5, while the proportion of biopsied aneuploid embryos developing to blastocysts correlated with the array analysis (single: 56.8%, double: 48.1% and complex: 27.3%). Embryos were transferred in 41 cycles resulting in 11 clinical pregnancies (26.8%).

Conclusion

The combination of embryo transport with biopsy and aCGH offers patients in rural centres access to the same technology as those in larger cities.

FC088

PARTIAL GENOME SEQUENCING - AN ALTERNATIVE TO MICROARRAYS FOR EMBRYO ANEUPLOID SCREENING

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It is becoming more evident that screening embryos for total chromosomes results in improved implantation rates and live birth rates. In the literature, embryo aneuploidy rates are routinely reported to vary from 40-75% according to the maternal age. Identifying and avoiding chromosomally unbalanced embryos logically should improve likelihood of successful pregnancies in women of all ages. Segmental aneuploidy in some couples, where one is a translocation carrier, further compromises the possibility of balanced chromosomes. Any alternative testing procedure should also be able to accurately identify segmental losses and gains for it to be a valid consideration.

While the microarray has become a standard for such screening, the cost per analysis can be quite prohibitive for some couples and reduces the possibility of wider implementation of aneuploid screening. Massively parallel sequencing offers an opportunity to reduce cost without loss of utility.

We used partial genome sequencing (PGS) to rapidly karyotype whole-genome-amplified samples from 25 human blastocysts and compared these with array-based comparative genomic hybridization (array CGH) data. Each embryo was analysed for 40,000 - 800,000 reads across the genome. There was concordance between the platforms. PGS, however, provided additional flexibility, scalability and reduced cost. The resolving power of PGS can readily be adjusted from screening whole-chromosome aneuploidies, to sensitive detection of segmental intrachromosomal copy number variations and mosaicism.

Next Generation Sequencing (NGS) has the potential to provide a uniform simplified approach to the screening of embryos and reduce associated costs making it an appealing opportunity for routine IVF use.

FC089

A CHANGE IN STRATEGY FOR PREIMPLANTATION GENETIC DIAGNOSIS (PGD) PATIENTS – COMBINING DETECTION OF SINGLE GENE DEFECTS WITH COMPREHENSIVE CHROMOSOME SCREENING IN ELECTIVE FREEZE ALL CYCLES

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Couples carrying single gene mutations often face a dilemma regarding their reproductive options. IVF-PGD has become a popular choice for such couples. PGD for single gene disorders is done using a combined approach of specific mutation(s) detection in conjunction with identification of the carrier chromosome(s) through familial STR linkage analysis.

Recently, general IVF patients have been able to use CGH to exclude chromosomally abnormal embryos with resultant improved implantation and pregnancy outcomes by lowering miscarriage rates and avoiding futile embryo transfers especially in patients of advancing maternal age, with repeated implantation failure or recurrent miscarriage. For various reasons, aneuploidy testing is typically considered incompatible with testing for single gene disorders.

Screening and selecting embryos for total chromosomes has revealed aneuploidy as the biggest contributing embryonic factor to achieving successful implantation. It is relevant for women of any age, regardless of their fertility status and so patients with an inherited risk from a single gene disorder could also benefit from screening aiding better selection of embryos and hence improved pregnancy outcomes.

We report here the routine application of combined testing for single gene and chromosome ploidy with results from 62 elective single embryo cryo cycle transfers. Outcomes show patients having combined testing with frozen cycle transfer exhibited improved implantation rates compared to patients having single gene testing alone and transfer within the stimulation cycle (64.5% vs 45.9% (P=0.0413), mean ages 32.8 and 33.7 respectively). Considerations around the effects of delayed embryo transfer on pregnancy outcomes will also be discussed.

FC090

IDENTIFICATION OF AN INCREASED INCIDENCE OF CFTR MUTATIONS IN MALE ART PATIENTS

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Cystic Fibrosis is the most common deleterious single gene disorder in Northern European Caucasians and has a live birth rate of about 1 in 2500 babies affected with Cystic Fibrosis (CF). CF is caused by two mutations in the Cystic Fibrosis Transmembrane Receptor gene (CFTR), usually one mutation inherited from each parent.

In the Australian population it is often quoted that the CFTR mutation carrier rate is between 1 in 25 to 1 in 30 people. We have previously shown that the CFTR mutation carrier rate is increased in the infertile population but is even further increased in the male patients that attend our clinic.

In 2500 male ART patient screens we have identified 164 CFTR mutations, which is a carrier rate of 1 in 15 or 6.6% of the male patient population (95% C.I. +/- 0.97%). Given a previously identified carrier rate of 1 in 24 female patients, that means 1 in every 360 couples coming for ART treatment are an at risk couple of having a CF affected baby.

Considering CFTR mutations are related to infertility, this raises the possibility that CFTR mutations have been under detected in live born affected individuals because of the decreased pregnancy rate amongst carriers. Is it possible that many carriers go undetected because screening takes place on new born babies rather than at the preconception stage? Could it be possible that this pattern is also replicated in other ethnic groups?

FC091

GENOME-WIDE COPY NUMBER VARIATIONS SCAN IN CHINESE PATIENTS WITH PRIMARY OVARIAN INSUFFICIENCY

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Objective: Premature ovarian failure (POF) is a heterogeneous disease defined by amenorrhea of at least 6 months duration, occurring before 40 years of age, with two FSH levels in the postmenopausal range, now referred to as primary ovarian insufficiency (POI). Currently genetic factors are thought to be the most common cause of POI. The aim of the present study is to investigate genetic causes in Chinese women with POI for genome-wide copy number variations (CNVs), focusing on novel autosomal microdeletions and microduplications. **Methods:** Genome-wide CNVs analysis using Affymetrix SNP 6.0 array was carried out in 30 Chinese POI subjects. And quantitative PCR (qPCR) were further performed for selected coding regions with microdeletions and microduplications in 30 POI subjects and another 40 POI cases. **Results:** A total of 101 CNVs were identified by SNP arrays, ranging in size from 0.1 Mb to 5.6 Mb. These CNVs included 8 novel microduplications and 12 novel microdeletions. 4 microdeletions identified in chromosomal regions 10q26.12, 10q26.3, 2p16.3, 6p26 and 2 microduplications which contained the coding regions 20p12.3 and 7p22.2 were verified by qPCR. **Conclusions:** We report the high-resolution rare CNV analysis revealing novel microdeletions/microduplications in Chinese POI patients. In the selected verified coding regions, we found five genes including *SYCE1*, *CYP2E1*, *NRXN1*, *PARK2* and *CARD11* may be involved in reproduction, thus representing potential candidate genes in POI.

Key Words: copy number variations, SNP microarray, premature ovarian failure, primary ovarian insufficiency, microdeletion, microduplication

FC092

CHROMOSOME SEGREGATION ANALYSIS AND ESTIMATION OF INTERCHROMOSOMAL EFFECT IN HUMAN EMBRYOS FROM CARRIERS OF CHROMOSOMAL REARRANGEMENT. MULTICENTRAL STUDY.

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Objective: To investigate meiotic segregation patterns and to estimate chromosomal imbalance for structurally normal chromosomes in cleavage-stage embryos from carriers of chromosomal rearrangement (Robertsonian translocation, reciprocal translocation, pericentric inversions).

Design: Retrospective multicentral study.

Materials and Methods: We analyzed 960 cleavage-stage embryos derived from 130 PGD-FISH cycles performed for chromosomal rearrangement carriers in 6 IVF centers between 2010-2013 were analyzed. 850 embryos from chromosomally normal individuals provided a control groups for comparative analysis of interchromosomal effect (ICE). For ICE analysis control and investigated groups were categorized by females' age (<35, 35-39, >39). Using FISH we determined the number copy for chromosomes involved in the rearrangement (3-6 loci per rearrangement) and the copy number of 7-12 structurally normal chromosomes (1-3 loci per chromosome). In order to examine segregation patterns we investigated some characteristics such as carrier's gender and rearrangement type. For reciprocal translocations we explored breakpoints and acrocentric chromosome involvement.

Results/Conclusion: The highest rates of normal/balanced embryos we observed in male carriers of Robertsonian translocations (40%) and in male (39%) and female (40%) carriers of inversions. Groups with reciprocal translocations showed significantly lower rates of normal/balanced embryos (male -17%, female -14%, P=0.001) than other groups. We found that some rearrangement characteristics correlate with segregation patterns. Carrier gender can affect the pattern but other characteristics, such as breakpoint, structural features are more significant. ICE was detected only in groups with female age <35 regardless of a carrier gender. This provides evidence that female age more than ICE affects the level of aneuploidy.

FC093

THE PREVALENCE AND MORPHOKINETICS OF HUMAN EMBRYOS DEMONSTRATING REVERSE CLEAVAGE WHEN VIEWED USING THE EMBRYOSCOPE™ TIME LAPSE VIDEO SYSTEM.

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Time-lapse technology has enabled embryo growth to be monitored closely in the routine setting. Various patterns of cleavage are seen, including reverse cleavage (RC) whereby blastomeres divide but then re-join. In the present study of 65 IVF/ICSI treatment cycles, 420 embryos were monitored using the Embryoscope™ time-lapse system (Fertilitech), with images taken at 10 mins intervals until Day 3. RC was more prevalent in embryos following ICSI (87/259, 33.6%) than IVF (32/161, 19.9%; $p < 0.01$), and happened more than once in 29/119 (24.4%) embryos. Of the 152 occasions of RC, 22 (14.5%) occurred at 1-cell, 67 (44.1%) at 2-3-cell and 63 (41.4%) at ≥ 4 -cell. Fewer embryos showing RC reached the 6-cell stage by Day 3 (63/118, 53.4%) compared to those not (228/302, 75.5%; $p < 0.00005$), and had a lower embryo grade (2.8 ± 1.1 vs 2.2 ± 1.2 , $p < 0.001$). RC embryos took longer to reach the 2-cell (28.2 ± 5.8 hrs vs 26.4 ± 3.7 hrs, $p < 0.005$) and 4-cell (41.1 ± 8.3 hrs vs 38.2 ± 5.8 hrs, $p < 0.005$) stages, and were slower to divide from 3 to 4 cells (3.9 ± 5.6 hrs vs 2.2 ± 4.3 hrs, $p < 0.01$) and 5 to 8 cells (11.9 ± 8.6 hrs vs 5.0 ± 5.1 hrs, $p < 0.001$). However, they progressed quicker from 4 to 5 cells (8.6 ± 6.4 hrs vs 10.3 ± 4.4 hrs, $p < 0.05$). From known implantation data, 0/11 embryos with RC implanted compared to 12/63 (19.0%) without RC. In conclusion, embryos with RC were more frequent following ICSI, were more likely to arrest before the 6-cell stage by Day 3, had division times which were significantly altered, and were not seen to implant.

FC094

STAGE SPECIFIC CYTOPLASMIC TEXTURE MEASUREMENTS ARE INDICATIVE OF THE PLOIDY STATUS OF PREIMPLANTATION HUMAN EMBRYOS

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Time-lapse monitoring of preimplantation embryos not only allows detailed timing of cleavage events but also allows morphological scrutiny at times specific to various developmental events. The aim of this study was to determine whether cytoplasmic texture of the oocyte/zygote is indicative of the ploidy status of resultant embryos.

A total of 194 embryos from 27 treatment cycles were biopsied on Day 3 to allow 24 chromosome CGH screening. All oocytes/embryos were cultured from the time of ICSI insemination to Day 5/6 of development in a time-lapse incubator (Embryoscope). Cytoplasmic regions ($630 \mu\text{m}^2$) were copied from images for each oocyte/embryo at three time points: post sperm injection (Inj), syngamy (Syn) and just prior to cytokinesis (Cyt). Second order co-occurrence matrix texture measurements were recorded for each of the cytoplasmic regions and analysed according to whether subsequent embryos were tested to be chromosomally normal (NAD) or abnormal (Abn).

Overall, 10/11 and 9/11 texture parameters differed significantly with developmental stage ($p < 0.001$) and ploidy status ($p = 0.007$ to $p = 0.038$), respectively. An interaction effect between the two variables was apparent: 1/11 texture parameters at the Inj stage and 0/11 parameters at the Syn stage differed significantly between NAD and Abn embryos. In contrast, 9/11 parameters differed significantly at the Cyt stage ($p = 0.005$ to $p = 0.034$).

These data demonstrate a stage specific difference between NAD and Abn embryos in cytoplasmic texture characteristics. The potential exists for texture parameters to be used in conjunction with developmental timing events to improve selection algorithms for chromosomally normal embryos.

FC095

MEIOTIC SPINDLE LOCATION IN COMBINATION WITH SPINDLE NORMALITY MAY IMPROVE PREGNANCY PREDICTION.

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Background:

Spindle location of oocytes injected using Polarised Light Microscopy (PLM) was compared with pregnancy outcome.

Materials and methods:

Patients (<38yrs) who had ICSI using PLM and with a single embryo transferred (SET) of blastocysts originating from normally spindled oocytes were included. These oocytes were classified into (1) pregnancy oocytes (PO) and (2) non-pregnancy oocytes (NPO). The spindle angle (SA) from the first polar body (1PB) was measured. The incidence of oocytes with SA $\leq 45^\circ$ and $>45^\circ$ was calculated.

Results:

96 oocytes were investigated. The pregnancy generating oocytes (PO) (N=53) had an average SA of 16.7° ; median=15; range= $0 - 55.4^\circ$. The non-pregnancy oocytes (N=43) average SA was 34.4° , median=26.8 and range= $0 - 116.8^\circ$ (T Test $P<0.05$). In the PO, 49/53 (92%) the SA $\leq 45^\circ$ [mean=14, median=13.4 and range $0-44.4$] vs 33/43 in the NPO - 33/43 (77%) [median=26.8; range = $0-44.9$, $P=0.005$]. 4/53(7%) in PO had SA $>45^\circ$ [mean=50.9; median=50.15; range = $48.1-55.4$] compared to NPO - 10/43 (19%) [mean=78.2; median=79.9; range = $47-117.8$ - $P=0.002$]. No pregnancy occurred if SA was $> 55.4^\circ$ despite a normal spindle and a good quality blastocyst (n=8).

Conclusion:

Normally shaped spindles with SA $\leq 45^\circ$ are more likely to implant. When SA exceeds 55° , the failure to produce a pregnancy is indicative of significant structural abnormality in the oocyte. Spindle angle measured by PLM adds another predictive factor in addition to spindle normality in the selection criteria of embryos more likely to implant.

FC096

TECHNOLOGY FOR RESCUE FERTILITY OF AZOOSPERMIC PATIENTS BY TRANSPLANTATION OF SPERMATOGENIAL STEM CELLS

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Manipulation of Spermatogonial Stem Cells (SSCs), includes enrichment/culture of donor SSCs and transplanting the SSCs into the testes of recipient, is an alternative approach for assisted reproductive technology (ART). 3 years ago, we have shown that transplantation of germ cells from a normal Mongolia sheep (donor) into an azoospermic poll dorset sheep (recipient) can rescue its fertility. However in andriatics for rescue fertility of an azoospermic patient, germ cell transplantation may be not so much effective, because SSCs density is low in the germ cells. It is known that purified SSCs are good source for testis transplantation, but in large animals and humans, the source of SSCs is rare. The methodologies for enrichment and proliferation remain developing. It hurdles the research and clinic trail of SSCs in ART. To break through this bottleneck, the purpose of present study is to establish stable and repeatable technology for long-term culture SSCs from large animals. The methods were set up for long-term culture of livestock SSCs. Immunofluorescence double staining were used for analyzing SSC marker-gene expression. Differentiation potency assay was performed in vitro, and in vivo. The results show that the long-term cultured livestock SSCs maintain the features of expressing marker genes, and possess the potency for differentiation into downstream germ cells in vitro or in vivo. All in all, the techniques for long-term culture of livestock SSCs are established, and lay down a foundation for application of SSCs in research and clinic trail in ART.

FC097

MORPHOKINETIC EVALUATION OF EMBRYOS: VARIABLES LINKED TO EMBRYO DEVELOPMENT AND IMPLANTATION

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Introduction

Time-lapse observations provide novel morphokinetic variables, which have been proposed to be utilized for identifying embryos with a high implantation potential. These variables are primarily based on the timing of cell divisions. Utilizing the largest time-lapse database in the world we show that implantation rate can be closely linked to morphokinetics, but that confounding factors need to be taken into account.

Materials & Methods

The importance of different morphokinetic variables was characterized on a database comprising more than 12,000 transferred embryos with known pregnancy information from 29 IVF centers.

Results

Direct division from 1 to 3 cells is linked to very low implantation potential (<9%). Further, 63% of embryos undergoing direct cleavage would not have been detected by conventional static monitoring. Timing from insemination to time of the first cleavage in ICSI cycles was directly linked to high implantation rates, while this variable was not as strong in IVF cycles (n=7929 for the comparison). Later cleavage stages (5-cell, 8-cell, interval between 3 and 5 cell) showed pronounced differences for division timings based on oxygen incubation conditions. Reduced oxygen levels caused a shift of optimal division ranges towards earlier times of division.

Conclusions

The analysis of morphokinetic variables within a large multicentric database revealed universal patterns for all variables investigated. However, optimal developmental timings will vary between clinics according to incubation conditions and clinical practice. Therefore, reported ranges may have to be adjusted for a specific clinic before being applied in the daily routine.

FC098

DISCRIMINATION OF MATERNALLY OR PATERNALLY DERIVED PRONUCLEI BY EPIGENETIC DIVERGENCE IN HUMAN ZYGOTES FERTILIZED ABNORMALLY

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Objective

This study focused on the epigenetic divergence of male and female pronuclei (PN), with respect to differences in DNA methylation status, to establish a method for PN discrimination.

Materials and Methods

Donated normally (n = 3) and abnormally (1PN: n = 9, 3PN: n = 14) fertilized oocytes were used in this study. These oocytes were performed whole mount immunofluorescence staining using the primary antibodies, anti-5mC to detect female PN (fPN) and anti-5hmC to detect male PN (mPN). Alexa Fluor 488 goat anti-mouse IgG and Alexa Fluor 568 goat anti-rabbit IgG were used as secondary antibodies. Images were obtained using an FV1000 confocal microscope.

Results

Our study showed that PNs derived from 3PN zygotes were the result of either polyspermy or failure of the second polar body extrusion. The average mPN diameter in 2PN and 3PN zygotes was slightly larger than that of fPN. A single PN analysis suggested fusion of the maternal and paternal genomes without the formation of a female and male PN.

Conclusions

In this study, we established a novel method to discern PN origin in human zygotes using immunochemical analysis. This system is very useful to reveal the patterns of abnormal fertilization. Further analysis is ongoing to elucidate the mechanisms of abnormal fertilization.

FC099

TRANSCRIPTOME ANALYSIS REVEALS PLACENTA SUBJECTED TO ASSISTED REPRODUCTIVE TECHNOLOGY TREATMENTS MAY HAVE WIDESPREAD GENOMIC EFFECTS

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Altered global gene expression of the placenta subjected to ART manipulation may subsequently affect perinatal outcomes and offspring's health. Variation in ART procedures disturb placental development which alter placentaion and cause the harmful perinatal outcomes and offspring's health. Four chronic villus samples were obtained from multifetal reduction patients under gone in vitro fertilization and embryo transfer due to oviductal factors only in the Center of reproductive Medicine in Third Hospital of Peking University from May 2010 to September 2013. Other four control villus samples were from induced abortion of natural pregnancies. We used a GeneChip Affymetrix HU-U133 Plus 2.0 Array to analysis the genes. Using qRT-PCR we certified microarray data from 10 dysregulated genes. Ten genes were localized precisely in the chronic villus as per immunohistochemistry. About six thousand of these genes were classified into six groups according to critical placental function: immune response; transmembrane transport; trophoblast invasion; steroid metabolism; oxidative stress; cell differentiation; and other functions. Genes involve in trophoblast invasion, such as FGB, MMPs and ITIH2, and those regulating trnsmenbrane transport, such as FABP1, APOB and TF were discerned to be differentially expressed. These genes products were expressed in the placental villus, either in the cytoplasm or in the membrane of syncytiotrophoblastic cells. Our finding may increase the understanding the roles of the differentially expressed genes in ART-treated placenta as well as investigate whether ART manipulations have some effects on placentation.

FC100

TOWARDS AN OBJECTIVE DEFINITION OF POOR RESPONDERS IN ASSISTED REPRODUCTION

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An objective definition of poor response should be based upon the cut-off points below which clinical pregnancy is significantly diminished. Infertile couples treated with ART were studied (n= 6454 cycles). Receiver-operator characteristic (ROC) curves were constructed to compare four predictors of clinical pregnancy. ROC curves revealed that basal AFC was the best predictor of clinical pregnancy (AUC = 0.926) followed by serum AMH (AUC = 0.771), number of oocytes retrieved (AUC = 0.681) and age of the female partner (AUC = 0.367). Cut-off points for AFC were 5, for AMH 2.7 ng/mL, for the number of oocytes 9 and for age of the patient 31 years. Subgroup analysis revealed that cut-off points for age are 30 years, 31 years, 29 years and 31 years for patients undergoing IVF, ICSI, fresh TeSA and frozen TeSA, respectively. Similarly, cut-off points for the number of oocytes retrieved were 6 oocytes, 8 oocytes, 8 oocytes and 7 oocytes for patients undergoing IVF, ICSI, fresh TeSA and frozen TeSA, respectively. It is concluded that the definition of poor responders in ART depends on the treatment modality. Cut-off points based on ROC curves should be used in defining poor response for each modality of ART.

FC101

ENDOMETRIAL BIOPSY PRIOR TO ARTIFICIAL HORMONE REPLACEMENT FROZEN EMBRYO TRANSFER IMPROVES PREGNANCY AND LIVE BIRTH RATE: A RETROSPECTIVE ANALYSIS

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This is a retrospective analyses of artificial FET using hormones in Singapore General Hospital between 2007 to 2011. Artificial cycles for FET consisted of mainly lucrin suppression followed by oral progynova starting from day 2 of menstrual cycle and supplemented cyclogest pessaries two weeks later. Subjects had endometrial biopsy on days 22 of the artificial cycle with an outpatient endometrial sampler the month prior to the cycle of embryo transfer. Controls were patients who did not have prior cycle endometrial biopsy. The dosage of progynova was adjusted when the histology of biopsy was not ideal.

There were 138 consecutive artificial FET cycles analysed between 2007 and 2011. This included 8 cycles that had no lucrin suppression The women's mean age was 33.6 years and a mean of 2.3 embryos were transferred. Endometrial biopsies were conducted prior to actual transfer cycle in 74 cycles and no prior biopsies in 64 cycles.

Pregnancy rate was 34% and livebirth rate was 22% in "biopsied" group and significantly higher compared to 22% and 12.5% in the "non-biopsied group.

Univariate logistic regression excluded confounding variables of age and number of embryos transferred. The only significant factor was the presence of endometrial biopsy ($p=0.004$). The variable of adjustment of progynova dosage was also found to be not significant in the analysis. ($p=0.448$)

The results of this retrospective study appears to show that endometrial injury as a result of biopsy results in doubling of pregnancy and almost tripled the livebirth rate similar to IVF cycles.

FC102

IS FRESH BEST?

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Introduction:

Freezing all viable embryos during an IVF cycle, rather than transferring a fresh embryo, is being adopted clinically for multiple reasons, namely to reduce the incidence of ovarian hyperstimulation syndrome (OHSS) and to improve birth outcomes for both mother and baby. However, there is concern pregnancy outcomes may be compromised utilizing this strategy.

Aim:

To determine whether a freeze only strategy in IVF compromises cumulative pregnancy rates.

Method:

A case controlled study of 181 patients undergoing freeze only cycles was conducted between January 2012 and August 2013 at Monash IVF. Controls were matched for age, number of egg collections, insemination type and usable embryos available for transfer and freezing on Day 5 and 6. Statistical analyses used Fishers exact test and two-tailed t-tests. Control patients were matched to case patients only once.

Results:

There was no difference between Case group and Control for mean maternal age (34.3vs34.5), number of egg collections (2.1vs2.0) and number of usable embryos (4.5vs4.2). The clinical pregnancy rate for the first embryo transfer for the frozen (Case) versus fresh (Control) group was 39.5% and 37.0% respectively. When looking at cumulative pregnancies from a single egg collection, the clinical pregnancy rate for the Case group was 58.6% and the Control group was 57.1%. There was no statistical difference seen between any of these results.

Conclusion:

The pregnancy rates achieved in freeze only cycles were no different to those seen in fresh embryo transfer cycles whether considering pregnancy rate from first embryo transfer or cumulative pregnancy rates.

FC103

DAY 4 AND DAY 5 BIOPSY: A COMPARISON OF ANEUPLOIDY AND PREGNANCY RATES

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Embryo biopsy at both the cleavage (day 3) and expanded blastocyst stage (day 5/6) is routinely used in clinical IVF for PGS/PGD analysis, however biopsy of morula/early blastocyst on day4 is not routinely performed. This study investigated the aneuploidy rates and subsequent pregnancy rates in a day 4 versus day5 biopsy program.

Patients undergoing IVF with PGS screening using 24sure most commonly for repeated implantation failure of good quality embryos were included in this study. Patients were randomly allocated to day4 ($n=53$ patients) or day5 ($n=22$ patients) based on day of oocyte retrieval. All oocytes were fertilised using ICSI and the embryos were vitrified after biopsy and subsequent screening was undertaken. In total 152 day 4 and 58 day 5 embryos were screened with no significant difference in maternal age between the two groups (37.4 ± 0.6 vs. 36.7 ± 1.2 for day 4 and day 5 respectively). There were no significant differences in aneuploidy rate between the two groups (77.6% and 81.0% for day 4 and day 5 respectively) and there was no correlation between grade at biopsy and ploidy status. Interim analysis of pregnancy rates between the two groups (for all ages) were not significantly different (43.5% vs. 35.7% for day 4 and day 5 respectively)

In conclusion day4 biopsy resulted in equivocal aneuploidy and pregnancy rates compared to a day 5 biopsy program. This demonstrates that blastomere biopsy on day4 does not impact on embryo viability and could be considered as a viable option to day 3 or day 5/6 biopsy.

FC104

DOES THE AMH CONCENTRATION PREDICT THE OUTCOME OF IVF IN WOMEN OVER 40?

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Introduction: AMH concentrations predict oocyte number but any association with livebirth rate, is unclear.

Methodology: Retrospective data review. 1439 stimulated ART cycles and 283 frozen embryo transfers were reviewed in women, aged 40-46, with AMH measurement within preceding three years (average 5.2mths). AMH assayed by Beckmann Gen II immunoassay. Cycles were categorised by AMH concentration as: A, >15pmol/L; B, 5-15pmol/L; C, <5pmol/L, and by age as 40, 41, 42, 43-44 and >45yrs. Outcomes studied were: number of oocytes; number of usable embryos; pregnancy /transfer, Livebirths per 100 cycles started.

Results:

Number of oocytes

Age	AMH>15pmol	AMH5-15pmol/L	AMH<5pmol/L
40	12.4 (±0.7)	7.8 (±0.4)	5.6 (0.3)
41	10.9 (±0.9)	8.9 (±0.4)	4.9 (0.3)
42	10.8 (±1.1)	7.4 (±0.4)	4.0 (0.2)
43-44	11.1 (±0.9)	7.2 (±0.4)	4.1 (0.2)

Livebirths per 100 stimulated cycle

Age	AMH>15pmol	AMH5-15pmol/L	AMH<5pmol/L
40	21.6	11.1	7.1
41-42	13.6	7.8	2.3
43-44	12.1	3.2	0.0

Mean number of usable embryos/cycle by AMH category was: A, 2.06 (±0.16); B, 1.63 (±0.05), C, 0.91 (±0.03). Clinical pregnancy per embryo transfer for 40 yearold women was: CategoryA, 25.0%, CategoryB, 15.3%, CategoryC, 10.1% and for 43-44 yearold women was: CategoryA 16.7%, Category B, 7.8%, CategoryC, 3.4%. Multivariate analysis demonstrated a significant association between AMH concentration and livebirth rate (p<0.01). Beyond 45yrs, too few pregnancies to assess predictive effects.

Conclusions: AMH concentration is strongly predictive of oocyte number. A higher AMH concentration is associated with a higher age-related livebirth rate, indicating that, while age remains the principle predictor, AMH also indicates oocyte quality.

FC105

ENDOMETRIAL SECRETIONS; PREDICTING RECEPTIVITY AHEAD OF EMBRYO TRANSFER

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Introduction: This study aimed to identify biomarkers of a receptive endometrium that can predict subsequent pregnancy. Previously uterine lavage samples collected in the early secretory phase of the menstrual cycle (n = 19 fertile/18 infertile) were analysed. Screening of 10 potential biomarkers identified three cytokines for further study; IL8, VEGF and G-CSF. Infertile women showed elevated levels of IL8 and G-CSF, with reduced VEGF, compared to fertile women. Receiver-Operator-Curve analysis produced Area-Under-Curve (AUC) values of 0.599, 0.658 and 0.760, respectively. Multivariate analysis produced an AUC of 0.969.

Methods: A small cohort of samples collected at hCG+2 from women undergoing ART treatment cycles (agonist stimulation) were analysed for the three potential markers. Immunohistochemical analysis of endometrial biopsies for G-CSF receptor was performed.

Results: Women in whom no pregnancy was achieved following embryo transfer in the same cycle, had elevated G-CSF and IL8 levels combined with reduced VEGF, in comparison to those who did become pregnant. Fertile women, showed greater G-CSF receptor immunoreactivity in the glandular epithelium, and lower levels in the stroma compared to the infertile group. The signal was absent from the glandular epithelium of women at hCG+2 in whom ART treatment was unsuccessful.

Conclusions: G-CSF receptor may serve as a useful marker in the initial assessment of endometrial based infertility. Together IL8, VEGF, G-CSF levels in uterine lavage at the time of oocyte pickup, provide a potential tool to predict whether the endometrium will be receptive to implantation when an embryo is transferred a few days later.

FC106

ONE TO NINE YEARS FOLLOW UP OF BABIES BORN AFTER TREATMENT BY AROMATASE INHIBITOR AN OFF LEVEL OVULATION INDUCING AGENT.

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Abstract :

The aim of this study was to evaluate the growth and development of babies born after treatment with aromatase inhibitor (Letrozole) an off level ovulation inducing agent.

This study was conducted in Infertility Care and Research Center (ICRC), Dhaka, Bangladesh over a period of 10 years from 2004-2013. Study population was babies born after treatment with letrozole. Three hundred and ten such babies' milestone were observed periodically from 1-9 years of their age according to CDC criteria and features were compared with milestone fixed for CDC children. Among them 154 are boys and 156 are girls. Average gestational age was 37.94 ± 1.38 weeks and birth weight was 2.94 ± .56 kg for male and 2.85 ± .44 kg for female babies. Twelve (3.87%) babies had minor form of congenital anomalies. Ten (3.23%) babies psychological development was more in comparison to that of CDC babies. Six (1.94%) babies' milestone was delayed in comparison to CDC children. Other babies' growth and mental development are comparable with that of CDC children.

In conclusion it can be said that babies born after treatment with letrozole are safe and their growth and development are comparable with that of babies of normal population according to CDC criteria.

FC107

PREVALENCE, CAUSES AND RISK FACTORS OF INFERTILITY AMONG NEWLY MARRIED COUPLES IN SHANXI PROVINCE, CHINA - A COHORT STUDY

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Objective: To describe the prevalence, causes and risk factors of infertility.

Design: Cohort baseline information was collected by face-to-face interviews with women who went to seek premarital health checkups from Nov 2009 to Jul 2012 in two maternal and child health care centers in a rural area of northern China. Information on time to pregnancy, reasons for not becoming pregnant, and the causes of infertility were collected during the follow-up.

Results: A total of 2035 couples were followed-up for at least 1 year. The 12-month infertility rate was 13.6%, and the 24-month rate was 8.4%. Infertility was due to a female factor alone in 36% of all cases with both wife and husband had infertility examination, 15% of cases were due to a male factor, 23% of couples had an infertility diagnosis in both partners, and 15% of couples had no demonstrable cause in either partner. The main causes of infertility for female were ovulation disorders, fallopian tube problems, and polycystic ovary. The main cause of infertility for male was sperm quality problems. Women living in rural village, having higher BMI, whose husband was a farmer or worker, and whose husband was older than 26 years at marriage were more likely to be infertile.

Conclusions: Infertility in northern rural China is higher than most Asia countries but lower than some European countries. Anovulation infertility is the commonest types. Environmental and occupational factors were identified to be associated with the risk of infertility, but further studies are needed.

FC108

OUTCOME OF LIVE BIRTH IN MASSIVE ADENOMYOSIS AFTER RESECTION

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Background. Mainly characterized by severe dysmenorrhea, pelvic pain, and infertility, adenomyosis usually managed by hysterectomy. However, for women wished to conserve their fertility, resection of the adenomyosis, by conservative or by Osada procedure, is the current chosen method for managing adenomyosis.

Objective. To investigate the live birth outcome of women with massive adenomyosis treated with resection procedure.

Methods. Retrospective study at Dr. Ciptomangunkusumo General Teaching Hospital, Jakarta, Indonesia was done by observational review on pregnancy and live birth outcome of the adenomyosis patient treated with resection between January 2010 and December 2012.

Results. From 49 patients undergone Adenomyosis resection, 37 of them are having severe adenomyosis based on the AFS, while the other 12 are having moderate ones. Twenty-three patients diagnosed with only Adenomyosis, 24 with adenomyosis and endometriosis cysts, and 2 with adenomyosis and endometrial polyp. From 39 women who complained of having infertility, 8 patients (20.51%) conceived naturally without any other intervention with 2 patients already delivered their babies by Caesarean Section without any sign of uterine rupture. The other 4 patients are still with the ongoing pregnancy, while the other 2 had miscarriage on the 3rd month of pregnancy. There are also 2 patients (5.13%) who are pregnant assisted by IVF and already delivered their babies by C-Section.

Conclusion. Resection of adenomyosis is a promising procedure for treating adenomyosis in women with infertility with more than 20% chance of having the pregnancy and delivery afterwards.

Keywords. Adenomyosis, adenomyosis resection, dysmenorrhea, infertility, live birth outcome

FC109

EFFICACY OF LOW MOLECULAR WEIGHTED HEPARIN (LMWH) TREATMENT IN UNEXPLAINED RECURRENT SPONTANEOUS ABORTION (RSA) PATIENTS WITH DECREASED UTERINE BLOOD FLOW

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Objective

To investigate whether low-molecular-weighted-heparin (LMWH) is effective in improve uterine blood flow and obstetric outcomes in RSA patients with decreased uterine blood flow.

Material and Methods

The prospective study population included 68 pregnant women (between 5-7 weeks gestation) with history of RSA and control were 35 healthy pregnant women (between 5-7 weeks gestation) without a history of infertility and RSA. Uterine color-pulsed transvaginal Doppler ultrasound was performed to evaluate uterine radial artery resistance index (RI) in RSA patients and healthy pregnant women. In RSA patients with decreased uterine radial artery blood flow (RI > 0.5), LMWH was administrated subcutaneously (range 40-80mg/day). Uterine radial artery blood flow was reassessed after one week following LMWH treatment. And we compared the viable pregnancy outcome (above 25 weeks gestation) between RSA patients with LMWH treatment and healthy normal control.

Result

Uterine radial artery RI was significantly higher in patients with RSA than healthy controls (0.59 ± 0.17 vs. 0.54 ± 0.12 , $p=0.010$). Uterine radial artery RI was decreased significantly with LMWH treatment in RSA patients (pretreatment RI: 0.62 ± 0.13 vs. post-treatment RI: 0.52 ± 0.14 , $p=0.000$). Viable pregnancy outcome was similar between LMWH treatment group and healthy control group (74.1% vs 80.0%, $p=0.519$).

Conclusion

LMWH seemed to be effective treatment to improve viable pregnancy rate in unexplained RSA patients with decreased uterine blood flow.

FC110

STUDY OF STRUCTURE AND ULTRASTRUCTURE, APOPTOTIC INCIDENCE AND FOLLICLE MATURATIONAL GENES EXPRESSION OF VITRIFIED-WARMED WHOLE RAT OVARY AFTER AUTOTRANSPLANTATION WITH UNILATERAL AND BILATERAL GONADECTOMY

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The aim of this study was to evaluate structure of ovary and ultrastructure of the initial follicle, apoptotic incidence rate and follicle maturational genes expression of vitrified ovary following autotransplantation.

The five groups of rat ovaries were divided as follows: control (nonVitrified nonTransplanted), nVTnG (nonVitrified Transplanted nonGonadectomy), VTnG, nVTG, VTG. These groups were evaluated by standard histology techniques and detailed semi thin study, EM and immunohistochemistry. Expressions of follicle maturational genes were studied by Real-time PCR.

In nVTG and VTG (gonadectomized) groups, the percentage of follicular maturation and ultrastructure of transplanted ovaries were in better condition as compared to other experimental groups especial non-gonadectomized (nVTnG and VTnG) groups. In gonadectomized groups, however, the incidence of apoptosis in primordial and antral follicles was higher, whereas the incidence of apoptosis in primary and preantral follicles was lower than the VTnG and nVTnG groups. Also the rate of TGF- β expression was lower in non-vitrified groups than the both of the vitrified groups. Expression of BMP-15 did not show any significant differences between all control and transplanted groups and the last GDF-9 was expressed at the highest level in control and secondly in non-vitrified groups comparing with vitrified experimental groups.

The results of this study showed that a combination of EG + DMSO and sucrose was more suited for follicular preservation, particularly at the initial stage could relatively restore ovarian follicular development after vitrification and autotransplantation. Also after gonadectomy the rate of follicular survivability and reservation was better than non-gonadectomized groups.

FC111

COMPARISON OF CLINICAL RESULTS FOLLOWING SPLIT CYCLES OF IN VITRO FERTILIZATION (IVF) AND INTRACYTOPLASMIC SPERM INJECTION (ICSI)

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Objective: To examine the effectiveness of split cycles in cases of no severe male factors, especially in the first cycles.

Materials and methods: Subjects were 258 patients (265 cycles) from January 2012 to February 2013. Retrieved oocytes were divided into two groups: IVF (1077 oocytes) and ICSI (1019 oocytes), and results were compared. Single embryo transfer was performed in all cycles.

Results: Rates of fertilization, D3 good quality embryo, blastulation, pregnancy and miscarriage in IVF and ICSI cycles were [65.6% (706/1077) vs. 78.1% (793/1019): p=0.01], [45.4% (279/614) vs. 48.6% (322/683): p=0.55], [49.8% (279/614) vs. 40.8% (252/619): p=0.04], [34.0% (16/47) vs. 24.1% (14/58): p=0.04], [18.8% (3/47) vs. 35.7% (5/58): p=0.65] respectively. In vitrified-warmed embryo transfer cycles, the rates of pregnancy and miscarriage were [50.0% (54/108) vs. 45.0% (54/120): p=0.65] and [18.5% (10/54) vs. 20.4% (11/54): p=0.41].

In mild male factor cases, the rates of fertilization and blastulation were [69.4% (93/134) vs. 89.8% (114/127): p=0.17] and [55.4% (36/65) vs. 39.2% (29/74): p=0.14].

Conclusions: Split cycles can be a choice in case of no severe male factor at the first treatment. In mild male factor, there was no significant difference in fertilization rate. There was also no significant difference between embryos derived from IVF and ICSI. Nevertheless, embryos derived from IVF had higher blastulation and pregnancy rates in fresh embryo transfer cycles. Also, in IVF cycles, 3.4% (9/265) were not fertilized and polyspermy occurred in 8.2% (88/1077) of cases, therefore, IVF should be performed in more oocytes of split cycles.

FC112

BIRTH WEIGHT FOLLOWING VITRIFIED-WARMED EMBRYO TRANSFER WAS HIGHER THAN THOSE IN NON-ART AND FRESH EMBRYO TRANSFER.

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Objectives: Effects of vitrification on the birth weight by the gestational age were assessed.

Materials & Methods: A total of 2,549 cycles (non-ART: 807; fresh embryo transfer (fresh-ET): 707; vitrified-warmed (cryo)-ET: 1,035) in which singleton pregnancy was confirmed between 2004 and 2011 was included in the analysis. We examined the birth weight and the rate of premature births in non-ART, fresh-ET, and cryo-ET by the gestational age. The data were compared using PLSD test following ANOVA. The model included the main effects of sex of baby, mode of delivery, and body height and BMI of mother.

Results: There was no difference in the rate of premature births among 3 groups. Although, the birth weight and the caesarean section rate in the cryo-ET were significantly higher than others ($P < 0.05$), the birth weight in the cryo-ET was significantly higher than others in the case of natural delivery. Moreover, the birth weight in the cryo-ET was significantly higher than others at 38-40 weeks of gestation ($P < 0.05$). Similarly, the birth weight in the cryo-ET was also significantly higher than others despite baby's sex, mother's height, and BMI.

Conclusion: The birth weight in the cryo-ET was heavy despite no difference between non-ART and fresh-ET partly because of its higher caesarean section rate. However, we had the same result even in the case of delivery by natural means. Furthermore, baby's sex, mother's height, and BMI didn't affect the result. Our data suggested that the vitrification procedure would affect the birth weight.

FC113

A PROSPECTIVE RANDOMIZED CONTROLLED TRIAL STUDYING THE NEED FOR HOSPITALISATION OF SURROGATES FOR A BETTER PREGNANCY OUTCOME

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Objective: With increased cross-border reproductive care, gestational surrogacy has become a viable option for many infertile patients. India has been a favoured location for pursuing such arrangements in a cost-effective manner, with supportive and liberal laws. The ultimate aim of subfertile patients travelling to foreign countries is to achieve a healthy pregnancy with maximum convenience and minimum cost.

Design: A prospective randomized controlled study in which 64 surrogate pregnancies at our centre were studied in detail, between 2006 and June 2013

Materials and Methods: This prospective randomized controlled study divided the surrogate mothers into two groups. A first group who had a supervised pregnancy within the hospital, and a second group of surrogates who stayed at their homes with their family. Surrogates were also grouped according to their age- 25 to 33 year age versus 33 to 38 year.

Results: The surrogates who were below 33 years of age and not hospitalised during pregnancy had more pregnancy-related complications like missed abortion, threatened abortion, IUGR, preterm delivery and LBW babies. Surrogates supervised in hospital during pregnancy had low complication rate. The difference in complication rate was statistically significant (7.4% for hospitalised-supervised surrogates versus 23.1% for non-supervised ones; p-value less than 0.05). Surrogates in the older age-group were more compliant with prolonged hospital stay and treatment.

Conclusions: Planned hospitalisation of surrogates leads to a better pregnancy outcome. Most surrogate mothers enjoy the freedom from domestic duties that hospitalisation offers, with a planned and nutritious diet. All these factors lead to better neonatal outcome.

FC114

CC-HMG combined with Growth Hormone in Ministimulation Protocol may Improve Clinical Outcome in Poor Ovarian Responders

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Objective: To investigate the role of highly purified Human Menopausal Gonadotrophin (HMG) and Growth Hormone (GH) in mini-stimulation protocol to improve outcome in poor ovarian responders

Design: Retrospective cohort study.

Patient(s): Women who attended Morula IVF Jakarta clinic in between January 2012-June 2013.

Methods: All patients were given clomiphene citrate 150 mg from day 3 to day 7 of menstrual cycle followed with 150IU hpHMG daily from day 8 until ovulation trigger. Two groups were observed where GH was given and the other arm did not received GH. In GH group, GH 8IU were given from day 1 of stimulation until stimulation was stopped. GnRH antagonist was used to suppress ovulation.

Outcome: The primary outcome was ongoing pregnancy rate per patient and secondary outcome was the embryo quality per patient.

Result: Among 51 eligible women (29 patients with GH and 22 patients without GH. No difference was observed in number of oocytes retrieved and number of embryos transferred in both group (2.21 versus 2.64) ; (1.24 versus 1.68) in group GH versus without GH, respectively. Total clinical pregnancy rate was 17,6 %. In patients older than 40 years old GH showed 4 times likelihood in producing top quality embryos (44.8% vs 13.6% (OR 3.6; p 0.05).

Conclusion(s): CC-HMG regimen in ministimulation protocol is an effective option in poor responders. Additional GH in ministimulation program provided higher number of top quality embryos in women older than 40 years old, although there were no difference in clinical or ongoing pregnancy rate.

FC115

ENDOSCOPIC EMBRYO TRANSFER AND IMPLANTATION(HEED AND SEED) - IMPROVING SUCCESS AND DECREASING RISKS

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INTRODUCTION

There has been little change in embryo transfer technique in past 30 years, even though it is a major bottleneck to the success of IVF procedure. Here we would like to share our experience with endoscopic embryo transfer and implantation.

MATERIALS AND METHODS

In the first group of patients HEED was done on day 2/3, and in the second group SEED was done on day 5/6 after oocyte retrieval. The hysteroscope was a 3mm flexible scope made by Storz, El Segundo, CA, USA and the KAM's catheter was made by IVF Scientific, Beverly Hills, CA USA.

RESULTS

There were a total of 59 patient starts with a total of 32 pregnancies. There were 14(24%) live births, 6 biochemicals, and 10 spontaneous pregnancies. The 2 ectopic pregnancies were confined to the HEED group.

DISCUSSION

HEED and SEED are objective and reliable techniques that assure correct placement of the embryo(s). Ectopic pregnancies from IVF will be minimized by using lower transfer volumes of 5 µl and visually confirmed positional placement of embryos 2 cm away from the uterine cornu and are eliminated with SEED. Patients with failed IVF, 'Implantation Failure' or at risk for ectopic pregnancy would particularly benefit from SEED. The use of endoscopic embryo transfers would greatly alleviate patient anxiety as they can see the transfer process on the monitor, and would decrease cost to the patient as they decrease the number of attempts at using IVF in achieving a successful targeted singleton pregnancy.

FC116

DUAL EMBRYO TRANSFER: BENEFITS AND RISKS.

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Introduction: Embryo transfer is a critical point of IVF procedure which may influence upon treatment outcome. The clinical pregnancy rate after IVF/ICSI is higher after embryo transfer at blastocyste stage. But in some cases prolonged cultivation leads to transfer cancellation because all embryos stop to develop after day 3 or 4. In order to avoid transfer cancellation and increase pregnancy rate we introduced in our practice the dual embryo transfer: first embryo transfer on day 3 and second embryo transfer on day 5.

Objective: to compare IVF outcomes after embryo transfer on day 3, on day 5 and after dual embryo transfer.

Material&methods: Retrospective study of 1133 consecutive IVF/ICSI cycles: group I – 545 cycles with embryo transfer on day 3, group II – 355 cycles with embryo transfer on day 5, group III – 233 cycles with dual embryo transfers.

Results: In group I, 545 embryo transfers resulted in 173 clinical pregnancies (31,7%), of these 7 were ectopic (4%) and 21% were multiple. In group II, 355 embryo transfers resulted in 154 pregnancies (43,4%), of these 5 were tubal (3,3%) and 23% were multiple. In group III, 233 embryo transfers resulted in 104 clinical pregnancies (44,6%), of these 4 were ectopic (3,9%) and 36% were multiple.

Conclusions: Dual embryo transfer is more effective than embryo transfer on day 3. Despite of the same number of transferred embryos the multiple pregnancy rate was also higher after dual embryo transfer.

FC117

QUALITY OF SIBLING CLEAVAGE-STAGE EMBRYOS GENERATED IN SYNTHETIC PROTEIN-FREE AND CONVENTIONAL PROTEIN-CONTAINING COMMERCIAL EMBRYO CULTURE MEDIA

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Objective: The objective of this study is to determine the impact of presence or absence of donor serum proteins in embryo culture medium (ECM) on quality of cleavage stage sibling embryos generated and on resultant pregnancies.

Materials and methods:

Analysis of laboratory data was undertaken on the quality of sibling embryos and pregnancies generated in the synthetic chemically defined protein-free medium (PFM) from Cellcura, Norway and the control Sage®Medium (SM) from Cooper Surgical, USA. The parameters investigated were: fertilization rate (FR), zygote arrest rate (ZAR), mean blastomere number (MBN), mean embryo grade (MEG). [Embryos graded 4 = excellent, 3=good, 2=average, 1=poor].

Results: Day 2 sibling embryo MBN and MEG for PFM and SM was 4.4 vs 3.9, $p=0.4691$; 3.2 vs 3.0, $p=0.0405$ respectively. On day 3 the mean blastomere numbers and embryo grades for PFM and SM were 6.6 vs 6.1, $p=0.2247$; 3.2 vs 2.9, $p=0.4318$, respectively. The clinical pregnancy rate (SAC& FHB) for the PFM was 43.3% (13/30) for all age (28 to 42yrs) groups combined.

Discussion and conclusion: Sibling embryo study in this small investigation in a newly established fertility center comparing outcome of sibling human embryo development in PFM and SM suggest the efficacy of both media to be similar in generating quality embryos. The pregnancy rate for the PFM appears good. The PFM is the only synthetic ECM available that is also certified Halal. It eliminates risk of disease transmission, is safe, completely chemically defined, offering the most advanced lot to lot functional consistency in ART.

FC118

INCIDENCE OF MOSAICISM IN CHROMOSOMALLY ABNORMAL HUMAN EMBRYOS DETECTED BY ARRAY-CGH.

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Introduction

Human embryo has many chromosome abnormalities which are aneuploidy such as trisomy and monosomy. Chromosomal abnormalities are took place during not only gametogenesis but also embryonic development. Mosaicism takes place when chromosome abnormalities occurred in embryonic development. The objective of the present study was to determine how much percentage of chromosomally abnormal embryos indicated by array-based comparative genomic hybridization (array-CGH) is mosaicism assessed by Fluorescence in situ hybridization (FISH) in order to offer precise diagnosis for PGD.

Materials and methods

Six good quality surplus chromosomally abnormal embryos vitrified on Day-3 were used after informed consent. Every blastomere from each embryo was analyzed by FISH to reveal mosaicism.

Result

Two of the six embryos were mosaic while four were not. Monosomy 5 embryo by array-CGH consisted of eight blastomeres. Seven of the eight had single signal and the other one had double signals by FISH of chromosome 5. Trisomy 13 embryo by array-CGH consisted of nine blastomeres. Eight had triple signals, but the other one had double signals by FISH of chromosome 13.

Conclusions

Mosaicism takes place one third (33%) of chromosomally abnormal Day 3 embryos. The data of the present study was lower than the other data reported. PGD of Day3 embryos should be reliable only if array-CGH is used from the present study.

FC119

SINGLE VITRIFIED EMBRYO TRANSFER OF BLASTOCYSTS CULTURED IN DEFINED MEDIA WITH RECOMBINANT HUMAN ALBUMIN: A RANDOMIZED TRIAL

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Human serum albumin (HSA), a common protein source for ART, may cause biological variation/disease transmission. Recombinant human albumin (rechA) could reduce these risks; however, the literature on rechA is limited. Here, we evaluated the efficacy of rechA-containing defined media for blastocyst culture. This study included patients undergoing elective cryopreservation of all embryos. Ninety-five patients received IVF/ICSI treatment between July 2012 and October 2012, which resulted in ≥ 6 2PN embryos 18 h after insemination. They were randomized into 2 groups for embryo culture in G1/G2 containing 5 mg/mL HSA (commercial media, group A) or 0.5 mg/mL rechA (group B). The rechA concentration was as per our previous study (ASRM 2012). After culture, good-quality embryos were vitrified by day 6. We evaluated the data for patients with vitrified blastocysts in groups A ($n = 34$) and B ($n = 32$). Patient age (years) was similar for groups A and B (35.7 [3.2] vs. 35.7 [3.8]). A total of 110 and 65 blastocysts were vitrified in groups A and B, respectively. Groups A and B underwent 47 and 37 cycles of single vitrified-warmed blastocyst transfer, respectively. All vitrified blastocysts survived after warming. Clinical PR/ET was 22/47 (46.8%) for group A and 26/37 (70.3%) for group B ($P = 0.05$). Ongoing PR/ET was 20/47 (42.6%) for group A and 19/37 (51.4%) for group B ($P = 0.51$). Embryo culture in the rechA-containing defined media yielded good-quality blastocysts, resulting in high PR after single cryopreserved ET. These results could help develop defined ART systems.

FC120

INFLUENCE OF BLASTOCYST PRE-FREEZE MORPHOLOGY & RE-EXPANSION POST WARMING ON PREGNANCY RATES IN VITRIFICATION CYCLES.

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OBJECTIVE: To study the effect of blastocyst pre-freeze morphology & re-expansion post warming on pregnancy rates in vitrification cycles.

Design: Retrospective study.

Setting: Tertiary care infertility centre.

Method: 100 vitrification cycles with single blastocyst transfers were analyzed between January 2012 to June 2013 using the Cryoloop. Based on their pre freeze morphology, blastocysts were grouped based on the following parameters:

- 1) Expansion state of the blastocyst - 2 groups: expanding & early hatching,
- 2) Inner cell mass (ICM): - 3 groups: Good, Average & Poor
- 3) Trophectoderm (TE): 3 groups: Good, Average & Poor.

Post warming, blastocyst's were categorized into 2 groups based on the re-expansion and non expansion of their blastocoele cavity.

Main outcome measure: Ongoing pregnancy rates

Results: Transfer of 100 vitrified warmed blastocysts in the study resulted in an implantation rate of 35% and ongoing pregnancy rate of 33%. 100% survival rate was achieved, with blastocoele re-expansion occurring in 88% of the cycles, resulting in a higher ongoing pregnancy rate in the re-expansion group (34.09% vs 25%; $P = 0.746$) as compared to the non-expanded blastocyst group, although statistically non significant. No difference in ongoing pregnancy rates was observed between blastocysts with good, average or poor ICM (33.3%, 30.3%, 50% respectively) or, with good, average or poor TE (34.8%, 29.8%, 42.9% respectively). The ongoing pregnancy rates for expanding & early hatching blastocysts were 38.98% & 24.39% respectively, which were not statistically significant.

Conclusion: Our analysis demonstrated that the pre-freeze morphology and re-expansion of a blastocyst post-warming has no influence on clinical outcome in vitrification cycles.

FC121

OOCYTE AND GRANULOSA-CUMULUS CELLS INTERACTIONS: LH RECEPTOR MRNA EXPRESSION ON THE OOCYTE QUALITY

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Background: Luteinizing hormone (LH) exerts its actions through its receptor (LHR), which is mainly expressed in theca cells and to a lesser extent in oocytes, granulosa and cumulus cells. The aim of the present study was the investigation of a possible correlation between LHR gene expression in granulosa-cumulus cells and ovarian response as well as assisted reproductive technology outcome.

Method: Human granulosa-cumulus cells obtained from 26 women undergoing oocyte retrieval for in vitro fertilization. RNA extraction and cDNA preparation was followed by LHR gene expression investigation through real-time polymerase chain reaction (PCR). The oocyte were classified into three groups of fertility ratio (low: <30%; moderate: 30%-60%; and high >70%). Furthermore, the gene expressions of LHR were compared by the fertility ratio of oocyte. **Result:** LHR mRNA expression were detected in all of granulocyte-cumulus tissue samples. LHR PCR products revealed increase in mRNA expression in the granulosa-cumulus with the low group of fertility ratio. LHR mRNA expression was lower in the high fertility ratio group than in the moderate group and low group of fertility ratio. Concerning LHR expression in granulosa-cumulus cells, a significant negative association was observed between LHR gene expressions with the fertility ratio.

Conclusion: LH play some role as regulators in granulosa-oocyte ovarian tissue, and that the present study provides a step towards a new role of LHR gene expression profiling as a biomarker candidate in the prediction of ovarian response.

Keyword: Granulosa-cumulus cells, LH Receptor, oocyte quality

FC122

BENEFICIAL EFFECTS OF RESVERATROL ON BOVINE OOCYTE MATURATION AND SUBSEQUENT EMBRYONIC DEVELOPMENT AFTER IVF

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Objective: To analyze the potential beneficial effects and mechanisms of action of resveratrol on the maturation of bovine oocytes that were incubated in different concentrations of resveratrol (0.1, 1.0 or 10µM) as germinal vesicle (GV) oocytes.

Design: *In vitro* prospective study.

Setting: University research laboratory

Patient(s): Animal models for human studies.

Intervention(s): *In vitro* culture in the presence of various concentrations of the antioxidant resveratrol.

Main Outcome Measures: The parameters of hormone levels, oocyte nuclear maturation, cumulus expansion, levels of intracellular glutathione and reactive oxygen species, embryonic cleavage, blastocyst formation, gene expression associated with mature bovine oocytes and cumulus cells and the level of sirtuin 1 gene expression were detected.

Results: Resveratrol significantly increased progesterone secretion and decreased estradiol-17β secretion by cumulus cells. The elevated levels of progesterone activated the *Mos*/MEK/p42 MAPK cascade in the oocytes. At a concentration of 1.0 µM, resveratrol significantly improved cumulus expansion, polar body formation, the (hatched)blastocyst rate and the mean number of cells/blastocyst. Meanwhile, resveratrol significantly reduced the level of ROS, increased the level of GSH. For the first time, the expression of the sirtuin 1 gene was identified in granulosa cells, cumulus cells, oocytes and blastocysts. Further studies revealed that resveratrol promoted sirtuin 1 gene expression.

Conclusion(s): Resveratrol promoted bovine oocyte maturation and subsequent post-IVF embryonic development by inducing progesterone secretion and antioxidant effect, *probably in a manner dependent on Sirtuin 1*.

MINI ORAL / POSTER ABSTRACTS



MINI ORAL ABSTRACTS

M001

NATURAL KILLER CELL ANALYSIS: FROM BLOOD, UTERUS, BOTH OR NOT AT ALL?

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Introduction

Since 2005 robust assays for natural killer cell analysis in both uterine biopsy (uNK) and peripheral blood (bNK) samples have produced one of the largest databases of its kind in the world. This study aimed to assess the level of correlation (if any) between the tests, and to try to determine which may be of greater clinical use.

Methods

A database was created for women presenting with infertility or miscarriages and tested for uNK and bNK cells. Clinico-pathological comparisons were made using previously published thresholds for 'high' NK cell levels, focusing on markers CD56+, CD57+ and CD69+.

Results

323 women (mean age 37.1) had uterine biopsies, of whom 98 (30.7%) had 'high' CD56+ levels. uNK CD56+ and CD57+ levels were not correlated. 249 women also had blood tests, with 26 women (10.4%) having 'high' CD56+ levels. bNK CD56+ and CD69+ tests were strongly correlated ($p < 0.0001$). There was no correlation between uNK and bNK CD56+ levels overall. However, in the 26 women with high bNK levels, 12 (46%) had high uNK CD56+ levels. And in 15 women with specifically high activated bNK levels (CD56+CD69+), 11 (73%) had high uNK levels – significantly higher than those with normal bNK levels (25%) ($p = 0.0002$).

Conclusions

High levels of activated bNK cells (CD56+CD69+) do strongly predict high uNK cell levels. It is proposed that the blood test could be used as a screening test for randomised trials of reproductive immune therapy.

M002

EPIDEMIOLOGICAL ANALYSIS OF THE RISK FACTORS AND THE PERIOD FOR ONSET OF OVARIAN INSUFFICIENCY AFTER SURGERY

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Objective: To determine the risk factors to induce ovarian insufficiency after surgical removal of benign ovarian cysts, and the period until the onset of ovarian insufficiency after the surgery.

Methods: Data were retrospectively obtained from ovarian insufficiency patients under fertility treatment in our hospital. Several epidemiological parameters cause of ovarian insufficiency after the ovarian surgery were analyzed based on their medical records.

Results: In the ovarian insufficiency patients ($n = 835$), 75 patients (9.0%) received ovarian surgery before onset of ovarian insufficiency. Among the 75 patients, 9 patients (12.0 %) received oophorectomy, whereas 66 patients (88.0 %) received cystectomy. Although 12 patients (16.0 %) received multiple surgeries (bilateral cystectomy), single operation was performed in other patients. In 57 patients (76.0 %) of patients, the surgical indication was the treatment of endometriotic cysts. The mean age of the patients at the time of surgery was 27.8 (± 5.5) years of age, and the mean period until the onset of ovarian insufficiency was 5.8 (± 3.8) years.

Conclusions: Our data indicated that both oophorectomy and cystectomy could induce ovarian insufficiency. In addition to the multiple ovarian surgery, single intervention could also induce ovarian dysfunction. Patients received cystectomy of endometrial cysts are likely to cause ovarian insufficiency. Because it takes long period to become ovarian insufficiency after ovarian surgery, it is important to inform the possibility of future ovarian dysfunction to the patients and monitor their ovarian reserve for sufficient time after the surgery.

M003

CLEAVAGE KINETICS ANALYSIS IS HELPFUL TO PREDICT THE BLASTOCYST TRACEABILITY

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Selecting the most competent embryo is a very important procedure to perform elective single embryo transfer (e-SET) and blastocyst vitrification preservation in ART. Methods: From June 2012 to January 2013, 45 patients who received IVF-ICSI in our clinic were studied. A retrospective time-lapse monitoring analysis of morphokinetic parameters for 79 Day 5 blastocysts (Group 1) were compared to 29 Day 6 blastocysts (Group 2) and 116 embryos with arrested development from the same pool of Group 1 (Group 3). Results: The results of morphokinetic parameters (reaching time) are 2PB extrusion time (3.1 ± 0.8 h, 3.0 ± 0.8 h, 3.1 ± 1.2 h post-ICSI), 2PN observation time (8.8 ± 2.2 h, 9.1 ± 2.6 h, 9.5 ± 3.0 h), syngamy (22.0 ± 3.2 h, 23.3 ± 3.2 h, 25.0 ± 4.6 h), two cell arrival time (24.9 ± 2.7 h, 25.7 ± 3.1 h, 28.0 ± 2.5 h), four cell arrival time (37.0 ± 4.6 h, 38.6 ± 3.9 h, 42.0 ± 8.8 h), 8cell arrival time (50.3 ± 4.4 h, 57.1 ± 8.6 h, 57.4 ± 9.9 h) and compaction arrival time (82.2 ± 7.6 h, 94.9 ± 13.2 h, 92.8 ± 15.2 h). Four cell arrival time from syngamy was significantly faster in Group 1 and 2 than in Group 3. Eight cell arrival time was significantly faster in Group 1 than in Group 2. Compaction arrival time in Group 1 is significantly faster than in Group 2 or 3. The length of time to four cell arrival time resulted in blastocyst formation. The length of time after eight cell arrival time resulted in a delay of blastocyst formation in Group 2. Conclusions: Morphokinetic parameters are helpful to make appropriate decisions for the disposition of each embryo. These data suggest that the time-lapse monitoring system can be used to link the timing of early embryo cell divisions and the probability of subsequent development of embryos into blastocysts.

M004

DEVELOPMENT OF A 5-POINT EMBRYOSCOPE ALGORITHM PREDICTIVE OF GOOD BLASTOCYST GRADE.

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Aim

Static embryo assessment at distinct time-points is conventionally used to select the best embryo for transfer, with critical stages between observations going unnoticed. This study compared good and poor quality blastocysts to establish if specific developmental time-points can be used to improve the process of embryo selection.

Methods

Oocytes were cultured between ICSI (Day 0) and Day 5 in an EmbryoScope using Vitrolife G5 media. Observations were made every 7 minutes and sperm entry timings were calculated for each egg. Expanded and hatching D5 blastocysts with ICM and trophectoderm grade AA or BA (good) were compared against C-grade (poor) ICM or trophectoderm blastocysts.

Results

Blastocysts at the same developmental stage with POOR grading showed an increased time to reach any growth stage compared with GOOD quality blastocysts. The five most significant predictors of D5 blastocyst quality are tabulated:

	S2	t8	tM	tSB	tEB
Grade:	Time 3cell-4cell	Time to 8cells	Time to Morula	Time to Blastulation	Time to Expanded Blastocyst
GOOD	28min (+/-22)	54.4hrs (+/-6.8)	89.3hrs (+/-6.2)	96.7hrs (+/-4.8)	108.8hrs (+/-5.0)
POOR	57 min (+/-60)	60.0 hrs (+/-9.9)	98.9 hrs (+/-10.7)	110.6 hrs (+/-10.7)	129.5 hrs (+/-12.1)
Pvalue	p=0.0013	p=0.0013	p<0.0001	p<0.0001	p<0.0001
GOOD Timing visible:	3AM D2	2PM D2-3AM D3	1AM D4-2PM D4	10AM D4-7PM D4	10PM D4-8AM D5

Conclusion

Two markers (s2 and t8) can be used to guide embryo selection for laboratories performing D3 culture. For D5 culture, a more advanced algorithm containing all five time-points can be used. A prospective randomized study using these developmental markers is underway.

M005

MORPHOLOGICAL DIFFERENCES BETWEEN EMBRYOS TREATED WITH IN VITRO MATURATION AND ROUTINE IVF DETECTED WITH TIME LAPSE MONITORING INCUBATOR

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Background: In preparation for intracytoplasmic sperm injection (ICSI), in vitro maturation (IVM) is hypothesized to be required for harvesting mature enough oocyte.

Objectives: The aim of this study is to evaluate the morphological differences of embryos treated with IVM in comparison with those which are treated with routine *in vitro* fertilization (IVF) method using time lapse monitoring incubator.

Method: This study is a preliminary study. After ovum pick-up, the immature oocyte was cultured in vitro under optimized condition to obtain M-II oocyte. ICSI was then conducted to both the oocyte which is treated with IVM and the one which is treated by routine IVF. Changes in the embryo's morphology, such as the time of cleavage and PN breakdown (PNB), were observed using time lapse monitoring incubator every 20 minutes until the third day.

Result: Embryo treated with routine IVF shows better development compared with the one treated with IVM. The PNB time of IVF and IVM embryos are similar at 23 hours after ICSI, with the cleavage time of 22 hours in IVF-treated embryo and 31 hours in IVM-treated embryo. IVF-treated embryos can reach 8 cells cleavage at 46 hours while IVM-treated embryos only reached 2 cells cleavage. Fragmentation is shown on both embryo, however the fragment in IVF-treated embryo is missing after 12 hours.

Conclusion: Embryos which is treated with IVM cleavage later than embryo treated with routine IVF prove in time lapse monitoring incubator.

Keyword: embryo, in vitro maturation, time lapse

M006

LIVE BIRTH RATES OF EARLY STAGE BLASTOCYSTS CULTURED OVERNIGHT FOLLOWING VITRIFICATION AND WARMING.

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The purpose of this study was to evaluate the influence of overnight culture on vitrified and warmed early stage blastocysts on survival, pregnancy and live birth rates. A retrospective analysis of warmed day 5 single blastocyst cycles was carried out between April 2010 and November 2011. The data examined was restricted to early blastocysts that had been vitrified on day 5 of culture and subsequently warmed approximately 16 - 24 hours prior embryo transfer. Two other groups were examined for comparative purposes: early stage blastocysts that were transferred in a fresh cycle and expanded or hatching blastocysts vitrified day 6. These were warmed on the morning of day 5 and transferred same day.

Of the 108 blastocysts warmed, 107 survived (99%) and were cultured overnight. Subsequent fetal heart rate (FHR) was 28.3% (30/106) and live birth rate (LBR) of 23.5% (25/106) per transfer. In comparison, fresh embryo transfers of similar embryos gave a FHR of 29.2% (71/243) and LBR of 25.9% (63/243). Day 6 embryos that were warmed and transferred on day 5 (88% survival) resulted in a FHR of 30.3% (40/132) and LBR of 25.8% (34/132). There were no statistical differences between the 3 groups.

Since all vitrified early stage blastocysts are routinely warmed the day prior transfer, it is difficult to select and compare a similar cohort of embryos cultured within 4 hours of transfer. The results show that warmed early stage blastocysts are not compromised with overnight culture and survival rates were better than those for day 6.

M007

COMPARISON OF PREGNANCY RATES BETWEEN IVM CYCLES WITH AND WITHOUT MATURE OOCYTES (MII) AT COLLECTION

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Introduction: It has been claimed that IVM cycles with MII at collection have lower success rate than without MII.

Objective: To compare the IVM outcomes between cycles with and without MII at collection.

Methods: A retrospective cohort study was conducted on 207 IVM cycles in PCOS patients at IVFAS, An Sinh hospital from July 2012 to June 2013. Patients had FSH (100 IU/day for 3 days) and hCG priming (10,000 IU). Oocyte maturity was assessed at collection. Patients were divided into 2 groups based on the presence of MII at collection. Embryo transfer was done on day 2. Transferred embryos derived from GV oocytes at collection, which matured through IVM process.

Results: A total of 207 IVM cycles were recruited, in which 63 cycles had and 144 cycles had not MII at collection. Cycles with MII at collection had significantly higher maturation rate and number of extra embryos for freezing compared to cycles without MII (66.4% vs 55.1%, 1.25 ± 2.2 vs 0.60 ± 1.3 , $p < 0.05$, respectively). No difference was found in the rates of fertilization, top-quality embryo, clinical pregnancy and implantation between two groups.

Conclusion: IVM outcomes are not compromised by the presence of MII oocyte at collection.

M008

RETROSPECTIVE ANALYSIS OF SERUM ANTI-MULLERIAN HORMONE LEVEL IN A COHORT OF PATIENTS WITH RECURRENT MISCARRIAGE

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Aim

To investigate the relationship between serum AMH level and recurrent miscarriage, and whether patients with recurrent miscarriage have a lower serum AMH level comparing to the general population based on the validated model published in the literature.

Methodology/ study design

A retrospective analysis of the AMH level in 71 patients with recurrent miscarriage; who presented to IVF Australia between 2010 and 2012. The AMH level was compared with the reference range derived from 4000 fertility screening patients. Medical records were reviewed to identify patient characteristics.

Results

71 patients were included in the study. The mean age is 35.8 years (25 to 45 years). The mean level of AMH is 15.8 pmol/L and median level is 10.4 pmol/L (0.5 pmol/L- 79 pmol/L).

23 patients were identified to have a low AMH level. The average age was 34.6 years. The mean AMH level was 3.8 pmol/L.

In these 23 cases, only five patients had identifiable factors which could possibly account for recurrent miscarriage; one patient had moderate titer of anticardiolipin antibodies, two patients had ultrasound evidence of uterine polyp or fibroids, one patient had type 2 diabetes, PCOS and endometriosis, one patient had hyperthyroidism and her partner had abnormal sperm DNA fragmentation index.

Conclusion

A higher proportion of patients with recurrent miscarriage had a lower serum AMH level compared with the age matched fertility screening population. The majority of patients with low AMH level had no identifiable causes for recurrent miscarriage. A low serum AMH may be a causal factor for recurrent miscarriage.

M009

OXIDANT AND ANTIOXIDANT STATUS IN NON- OBESE WOMEN WITH AND WITHOUT POLYCYSTIC OVARY SYNDROME

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Introduction: Polycystic ovary syndrome (PCOS) is the most common endocrinopathy in women. Obesity is a relatively common feature of PCOS and its relationship with oxidative stress has already been identified. The present study is to investigate the relationship of PCOS per se and the oxidant and anti-oxidant status.

Method & Material: A case control study was carried out on 29 non-obese patients with polycystic ovary syndrome (BMI ≤ 25) and 100 non-PCOS women matched for BMI, all aged 17 -40 years. Total oxidant status (TOS) and total antioxidant status (TAS) levels were compared in the groups using SPSS V17 for statistical analysis. Pvalue<0.05 was considered as significant.

Findings: The logarithm of the mean of Malondialdehyde (MDA) as an indicator of TOS was 1.29 ± 0.5 in PCOS patients and 1.25 ± 0.3 in the control group (Pvalue: 0.7). On the other hand, log of the mean of total antioxidant status (TAS) was 0.48 ± 0.07 in PCOS versus 0.48 ± 0.08 in the control group (Pvalue: 0.73). Mean of MDA revealed positive correlation with TSH, $r = 0.5$ ($p = 0.005$), while mean of TAS had negative correlation with FSH, $r = -0.7$ ($p = 0.002$).

Conclusion: Based on the above results, TAS and MDA levels are not different in non obese women with and without PCOS and increased oxidative stress in PCOS patients can be secondary to obesity rather than PCOS per se.

Key words: Polycystic ovary syndrome, oxidative stress, antioxidant status, non- obese.

M010

REGULATION OF SERUM AMH IN PATIENTS WITH PCOS: RELEVANCE OF INSULIN RESISTANCE INDICES

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Objective: Elevation of serum anti-Müllerian hormone (AMH) levels among polycystic ovary syndrome (PCOS) patients has been reported. However, the regulatory factors of AMH in PCOS remain unknown. We examined correlations between AMH values and various indices of insulin resistance in PCOS women.

Methods: 40 infertile Japanese women compatible with 2003 Rotterdam criteria for PCOS were recruited under informed consent. The subjects underwent 75g oral glucose tolerance test (75gGTT, sampling at 0, 60, 120 minutes), and 2-step ELISA for AMH (sensitivity 0.7 pmol/L, inter & intra-assay C.V. :12.3%, 14.2%, respectively). $p < 0.05$ was considered as statistical significance.

Results: Average AMH level of the subjects (Age: 31.9 ± 3.9 years, mean \pm S.D.) was 50.4 ± 31.1 pmol/L, which was higher than normo-ovulatory age-matched women (16.7 ± 10.5 pmol/L, $n = 16$). AMH levels had significant positive correlation with total ovarian volume by ultrasound (18.2 ± 11.4 cc), but not with BMI (21.1 ± 3.0 kg/m²), waist-hip ratio (0.85 ± 0.06), and levels of serum LH (5.5 ± 3.6 IU/L), total testosterone (67.3 ± 32.8 ng/dL). Although fasting glucose levels (83.1 ± 4.9 mg/dL) were positively correlated with total ovarian volume ($r = 0.41$), AMH levels had no significant correlation with fasting insulin (4.3 ± 2.3 IU/L), fasting glucose/insulin ratio (22.8 ± 8.4), HOMA-IR (0.90 ± 0.51), and the sum of insulin levels (98.6 ± 42.1 IU/L) during 75gGTT.

Conclusions: Increased serum AMH level of PCOS and its positive correlation with total ovarian volume implies that determination of serum AMH level would aid in confirming diagnosis of the common ovulatory disorder. Absence of relationship between AMH and various clinical indices of insulin resistance may suggest alternative regulatory factors of AMH gene expression in the follicular compartment.

M011

METABOLIC RISKS OF THE LEAN PCOS WOMAN.

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Objective : To compare the lean (BMI <25) and the non-lean (BMI ≥25) PCOS women with respect to insulin resistance , metabolic syndrome and dyslipidimias.

Study population: 207 lean PCOS and 314 non-lean PCOS women.

Methods : This was a cross-sectional analysis of 528 PCOS women enrolled for a PCOS phenotypes study in a private practice setup. PCOS was diagnosed based on the Rotterdam(2003) criterias. Fasting blood sugar, fasting insulin, HOMA-

IR, serum lipids, thyroid function tests, prolactin, FG score, systolic and diastolic blood pressure was compared between the two groups. Insulin resistance was assessed based on multiple HOMA-IR cut-offs. Prevalence of metabolic syndrome (NHLI ATP III 2005) was checked.

Results: Lean PCOS women had statistically significantly lower levels compared to non-lean PCOS women for the following parameters: W/H ($P < 0.0001$), Fasting insulin ($P = 0.0006$), HOMA-IR ($P < 0.0001$), total cholesterol ($P = 0.03$), triglycerides ($P = 0.0068$), Vldl ($P = 0.008$), systolic blood pressure ($P < 0.0007$), diastolic blood pressure ($P = 0.0002$).

Using a HOMA-IR cut-off of ≥ 3.8 , ≥ 3.5 and ≥ 2.5 , the prevalence of insulin resistance was 7.79%, 7.79% and 8.44% respectively in lean PCOS and 34.89%, 35.67% and 58.63% respectively in non-lean PCOS women.

Metabolic syndrome was present in 3.25% of lean against 25.6% of non-lean PCOS women.

Conclusion: Diabetes mellitus, insulin resistance and metabolic abnormalities are all significantly lower in lean PCOS women. However, in view of abnormal waist circumference and W/H ratio with normal BMI and a very high prevalence of diabetes in the Indian population, we would recommend testing all lean PCOS women for glucose intolerance and metabolic syndrome.

M012

IMPAIRED FASTING GLUCOSE (IFG) AND GLUCOSE TOLERANCE TEST (IGT) OCCUR ACROSS THE SPECTRUM OF BMI AND PHENOTYPES OF PCOS PATIENTS

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Background Although not a part of diagnostic criteria for PCOS, insulin resistance, IFG and IGT are considered to play a key role in etiology of PCOS. Early detection in this population could reduce the incidence of diabetes mellitus and other metabolic diseases. The aim of this study was to compare fasting glucose level (FPG), insulin resistance (HOMA-IR) and Glucose tolerance test (GTT) across the spectrum of BMI and phenotype of PCOS patients.

Method This is a cross sectional study of 108 patients diagnosed with PCOS based on Rotterdam criteria. Patients were interviewed and examined for menstrual history, clinical manifestations of hyperandrogenism, and ovarian ultrasound assessments. OGTT, FPG levels and insulin resistance index (HOMA-IR) were examined for those meeting criteria. Kruskal wallis and Mann whitney test were used to analyze the difference of blood glucose measurements and insulin resistance in different PCOS phenotypes.

Result Normal glucose metabolism was present in majority of cases 83(76.1%), mean age: 29.03 ± 4.1 years; BMI: 26.5 ± 5.0 kg/m². Diabetes was diagnosed in four patients (3.7%). FPG, OGTT and insulin resistance were significantly higher in obese patients. The most common PCOS phenotype was PCO and non-ovulatory (56%). However, the median of GTT (115(68-312mg/dl) and HOMA-IR (2.6 (0.4-13.9)) were significantly higher in the phenotypes of non-ovulatory, PCO and hyperandrogenism compared to PCO and non-ovulatory (102(69-203mg/dl) and (1.8(0.4-9.3)). IFG and IGT were significantly higher in obese patients.

Conclusion: IGT, insulin resistance and IFG occur across the spectrum of BMI and PCOS phenotypes.

Keywords PCOS phenotypes, IFG, IGT, BMI

M013

THE EFFICACY OF EMBRYO CULTURE IN MEDIUM SUPPLEMENTED WITH GRANULOCYTE MACROPHAGE-COLONY STIMULATING FACTOR (GM-CSF) ON OUTCOMES IN REPEATED IMPLANTATION FAILURE (RIF) PATIENTS

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Introduction: GM-CSF is a lymphohematopoietic growth factor that plays a major role in fetal development. It has been found that RIF patients have a significant reduction of serum GM-CSF.

Objective: To explore the efficacy of embryo culture in GM-CSF supplemented medium on outcomes in RIF patients.

Methods: A case series was conducted at An Sinh hospital from August 2012 to June 2013 on 60 RIF patients. RIF patient was defined as who failed ≥ 3 embryo transfers (ET) with ≥ 6 top-quality embryos transferred. After ICSI, the injected oocytes and later on, the cleaving embryos were cultured in GM-CSF supplemented medium at 37°C, 7%CO₂, 5%O₂ until day of ET

(day 2). Outcome measures included number of top-quality embryos, rates of fertilization, implantation, clinical and ongoing pregnancy.

Results: Mean patients' age was 35.77 ± 4.61 and mean previous failed transfers was 4.20 ± 1.25 . The mean number of top-quality embryos was 1.70 ± 1.98 . The fertilization, implantation, clinical and ongoing pregnancy rates were 77.5%, 14.1%, 33.3%, and 30%, respectively.

Conclusions: Embryo culture in GM-CSF supplemented medium improves the implantation and ongoing pregnancy rates in RIF patients. Further RCTs are awaited to confirm the benefits of GM-CSF supplemented medium in RIF patients.

M014

ENDOMETRIOSIS: HOW MUCH DOES IT COST AUSTRALIA?

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In Australia, it is estimated that 560,000 women in their reproductive years are affected by endometriosis. Endometriosis is frequently associated with decreased health-related, Quality of Life (HRQoL), increased chronic pelvic pain and increased pain-associated distress and disability. Decreased HRQoL and increased pain is associated with rising health costs to society.

This study explored the cost of endometriosis to Australia. Australian population and healthcare data were obtained from the Australian Bureau of Statistics website and the Australian Institute of Health and Welfare website, specifically: Australian Hospital Statistics 2010-2011 and Chronic Disease Statistics. These data were mined to identify the Australian-equivalent, cost-of-illness, outcome measures that have been published for European and North American endometriosis populations.

This study suggests that endometriosis costs Australian society \$7.7 billion annually. Approximately two thirds of these costs are attributed to lost productivity, with the remainder – about \$2.5 billion - being direct healthcare costs. These costs may be directly borne by the healthcare budget, government subsidies, private health insurance, employers and individuals or households. Indirect costs are often met by support organisations, familial and social networks. Other non-healthcare costs may include social withdrawal, diminished life opportunities, psychological disturbances such as low mood and physical inactivity.

Detailing the burden of endometriosis from a cost perspective is an important tool in optimising funding allocation and government support. In Australia, available data strongly suggest that endometriosis adds a significant 'silent' burden to the nation's budget. A cost-of-illness study in Australia is a key area for future research.

M015

DEVELOPMENT OF INFANTS DERIVED FROM METAPHASE II OOCYTES WITH SMOOTH ENDOPLASMIC RETICULUM CLUSTERS (SERC)

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Objective: To examine the impact of smooth endoplasmic reticulum clusters (SERC) in human metaphase II (MII) oocytes on embryo development, clinical pregnancy rate, and infant malformation rate.

Design and method(s): A total of 2158 patients (3578 cycles) underwent ICSI treatment at Kyono ART Clinic from January 2007 to December 2011. Prior to ICSI, MII oocytes were classified according to the presence of SERC. In 212 patients (252 cycles) at least one oocyte in the same cohort of retrieved oocytes had SERC. In these patients, a total of 1557 mature oocytes were divided into two groups: Group A (322 oocytes) was SERC positive while Group B (1235 oocytes) was negative. The remaining 1946 patients (3326 cycles) had no SERC in their 14000 mature oocytes; these were classified as Group C.

Result(s): Rates of fertilization, blastulation, implantation, clinical pregnancy and miscarriage following fresh embryo transfer were [64.0% (206/322), 40.3% (56/139), 7.1 (3/42), 8.3% (3/36) and 33.3% (1/3)], [73.1% (903/1235), 43.5% (324/745), 19.3% (28/145), 23.7% (27/114) and 33.3% (9/27)] and [70.3% (9848/14000), 42.6% (2933/6880), 16.6% (422/2547), 17.8% (407/2147) and 30.2% (123/407)] in Groups A, B, and C, respectively. In Group A, 3 patients became pregnant (1 patient had a miscarriage) after transfer of fresh embryos, while 12 patients conceived by single vitrified-warmed blastocyst transfer. Consequently, 14 healthy babies were delivered with no malformations in Group A.

Conclusion(s): The presence of SERC in MII oocytes had no impact on pre- or post-implantation development as well as neonatal outcome including early growth of infants.

M016

INVESTIGATING THE POTENTIAL USE OF CHLAMYDIAL SEROLOGY DURING THE INITIAL INFERTILITY INVESTIGATION

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Introduction: *Chlamydia trachomatis* infection results in reproductive tract damage in some women causing infertility due to tubal damage or occlusion. Currently, this aetiology for female infertility is diagnosed using laparoscopy and hysteroscopy methods. In some cases this technique will not detect more subtle damage caused by chlamydial infection to the fallopian tube and hence, may miss some chlamydial female factor infertility. Therefore, a serological based assay may be a less invasive yet effective first line of investigation. This project aimed to evaluate a new serological diagnostic assay (QUT Prototype Peptide 11 Assay) for chlamydial infertility and compared to the only commercially available serological diagnostic assay for identification of chlamydial tubal factor infertility (MEDAC, cHSP60 +MOMP IgG ELISA).

Method: A case:control approach was used and participants were recruited with either tubal factor infertility (case), and infertile with patent tubes (control). To date serum from four participants with known tubal factor infertility and 23 infertile with patent tubes have been analysed.

Results: Neither assay successfully detected any of the four participants who were in the tubal factor infertility cohort. There are other causes known of tubal infertility which may be the aetiology for these four cases. The MEDAC infertility assay was positive for two of the 23 participants from the infertile with patent tubes cohort.

Conclusion: Ultimately the research team aims to conduct a larger screening study to evaluation the effectiveness of across the board implementation of chlamydial serology compared to laparoscopy and hysteroscopy methods during the initial infertility investigation.

M017

BODY MASS INDEX (BMI) AS A PREDICTIVE FACTOR FOR MENARCHE IN INDONESIAN FEMALE STUDENTS

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Introduction: Menarche is the first menstrual cycle that indicates puberty in young girls. There is no exact age of menarche, depending on various factors such as biological, nutritional, and socioeconomic factors. Body mass index (BMI) is one of the factors that show nutritional status in human. This study hypothesized that BMI affects the onset of menstrual cycle in female students.

Objective: This study aims to determine whether BMI has a correlation with age of menarche in Indonesian female students.

Methods: A cross-sectional study was conducted and data were obtained from 404 questionnaires that have been completed by middle and high school's female students in Jakarta, Indonesia. There were ±15 items in a questionnaire, including demographic data, menarche, menstrual cycle, dysmenorrheal pattern, treatment, and other menstrual symptoms. Statistical analysis for the study was conducted using nonparametric test of SPSS program.

Results: From 404 data, the sample's distributions were not normal. The average number of BMI was 19.33 (12.50-35.84), with 11-13 years old (84%) as a median age of menarche in Jakarta. There was a negative correlation between BMI and menarche.

Conclusion: This study shows there was a negative correlation between BMI and menarche, which indicates that the higher BMI gets, the earlier onset of menarche. However, other factor such as biological, genetical, and environmental factor could give influence to the onset of menstrual cycle.

Keywords: BMI, menarche, menstrual cycle, endocrinology

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P001

COMPARISON OF HYALURONAN BINDING ASSAY SCORE OF SPERMATOZOA USING THE SWIM-UP TECHNIQUE AND DENSITY GRADIENT CENTRIFUGATION

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Purpose To compare the hyaluronan binding assay (HBA) scores of sperm preparation using two different methods, the swim-up technique and density gradient centrifugation (DGC).

Methods This experimental study used semen specimens from 54 volunteer subjects with normal semen analysis according to the 2010 World Health Organization criteria. Each semen specimen was split into two portions: one was prepared using the swim-up method and the other the DGC method. The prepared sperm were counted in the sperm-HBA slide to determine bound and unbound motile sperm. The HBA scores between the two methods were compared using matched analysis.

Results The HBA scores by either preparation method were >80%. There was no statistically significant difference in HBA scores following the swim-up preparation [median 97%, interquartile range (IQR) 94, 98] and density gradient concentration [96%, IQR 95, 98] ($P=0.96$). Ten of the 54 specimens received the same HBA scores following the two methods and none differed by more than $\pm 7\%$.

Conclusions Both preparation methods gave high HBA scores with no apparent difference in the proportions between methods.

Key words hyaluronan binding, spermatozoa, swim-up, density gradient centrifugation

P002

DISTINCT EFFECTS OF NON-INVASIVE AND INVASIVE ASSISTED REPRODUCTION TECHNOLOGIES ON THE ADAPTABILITY TO THE ENVIRONMENT TEMPERATURE DYNAMICS CHANGES OF THE OFFSPRING

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ART, including IVF, ICSI and PGD, has potential effects on the brain and nervous system. Regarding the important role of the brain and nervous system in the process of adjusting the adaptability to the environment dynamic changes, herein we developed a mouse model of ART and treated the mice under higher temperature condition to study the effects of environmental temperature change on ART offspring. The mice in the IVF and control groups had an increased heart rate and blood pressure; however, it could quickly restored to normal levels. The mice in the PGD and ICSI groups required more time to recover. To discuss the possible mechanism, the gene expression of brain tissues were detected by mRNA transcriptomics. More differentiated expressed genes were screened in the PGD and ICSI groups. Stress-related genes were screened using a bioinformatics method and identified with RT-PCR. The expression of HSP27 and HSP70 had dramatically changed, which suggested potential effects on the MAPK signal pathway. By identifying the gene and protein expression in the MAPK -P38 and MAPK -JNK pathways, we found that the former was impaired in IVF mice, but the latter was damaged in ICSI and PGD mice. In conclusion, the present study suggests reduced adaptability to the environment dynamics changes for ART offspring. The molecular mechanisms are different between non-invasive and invasive ART. Invasive ART should be limited in the clinic until before safety concerns are clarified.

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P003

ADVANCED LIFE SUPPORT IN OBSTETRICS (ALSO) WORKSHOP ON YEAR-ONE POST GRADUATE STUDENTS (PGS) - A PRE AND POST WORKSHOP ANALYSIS

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The study is being conducted in Aga Khan University Hospital in obstetrics and Gynaecology department on year one residents admitted in the year 2013. The ALSO Provider Course is a part of on-going training for Obgyn faculty, residents and labour room staff along with participants from Family Medicine and Emergency Medicine. The aim of this study is to identify the changes, if any, in the knowledge and skills taught to manage emergency obstetrics situation, after attending ALSO Provider Course. A total of 10 candidates have been taken as post graduate students in the year 2013 and all will be included in the study. Each candidate is asked to fill in survey questionnaire, give feedback on each facilitator's performance

on a five point grade scale, and appear in a pre and post written exam and viva. So far the data for 7 participants have been collected. There was a great progress observed in the pre and post results for both written and viva exam. All the candidates scored higher in the post written and viva. There was a minimum 23% increase in total Post exam marks and a maximum of 47% increase at p-value .018. The results are subjected to change.

P004

IMPACT OF ASPARTATE AND SERINE ON MOUSE EMBRYO DEVELOPMENT IN VITRO

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Objective

The objective is to determine the optimal concentrations, tolerance levels and effects of aspartate and serine on mouse embryo development in vitro.

Methods

Earle's Basal salt solution was supplemented with 5mg/ml of bovine serum albumin, antibiotics penicillin/ streptomycin sulfate, and pyruvate/ lactate. Treatments consisted of culture media supplemented with graded amounts (0, 250,500,750 and 1000mM) of aspartate and serine. Quakenbush sibling mice zygotes were randomly apportioned to individual treatments. The numbers of replicates were 5 and 4 respectively. The end point was the development of day 6 (expanded, hatching, hatched) embryos.

Results

The proportion of day 6 embryos developed were only marginally improved 29.1, 34.2, 41.8, 40.3, 32.9% with the addition of graded amounts (0, 250,500,750 and 1000mM respectively) of aspartate with maximum improvement seen with treatment supplemented with 500mM but this was not significant. Whereas all treatments supplemented with serine resulted in insignificant increases in embryonic death and was harmful at all concentrations tested (%dead: c =51.6, 250mM=58.3%, 500mM=53.3%, 750mM=75% and 1000mM=61.7% respectively; $r = 0.6271$; $p=0.2575$).

Conclusion

Aspartate do not appear to be of benefit while serine in general appear detrimental but these effects were insignificant. These responses were obtained under the confounding influences of bovine serum albumin that is present in conventional embryo culture media (ECM). The present findings showed the addition of these two amino acids in conventional ECM containing serum proteins may not be useful and may even be harmful. This suggests the practice of supplementing ECM with aspartate and serine must be re-examined.

P005

COMPARISON OF SUCCESSFUL PREGNANCY RATE WITH REFLUX INCIDENCE BETWEEN FLEXIBLE AND RIGID CATHETER IN INTRA UTERINE INSEMINATION

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Objective : To evaluate comparison of succesful pregnancy rate with reflux incidence between flexible and rigid catheter in intra uterine insemination.

Methods : The study design was an analytic observational with cohort study design to evaluate the comparison of succesful pregnancy rate with reflux incidence between flexible and rigid catheter in intra uterine insemination were performed at the Halim Fertility Centre and Stella Maris – Women and Children Hospital in Medan from July until September 2013.

Result : There is no difference between the general characteristics of 30 cases of rigid catheters and flexible catheter, both in terms of female partner age, duration of infertility, and type of infertility. From insemination cycle's characteristics , we found no significant difference in the number of follicles, endometrial thickness and number of sperm used in both groups. Reflux incidence was higher in rigid catheter group 8 (26.7 %) compare to only one in flexible catheter ($p < 0.05$). The Clinical pregnancy rate was similar between reflux group and non reflux group ($p > 0.05$).

Conclusion : There is no difference in the success rate of intra-uterine insemination with reflux And non reflux group using either rigid or flexible catheter.

Key Words : Intra uterine insemination, Rigid Catheter, Flexible catheter, Reflux.

P006

INFLUENCE OF ANTI-MÜLLERIAN HORMONE LEVELS ON OOCYTE YIELD AND PREGNANCY RATE

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Background: Anti-Müllerian hormone (AMH) is a member of the transforming growth factor superfamily. In the fertility setting AMH is used for ovarian reserve assessment. Ambiguity remains as to whether AMH is clinically beneficial in predicting assisted reproductive technology outcomes. With current research divided, we investigated the influence AMH levels have on the number of oocytes retrieved and pregnancy rate.

Design: Retrospective cohort study

Method: De-identified patient records from 710 females aged 21-47 were collected at Concept Fertility Centre (2010-2013). AMH levels were log-transformed. All analyses were adjusted for maternal age. One-way between groups analysis of covariance generated adjusted means. Linear regression investigated oocyte retrieval and AMH levels.

Patients: All presenting females with an AMH level were included.

Results: Results demonstrated a positive correlation between AMH levels and the number of oocytes retrieved ($B=3.77$, $t=9.81$, $P<0.0005$). The association between AMH levels and the number of oocytes retrieved was examined further by dividing patients into two groups according to their response to ovarian stimulation (poor responders = 0-4 oocytes and normal or high responders >4 oocytes retrieved). Significantly higher levels of log-AMH were found in the normal or high responders ($M=0.92$, $SE=0.025$) compared to the poor responders ($M=0.63$, $SE=0.025$, $F_{1,707}=68.570$, $P<0.0005$). Significantly higher log-AMH levels were also found in patients who were clinically pregnant at 8 weeks ($M=0.97$, $SE=0.053$) compared to those who were not ($M=0.75$, $SE=0.019$, $F_{1,707}=15.0$, $P<0.0005$).

Conclusion: In this cohort study, results indicate AMH levels influence oocyte yield and pregnancy rate, demonstrating a place for AMH in the fertility setting.

P007

EFFECT OF TOTAL INTRAVENOUS ANESTHESIA AND REGIONAL ANESTHESIA ON IVF OUTCOMES

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Effect of Total Intravenous Anesthesia and Regional Anesthesia on IVF Outcomes

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Objective: To compare the influence of different anesthetic methods on reproductive data. Primary outcome was clinical pregnancy rate per embryo transfer (CPR). Secondary outcome measures were live birth rate per embryo transfer (LBR), fertilization rate (FR), oocyte maturation rate (MR) and oocyte degeneration rate (DR).

Methods: We conducted a retrospective study involving 87 patients who underwent oocyte retrieval procedure between 2009 and 2011. Total intravenous anesthesia (TIVA) was done using Propofol and Midazolam and regional anesthesia (RA) was achieved Lidocaine injection into the subarachnoid space. The types of anesthesia used was dependent on choice of anesthetist in charge.

Results: Our results showed a statistically significant difference in CPR between patients using TIVA and RA (41% vs 69%; $p < 0.05$).

LBR was also higher in RA group compared to TIVA (54% vs 33%).

FR and MR were similar between two groups (68% vs 63%; 76% vs 88%, respectively).

DR was also significant lower in RA group (5.4% vs 8.4%, $p=0.002$).

Conclusion: Although the number of cases reviewed was small, we found significant difference in CPR and DR, we have since started using RA for all OPU cases. Our aim is to limit the accumulation of anesthetic drugs in the follicular fluid to prevent its detrimental effect on oocyte.

P008

LH RECEPTOR GENE EXPRESSION IN GRANULOSA CELLS IN POOR RESPONDER WOMEN UNDERGOING IVF TREATMENT

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Introduction

In infertility women that undergoing stimulation protocol, only a subset of them seem to benefit from addition of LH to stimulation protocol. Such as in poor responder patient. To know the differences response, genetic studies needed for helping us to individualize the therapeutic approach. Luteinizing hormone (LH) play an important role in folliculogenesis and ovulation. The action of LH is mediated by binding to its receptor (LHR) which is located on the cell membrane. Infertility and abnormal steroid can be monitored by the expression based on LHR mRNA.

Objective

The aim of the study is to compared LHR gene expression in poor responder and normal patient with male factor undergoing IVF treatment.

Method

Granulosa and cumulus cells were collected from 10 patients undergoing IVF treatment on the day egg collection, Five patients with male factor and 5 patient with poor responder. The RNA were extracted than reverse transcribed to cDNA and followed by LHR gene expression using Real Time PCR.

Result

The LHR gene expression in poor responder patient has no differences with male factor patient. A significant negative association was observed between LHR gene expression from both patient ($P < 0.05$).

Conclusion

LHR gene is mostly expressed in Granulosa cell in both poor responder women and male factor patient undergoing ICSI treatment. No significant differences between those patients.

Keyword

LHR, poor responder, granulosa cells

P009

RAPAMYCIN MODULATES MATERNAL MRNA DEGRADATION AND IMPROVES EMBRYO VIABILITY IN CLONED MOUSE EMBRYOS

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Although somatic cell nuclear transfer (SCNT) has been successful in many species, the overall rate of success has remained quite low. We found that autophagy, an essential process for preimplantation development, shows a low expression level in 2-cell stage SCNT when compared with fertilized counterparts. The present study investigates embryo viability and gene expression of SCNT mouse preimplantation embryos in different concentrations of rapamycin (an activator of autophagy). Our results indicated that 1 nM rapamycin treatment for 10 h following oocyte activation significantly enhanced the development of SCNT embryos to the blastocyst stage when compared with the control (59.3% vs. 41.5%, $P < 0.05$). Furthermore, the treatment of rapamycin improved total cell number and also decreased the apoptotic index, stress granules in blastocyst stage embryos. Rapamycin treatment resulted in low mRNA expression levels of four maternal genes, plat, gdf9, mos and h1foo in 2-cell and 4-cell stage embryos were closer to the fertilized group rather than untreatment group. These results indicate that rapamycin-treatment can accelerate mRNA degradation of maternal genes and improve the efficiency of preimplantation development of cloning embryos.

P010

EFFECTS OF DIMETHYLSULPHOXIDE ON ASYMMETRIC DIVISION AND CYTOKINESIS IN MOUSE OOCYTES

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Dimethylsulphoxide (DMSO) is used extensively as a cryoprotectant and as one of the most common solvents for the in vivo administration of several water-insoluble substances. DMSO also has various biological and pharmacological activities. However, the effect of DMSO in mouse oocyte meiotic maturation remains unknown. The present study investigated that asymmetric division and cortical reorganization were disrupted by exposure of oocytes to 3% DMSO at 37 °C. In DMSO treatment oocytes, we observed symmetric division during in vitro maturation. Moreover, ethylene glycol and glycerol treatment also causes failure of asymmetric division. Oocyte polarization was not established due to the failure of an actin cap formation and spindle migration. These features were among the main causes of abnormal symmetric division. However, an analysis of the mRNA expression levels of genes related to asymmetric division revealed that no significant difference in expression of factors was observed between the DMSO-treatment group and the control group. Furthermore, after two sperm heads injection to the oocytes of 2-cell-like M2 respectively, the embryo had ability to extrude the second polar body respectively and start to cleavage. The big polar body M2 also had development ability after parthenogenetic activation. We conclude that DMSO probably affects asymmetric division and cytokinesis completion through failure of actin polymerization, spindle organization.

P011

PREDICTIVE FACTORS FOR PREGNANCY IN IVF: AN ANALYSIS OF 1107 CYCLES

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Objective: To determine predictive factors for pregnancy after IVF.

Method: The subject of this study were one thousand one hundred and seven IVF cycles in 1083 couples who underwent controlled ovarian hyperstimulation in IVF cycles between January 2005 and June 2013. Categorical variables were compared using Chi Square test and continuous variables were analyzed using Independent t-test, $p < 0.05$ was considered statistically significant. Multivariate logistic regression analysis was done to test correlations between clinical variables and the occurrence of pregnancy.

Results: The women's age significantly influenced pregnancy rate since women under 35 years old has the best chance for pregnancy (58.3%). Endometrial thickness on the day of hCG administration also significantly influenced pregnancy in IVF ($p < 0.004$) because 58% of pregnancy occurred if endometrial thickness ≥ 11 mm. Number of embryo > 8 cells after fertilization also has significance on pregnancy. Logistic regression done revealed the couple with best chance of pregnancy can be described as follows: women with endometrial thickness ≥ 11 mm, number of embryo > 8 cells after fertilization more than 2 cells and age under 35 years old.

Conclusion: This study enabled the characterization of many prognostic factors for pregnancy.

Keywords: in vitro fertilization, clinical pregnancy, age, embryo, endometrial thickness

P012

TOWARDS A "ZERO" OHSS RATE

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Introduction:

Ovarian hyper stimulation syndrome (OHSS) is a life-threatening complication of ART. The incidence of moderate/severe OHSS is reported to be as high as 3-8% of IVF cycles. The best treatment remains prevention, and a dopamine agonist has been shown to have benefit in women at high risk of OHSS. Despite the good quality evidence to support its role, it is not routinely used in all Australian fertility units.

Aim:

To review the effect of adding a dopamine agonist to a standard prevention protocol to reduce OHSS rates in a single fertility centre, and compare to the nationally reported rates of OHSS.

Method:

Retrospective cohort analysis of patients in a single fertility centre (2012-13) who had evidence of hyperstimulation and were prescribed cabergoline as part of standard management for prevention of OHSS. Parametric and non-parametric influence was used for interpretation.

Results:

Demographics reported in Table 1. Mean oocytes collected was 20 (IQR=18-23). Mean oestrogen pre-trigger was 13663 pmol/L (IQR=8019-19128pmol/L). 0% cases of moderate/severe OHSS in patients with >20 oocytes collected. ANZARD 2013 reported 4.3% hospitalization for OHSS with >20 oocytes collected.

Conclusion

Preliminary analysis suggests that the addition of cabergoline to standard practice may further reduce moderate/severe OHSS rates. Increasing the sample size to allow multivariate analysis is ongoing.

Table 1: Demographics of patients treated with cabergoline for OHSS (2012-13)

n=32	Mean	IQR
Age (years)	33.00	30.00-36.50
BMI (kg/m ²)	22.96	20.54-23.50
AMH (pmol/L)	32.08	12.20-44.10
Antral follicle count (AFC)	15.61	11.00-20.00
Length of subfertility (months)	20.06	9.00-29.25

P013

CHANGES IN SPERM MEMBRANE STRUCTURE AND FUNCTION INDUCED BY EXOGENOUS PHOSPHOLIPIDS

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Egg yolk is widely used as cryoprotectant for sperm cells. Recently phospholipids isolated from egg yolk (1) and other sources (2) are also used, even commercially available phospholipids are being tried (3). In this study we investigated whether exogenous phospholipids can manipulate sperm membrane structure and function.

Phospholipids, such as phosphatidyl choline, phosphatidyl ethanolamine, phosphatidyl serine, phosphatidyl inositol, sphingomyeline were isolated and purified from chicken egg yolk and liver of sting ray by chromatographic methods. They were integrated with sperm cells in liposomal form. Change in sperm lipid composition was studied by thin layer and gas chromatography. Membrane fluidity was calculated from fluorescence intensity studies, measured by spectrofluorimeter using diphenylhexatriene probe (4). Sperm motility was studied microscopically.

Changes were observed in motility, membrane fluidity and fatty acids on integration of exogenous phospholipids. Also different phospholipids induced different changes which were compared and correlated.

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P014

BIRTHWEIGHT PERCENTILES BY GESTATIONAL AGE FOR BIRTHS FOLLOWING FRESH AND THAW CYCLES IN AUSTRALIA AND NEW ZEALAND, 2002-2010

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Aim: Births following Assisted Reproductive Technology (ART) treatment were reported to have a higher proportion of low birthweight and preterm birth compared with the general birth population. The aim of this study was to establish the birthweight percentiles for singletons following fresh and thaw cycles and to compare ART-specific birthweight percentile with the general population for identifying the impact of ART treatment on fetal growth.

Method: Population study using 46,626 liveborn singletons following fresh cycles and 24,276 liveborn singletons following thaw cycles from the Australian and New Zealand Assisted Reproduction Database between 2002 and 2010 in Australia and New Zealand. Univariate analysis was used to determine the birthweight percentiles for singletons following fresh and thaw cycles and binomial test was used to compare the proportion of small for gestational age (SGA) and large for gestational age (LGA) births following ART treatment to general population.

Results: The mean birthweight for liveborn singletons following thaw (3,413g) cycles was significantly heavier than liveborn singletons following fresh (3,280g) cycles ($p < 0.0001$). Compared with general population, liveborn singletons following fresh cycles had a significantly higher proportion of SGA births for both male (10.6%, 95% confidence interval (CI): 10.2%, 11.0%)

and female (10.9%, 95% CI: 10.4%, 11.3%) infants. In contrast, the proportion of SGA births was significantly lower among male (6.1%, 95%CI: 5.7%, 6.6%) and female liveborn singletons (6.7%, 95%CI: 6.2%, 7.2%) following thaw cycles.

Conclusion: Caution should be used when using the birthweight percentiles established from general population for the detection of ART births at high risk.

P015

THE CORRELATION BETWEEN FOLLICLE STIMULATION DURATION AND FERTILIZATION RATE, POST-ICSI OOCYTE DEGENERATION RATE AND CLINICAL PREGNANCY RATE (CPR)

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Objective: To evaluate if prolonged follicle stimulation duration are related to fertilization rate, post-ICSI oocyte degeneration rate and clinical pregnancy rate (CPR) in women undergo IVF cycles stimulated using GnRH agonist (GnRHa) protocol.

Study methods: We performed a retrospective review on 222 IVF±ICSI cycles in TMC Fertility Centre from 1st January 2012 to 31st December 2012. All patients were below 38 years old and had undergone ovarian stimulation using GnRHa protocol. Patients were given final oocyte maturation with human chorionic gonadotrophin (hCG) when the follicular cohort consists of at least 3 follicles of 18-20mm in diameter. Oocyte retrieval was performed at approximately 37 hours post-hCG administration. ICSI was performed at 40-42hours post-hCG administration and fertilization assessment was performed at 16-18hours post-insemination/injection. Patients were categorized according to follicle stimulation duration. In group A (n=182), the follicle stimulation duration was 12 days or shorter, and in group B (n=40) it was 13 days and later.

Results: The mean number of oocyte retrieved for both groups was similar (13.0). The fertilization rate in group A (67.6%) was significantly higher compared to group B (61.3%), ($P=0.0156$). In contrast, the post-ICSI oocyte degeneration rate in group B (12.1%) was significantly higher compared to group A (8.1%), ($P=0.011$). The CPR appeared to be higher in group A (65.4%) compared to group B (57.1%), however the difference was not significant.

Conclusion: Prolonged follicle stimulation duration results in lower fertilization rate and higher post-ICSI oocyte degeneration rate in GnRHa cycles. However, it does not affect the CPR.

P016

CHROMOSOMAL ABERRATIONS IN IN-VITRO MATURED OOCYTES INFLUENCE ON IMPLANTATION AND ONGOING PREGNANCY RATES IN MOUSE MODEL UNDERGOING INTRACYTOPLASMIC SPERM INJECTION

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Implantation failure and early pregnancy loss have been reported to be closely related to the quality of mammalian oocytes, however the pregnant outcome of the embryos from in-vitro matured oocytes remains unknown. In this study, we examined the spindle assembly and chromosome segregation during differentiation maturation durations of in-vitro maturation of mouse oocytes, and analyzed the resulting implantation and pregnancy outcome to clarify the relations between the spindle and chromosome of in-vitro matured oocytes and the implantation and early pregnancy. We performed a controlled prospective study using female ICR strain mice superovulated with pregnant mare's serum gonadotrophins. Cumulus-enclosed germinal vesicle (GV) oocytes were collected and randomly cultured in in-vitro maturation medium. At three different hours from the onset of the culture, oocytes were removed from in-vitro maturation medium and one part of them were fixed and stained to alpha-tubulin and chromatin using confocal laser scanning microscopy, and the other part of them were fertilized after injected a sperm *in vitro*, and the resulting embryos were transferred into the pseudo-pregnant female mice. The rates of in-vitro maturation was not significant among three experiment groups and control group, however there were significant difference in spindle assembly and chromosome segregation. Accordingly, significant improvements

in post-implantation development were observed among these different groups. The present study indicates that the spindle and chromosome assembly of in-vitro matured oocytes can be improved with appropriate maturation duration, and enhance the post-implantation development potential of the resulting embryos, and decrease the implantation failure and early pregnancy loss.

P017

GONADOTROPINS-MEDIATED DYNAMIC ALTERATIONS DURING OOCYTE MATURATION IN VITRO: ANIMAL MODEL

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Gonadotropins (Gns) have been widely used in human oocyte *in vitro* maturation (IVM) in assisted reproduction technology (ART). However, there is no any specific dose and proof available on the concentration normally implemented in IVM media. Particularly, the specific effect of Gns on oocyte molecular and genetic events is poorly understood. We aimed to examine the dose-related effects of Gns on nuclear maturation, DNA integrity, mitochondria, especially the genetic and epigenetic dynamic alterations using bovine oocyte as ART animal model. We found that high dose Gns is absolutely detrimental include influence gene expression and imprint genes methylation in oocytes. Low doses of Gns is better than other dose groups according to above indicators in oocytes. 7.5 IU/ml dose is not safe although used in some labs after our exploring the oocytes internal molecular cascade and methylation. Our work is the first study on molecular events of Gns-regulated oocyte maturation in dose-response manner- from genetic to epigenetic levels. This will be great benefit to understand the abnormal imprint alterations undergone ART treatment in clinic, and potentially provide very useful guide for routine stimulation protocols considering the Gns' safety.

P018

GNRH AGONIST LONG PROTOCOL VERSUS SHORT PROTOCOL IN ADVANCED MATERNAL AGE WOMEN 35 YEARS OR MORE UNDERGOING ICSI: INITIAL STUDY

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ABSTRACT

Objective: To compare results of GnRH agonist long protocol vs. short protocol in women thirty five years or older undergoing IVF cycles (January-October 2013)

Setting : Fertility Clinic Melati Mother and Children Harapan Kita

Materials and methods: Participants & Interventions: a total of 30 women above 35 years old were included in this study. Eighteen women received long protocol while twelve women received short protocol. All women had FSH level less than 12 before start of treatment. Standard ICSI program was done and follow up for all cases. Women with previous poor response were excluded from the study.

Results: In total, among 30 participants, there was a variation in regarding both protocols, long protocol achieved pregnancy rate of 10.7 % while short protocol achieved 9,1 % pregnancy rate ($P > 0.05$). The matur oocytes more than 3 is cut off point to succeed the pregnancy that significantly related with the protocol ($P < 0.05$). No cases of severe OHSS were reported. Cost of drugs were significantly reduced with the short protocol ($P < 0.05$)

Conclusion: This Centre study shows that the long protocol and short protocol doesn't have different significant to get amount mature oocyte and pregnancy rate in women with age of thirty five or above. The study with larger sample is needed.

P019

EXPRESSION PROFILING IN PERIPHERAL BLOOD OF KLINEFELTER'S SYNDROME PATIENTS

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Objective: To investigate different expression genes (DEGs) in peripheral blood samples of Klinefelter's syndrome (KS) patients.

Design: Case-control study. The KS group consisting of 5 patients were compared with the group of 5 healthy controls.

Setting: Healthy volunteers were diagnosed without any symptoms of KS.

Patients: KS patients were identified by karyotype analysis, and represented corresponding symptoms.

Interventions: Neither KS group nor control group were treated by any interventions before collecting peripheral blood samples.

Main outcome measurements: Microarray and qRT-PCR

Results: Under thresholds of fold change >2 and q value <0.05, 21 DEGs were selected from 717 expressed genes (480 up-regulated, 237 down-regulated). Among these DEGs, FABP1, ADH1C, and ALDH1L1 that associate with metabolism regulation, were most significantly down-regulated in KS, which was verified by qRT-PCR. Additionally, it was not detected that X chromosomes owned any DEGs.

Conclusions: The down-regulated DEGs identified in this study are involved in metabolism regulation. Therefore, KS not only displays on variable genotype, but more importantly impacts on key functional genes of metabolism regulation.

P020

SYNTHESIS OF THIADIAZOLOTIAZINONE DERIVATIVES AND ASSESSMENT OF THEIR REPRODUCTIVE TOXICITY

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6-tertiarybutyl-4-Amino-3-mercapto-1,2,4-triazin-5-ones are used in pharmaceutical preparations of anti-inflammatory, CNS stimulants, sedatives, antianxiety, antimicrobial, antifungal, anti-HIV infection and anticancer agents. These compounds are also potential herbicides and used in agriculture set up. Due to the increased resistance of microbial organisms or pests towards triazinone derivatives the synthesis of new generation drugs with high antimicrobial and antifungal efficiency is of paramount importance. In the present study we synthesized various derivatives of 4-amino-3-mercapto-4H-1,2,4-triazin-5-ones and named them as E1, E2, E3, E4, E5, and E8. These agents were analyzed for male reproductive toxicity in Swiss albino mice. No significant difference in the activity of 4-amino-3-mercapto-4H-1,2,4-triazinone and its derivatives on sperm count, motility and nuclear maturity was observed in mice. In addition, when DNA damage was assessed in bone marrow cells of these mice E5 derivative had marginally higher clastogenic effect when compared to rest of the groups. Therefore,

the compound E4 is more preferable larvicidal agent than the parent compound to minimize the toxic health hazards to non-target organisms, including human beings.

P021

DYNAMIC PROTEOMIC PROFILES OF IN VIVO AND IN VITRO PRODUCED MOUSE POST-IMPLANTATION EXTRA-EMBRYONIC TISSUES AND PLACENTAS

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As the interface between the mother and the developing fetus, the placenta is believed to play an important role in assisted reproductive technologies (ART) induced aberrant intrauterine and postnatal development; however, the mechanisms underlying this aberrant placentation are still unclear. This investigation provides the first comparative proteomic analysis between extra-embryonic tissues and placentas after *in vivo* fertilization and development (IVO) and *in vitro* fertilization and culture (IVP). We identified 165 and 178 differentially expressed proteins (DEPs) between IVO and IVP extra-embryonic tissues and placentas at embryonic day 7.5 (E7.5) and E10.5, respectively. Many DEPs were functionally associated with genetic information processing, such as impaired *de novo* DNA methylation, as well as post-transcriptional, translational and post-translational dysregulation. Global hypomethylation, as well as lower correlation between transcriptome and proteome in IVP groups, provided further confirmation of these novel findings. In addition, numerous DEPs were found to be involved with mitochondrial function, cytoskeleton organization and transport, as well as vasculogenesis and angiogenesis. These disturbed processes and pathways are likely to be associated with embryonic intrauterine growth restriction, enlarged placenta, and impaired labyrinth formation. This study provides a direct and comprehensive reference for further exploring the placenta involved mechanisms underlying ART induced aberrations.

P022

IN VITRO FERTILIZATION AND CULTURE INDUCED XIST DEPRESSION IS INVOLVED IN THE SKEWED SEX RATIO AT BIRTH

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Recently, epidemiological analysis showed that assisted reproductive technology (ART) processes could also lead to a skewed sex ratio at birth. However, the underlying mechanism is still unclear. Using mice as model, our study demonstrated that *in vitro* produced (IVP, including *in vitro* fertilization and culture) embryos would result in a higher male birth rate, compared with *in vivo* produced (IVO) embryos. Further analysis showed that the skewed sex ratio (1.3:1) had already occurred during early post-implantation development period (before E7.5) due to a female biased lethality. Moreover, our "omics" data showed that the IVP embryos were characterized with a higher transcriptional activity and a lower level of DNA methylation on X chromosome, which implies that IVP underwent an incomplete X chromosome inactivation (XCI). To prove this hypothesis, we detected the XCI status of IVP embryos at morula and blastocyst stage: the depressed *Xist* expression in IVP embryos was confirmed by both quantitative RT-PCR and RNA FISH analysis; immunofluorescence analysis of H3k27me3 at blastocyst, a well-known indicator of XCI, showed a same result. In addition, we explored the possibility to alleviate the deficient *Xist* expression by supplementing retinoic acid (RA), which could promote *Xist* activation, in *in vitro* culture media. RA at 5 nM for 36 hours could significantly increase *Xist* expression, without an obvious side-effect on the developmental capacity. Our study proposed that ART processes could lead to the depressed *Xist* expression, which results in a female biased embryonic lethality during post-implantation period, and eventually increased the male birth rate.

P023

LOW DOSE HCG FOR LUTEAL PHASE RESCUE IN GnRH ANTAGONIST CYCLES WITH AGONIST TRIGGER FOR PCOS CASES FOR ICSI.

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OBJECTIVE-The reproductive outcomes following the administration of low dose hCG on the day of oocyte retrieval, on the day of embryo transfer and 3 days later were evaluated in cases of PCOS. **DESIGN:** Prospective study. **MATERIALS AND METHODS:** 100 infertile women with PCOS who are at high risk of ovarian hyperstimulation syndrome (OHSS), having total of dominant follicles (more than 16mm) in 14 nos on the last day of stimulation was included. They underwent ICSI treatment with a GnRH antagonist protocol. Final oocyte maturation was achieved with a single bolus of 2mg of Luprolide acetate and patients received IM bolus of HP hCG 1500IU s/c right after oocyte retrieval, on embryo transfer day, 3days after embryo transfer. **RESULTS:** The incidence of clinically significant ovarian hyperstimulation syndrome was 12%(6/50), admission treatment was required only in three cases(6%). Implantation rate was 36% and clinical pregnancy rate was 44%(22/50). **CONCLUSION:** Low dose hCG successfully rescues the luteal phase in GnRH agonist trigger cycle, provides excellent reproductive outcomes. The incidence of ovarian hyperstimulation syndrome was much decreased, hospitalization treatment was required in rare cases.

P024

LIVE BIRTH RATE AFTER GnRH AGONIST TRIGGERING AND INTENSIVE LUTEAL STEROID SUPPORT IN STIMULATED CYCLES AT HIGH RISK OF OHSS

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Introduction: GnRH agonists are highly effective in inducing an LH surge. This characteristic has been exploited clinically in OHSS high risk patients. There is a lack of consistency in the literature regarding the pregnancy outcomes of GnRHa triggering and intensive luteal support.

Objective: To investigate the pregnancy outcomes of OHSS high risk patients having GnRHa triggering and intensive luteal support.

Materials and methods: An observational study was conducted at An Sinh Hospital, HCMC, Vietnam from April 2010 to April 2012. Three-hundred eighty-three patients undergoing COS using a GnRH antagonist protocol, who had ≥ 15 follicles of ≥ 12 mm on the day of trigger, were included. Triptorelin 0.2mg was injected subcutaneously to trigger endogenous LH surge. Luteal support included intramuscular progesterone 50 mg/day, progesterone gel 2 applicators/day and oral estradiol valerate 2 mg t.i.d, from the day of oocyte pick-up until pregnancy testing.

Results: Mean age of patients was 31.04 ± 3.7 . Clinical pregnancy, implantation, miscarriage and live birth rates per ET were 35.5%, 15.1%, 5%, and 29.9%, respectively. One case of severe late OHSS occurred.

Conclusions: This large observational study confirms that GnRHa trigger combined with intensive luteal steroid support achieves satisfactory live birth rate with minimal OHSS risk.

P025

THE EFFECTS OF OOCYTE RETRIEVAL TIMING ON THE RATE OF MATURE OOCYTES AND QUALITY OF OOCYTES IN STIMULATED CYCLES USING GNRH AGONIST FOR TRIGGERING MATURATION

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Introduction: The GnRH agonist (GnRHa) induced LH surge is significantly shorter than that observed during hCG trigger cycle. Therefore, the optimal oocyte retrieval (OR) timing requires to be re-determined.

Objective: To investigate the effects of OR timing on the rates of mature and poor-quality oocytes in GnRHa triggering cycles.

Methods: A retrospective cohort study was conducted on 365 GnRHa triggering cycles, including 196 donor cycles and 169 non-donor cycles at IVFAS, An Sinh hospital from January to August 2012. Oocyte retrieval was performed at different timings of 31, 32, 33, 34, 35, 36, 37 hours after GnRHa administration. Primary endpoints were the rates of mature and poor-quality oocytes at different timings of OR.

Results: No significant difference was found in the rate of mature oocytes at different OR timings in both donor and non-donor cycles. The rate of poor-quality oocytes was significantly higher in cycles that had OR later than 36 hours compared to before 36 hours in both donor cycles (14.1% vs. 5.4%, $p=0.003$) and non-donor cycles (7.8% vs. 4.1%, $p=0.01$).

Conclusions: In GnRHa triggering cycles, the appropriate timing for OR would be between 31 and 36 hours after the GnRHa injection.

P026

SLOW FREEZING TECHNIQUE IS BETTER THAN VITRIFICATION IN THE SURVIVAL OF POST THAWED EMBRYOS, IS IT A MYTH OR A FACT?

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Objective: To study the survival rate of thawed embryo following slow freezing and vitrification techniques.

Design: A retrospective study

Setting: The Saudi Center for Assisted Reproduction, Abha, Saudi Arabia

Method: 516 freezing cycles from June 2005 till June 2012, cryopreservation either by slow freezing or vitrification methods was carried out.

Main outcome measures: Survival rate of thawed embryos following cryopreservation by both techniques was assessed as well as the pregnancy rate.

Results: Survival rate of Grade I, Grade II post thawed embryos following slow freezing cryo-technique is statistically significant and higher than vitrification with a P value of less than 0.001. Positive pregnancy rate following slow cooling was found in 34.8% as compared to 29.5% in vitrification which is also statistically significant less than 0.001.

Conclusions: Survival rate of post thawed embryos of Grade I and II were better with slow freezing cryotechnique than vitrification and also the pregnancy rate was found to be high and statistically significant with slow freezing than with vitrification.

P027

COMPARISON OF MCOMPARISON OF MECHANICAL ARTIFICIAL SHRINKAGE METHODS IN MOUSE BLASTOCYST VITRIFICATION

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Purpose: This study was designed to investigate which mechanical artificial shrinkage method, induced by a aspiration, puncture or pipetting, results in better survival and reexpanding rate in vitrified-warmed mouse expanding blastocysts.

Methods: In each group, 50 mouse blastocysts were used. Before vitrification, the blastocoelic cavity was collapsed by direct aspiration by ICSI pipette, puncture by micro-needle and pipetting. Post-warm blastocyst survival and re-expanding rate were examined in each artificial shrinkage method. Re-expanding rate was checked at 1, 2 and 4hour after warming.

Results: Survival rates were 95% in aspiration group, 93% in micro-needle puncture group and 92% in pipetting group. Re-expanding rates were 89%, 84% and 82% in each groups. In the aspects of re-expanding time, direct aspiration group shows more rapid re-expanding than other two groups. At 1 hours after warming, in direct aspiration group, 85% of survival mouse blastocysts undergoes hatching state, but in other two methods, the hatching rates were 70% in micro-needle puncture group and 55% in pipetting group. Direct aspiration method showed the shortest time for AS.

Conclusions: AS is important procedure for blastocyst vitrification. Several different mechanical AS methods were introduced. In our study, we can confirmed the direct aspiration of blastocoelic fluid is very effective and easy way for AS in mouse blastocyst.

P028

A CASE CONTROL STUDY OF MELATONIN IN MONOTHERAPY AND COMBINATION WITH CO-ENZYME Q10

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Aim:

Oxidative stress and mitochondrial dysfunction have been implicated in poor oocyte and therefore embryo quality. Melatonin is a broad spectrum antioxidant and free-radical scavenger. CoQ10 is essential for ATP production and antioxidant defense. We examined IVF outcomes in women given melatonin alone or with CoQ10.

Method:

Retrospective analysis of 30 women given antioxidant therapy; melatonin 4mg alone (n=13) or with CoQ10 150mg (n=17). Three controls per case were matched for age, cycle number, stimulation type and dose (n=90). Primary outcomes were follicles >11mm, oocyte number, MII number and oocyte maturation. Secondary outcomes included utilisation ratios, fertilisation and pregnancy rates. Treatment group was additionally analysed according to age (<40,>40) and previous IVF response (poor/non-poor responder). T-tests used for continuous data, Fisher's exact test for categorical data; p<0.05 considered significant.

Results:

No significant improvements occurred in any treatment group. Trends were seen towards increased MII oocytes (4.5 vs 5.7) and oocyte maturation (68% vs 75%) in all treated patients, with no change seen in the controls (78% vs 77%). Poor responder patients (n=12) showed a significant improvement in MII egg number (1.8 vs 4.25, p=0.009, 95% CI 0.71-4.22), and in oocyte maturation (50% vs 83%, p=0.005) not seen in the remainder of the group. No other changes were seen.

Conclusion:

Antioxidant therapy in poor-responder patients led to a significant increase in mature egg numbers and oocyte maturation not seen in the general IVF patients. Addition of CoQ10 to melatonin did not improve outcomes. Further study is required to assess result validity.

P029

THE CLINICAL OUTCOMES OF LH SUPPLEMENTATION IN GNRH AGONIST LONG PROTOCOL IN KOREAN WOMEN

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Objective

There have been a lot of debates about the use of additional luteinizing hormone (LH) on controlled ovarian stimulation (COS). The purpose of this analysis was to verify the outcome of additional LH supplementation depending on age of patient time of initial injection on the patients undergoing GnRH agonist long protocol therapy.

Materials & Methods

A total of 213 patients was divided into three groups for LH injection. Group A (n=91): FSH only injection, Group B (n=90): FSH+early follicular phase LH additional injection, Group C (n=32): FSH+mid follicular phase LH additional injection. Each group was divided again by age.

Results

The number of retrieved oocyte was significantly lower in the Group 2 than in Group 1 and Group 3 ($P<0.001$). The number of mature oocyte was significantly lower in the Group 2 ($P<0.001$). The number of blastocyst was significantly lower in the Group 2 ($P<0.001$). Clinical pregnancy rate was lower in the Group 2, even though there was no significant difference statistically ($P=0.011$).

Conclusions

An advantage of additional LH injection was not proved in this study. However, the number of retrieved and mature oocyte in mild-follicular phase LH additional injection group was not low compared to those of in FSH only injection group. Though it was not statistically valid, clinical pregnancy rate was highest in the mid follicular LH additional injection group. Therefore, further prospective investigations are necessary to validate effects of LH additional injections in mid follicular phase.

P030

NEONATAL DATA AFTER TRANSFER OF BLASTOCYST VITRIFIED USING A CLOSED SYSTEM

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Objectives: Closed vitrification system (CVS) may enable the risk of contamination to be minimised. We have revealed that human embryos are vitrified using a CVS without impairment of developmental competence to the blastocyst stage and to the fetal stage (Hashimoto et al., 2013). In this study, the neonatal data after the transfer of blastocyst vitrified using a CVS were investigated.

Methods: Human blastocysts were vitrified using either a CVS (Rapid-i®) or an open vitrification system (OVS; Cryo-top®). Single blastocyst transfer was performed after warming. Neonatal data after the transfer of blastocyst vitrified using the CVS (n = 47) were compared with that using the OVS (n = 101).

Results: There were no differences between the CVS and the OVS in mother age (CVS: 34.9 y vs. OVS: 34.4 y), mother BMI (CVS: 20.3 vs. OVS: 20.1) and implantation rate (CVS: 43.2%, n = 139 vs. 44.8%, n = 317). There were also no differences between the CVS and the OVS in the gestational age (CVS: 271.6 days vs. OVS: 275.2 days), birth weight (CVS: 3027.7 g vs. 3048.7 g), the Apgar score (CVS: 9.1 vs. 9.2), the occurrence of congenital anomaly (CVS: 2.1% vs. OVS: 0%), and the maternal complication (CVS: 12.8% vs. OVS: 15.0%). The neonatal data for blastocysts vitrified using the CVS was similar to that with the OVS. Data of the present study showed that a CVS didn't affect the neonatal data compared with an OVS.

P031

GNRH AGONIST VERSUS ANTAGONIST IN NORMAL RESPONDERS - INDIAN SCENARIO

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Introduction-

For many years now, ART treatment involves using GnRH analogues. GnRH agonist use with long protocol has been considered as gold standard for normal responders since a long time. In contrast GnRH antagonist have been used for poor responders or older women. Recently many studies abroad have been done on evaluating agonist versus antagonist in normal responders. GnRH agonist administration causes gonadotropin suppression via pituitary desensitization after an initial short period of hyperstimulation. In contrast, Antagonist causes immediate and rapid suppression by competitive occupancy of GnRH receptors.

Material and Methods

We studied cases from various centres and compiled the results of Agonist versus Antagonist in normal responders.

Study was done from DEC 2011 till DEC 2013. Strict inclusion and exclusion criteria were made and cases were selected. Main outcome measure being Pregnancy rate. Other outcome measures were Fertilization rate, OHSS rate, Abortion Rate and multiple pregnancy rate.

Conclusion-

The result of the study did show the two protocols were very similar in outcomes.

Immediate mode of action, shorter duration, low incidence of hospital admission did make Antagonist a better choice.

There was no significant difference in rate of live births.

GnRH Antagonist protocol is a 'PATIENT FRIENDLY PROTOCOL'

P032

CUMULATIVE PREGNANCY RATE OF CORIFOLLITROPIN ALFA (ELONVA) IN LOWER BODY WEIGHT IVF PATIENTS

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Introduction: One Elonva injection can replace 7 daily recFSH injections, reducing patients' physical stress. Data on Elonva use in lower body weight patients is still limited.

Objective: To investigate the efficacy and safety of Elonva for controlled ovarian stimulation (COS) in lower body weight IVF patients.

Methods: A case series study was conducted on 132 patients at IVFAS, An Sinh Hospital from July 2012 to May 2013. COS was performed using GnRH antagonist protocol. Elonva was injected on cycle day 2. GnRH antagonist 0.25mg was started from day 5 of stimulation. hCG was administered when at least 2 lead follicles reached 17mm. In patients having more than 15 developing follicles, hCG was replaced by GnRH agonist.

Results: A total of 71 oocyte donors and 61 IVF patients had Elonva used for COS. Mean number of oocytes retrieved and embryos were 15 and 9, respectively. The rates of ongoing pregnancy and implantation were 34.1% and 23% in donor cycles, and 30.8% and 18% in IVF patients. The cumulative pregnancy rate after 3 transfers from an Elonva cycle was 60% (in both donor cycles and IVF patients).

Conclusions: Elonva is effective and convenient for COS in lower body weight IVF patients.

P033

DEFINING THE OPTIMAL TIME FOR OCCYTE RETRIEVAL IN LONG PROTOCOL, FLARE-UP AND ANTAGONIST PROTOCOLS: A PROSPECTIVE STUDY

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Objective: To determine the optimal time for oocyte retrieval in ART.

Materials and Methods: A prospective study recruited 1049 patients undergoing ICSI/ET. 10000 IU of uhCG was used for trigger. The interval time between hCG injection and egg retrieval (T) was divided into four groups: T1 ≤35 h; 35h< T2 ≤36h; 36h<T3 ≤37h; T4 >37h.

Results: 663 patients underwent long protocol, 208 patients underwent flare-up protocol, and 178 underwent antagonist protocol were enrolled. In long protocol, MII oocyte rate was significantly lowest in group of T1 (86.6%) compared to groups of T2, T3, and T4 (97.7%, 97.7% and 92.6% respectively). The fertilization rate was significantly highest in group of T2 (82.9%). The implantation and clinical pregnancy rates were lowest in group of T1 (9.68% and 37.5%, respectively). In flare-up protocol, MII oocyte rate was significantly lower in groups of T1 and T4 (92.2% and 81.2%) compared to groups of T2 and T3 (95.4% and 95.4%). The fertilization rate was also significantly lower in groups of T1 and T4 (70.9% and 69.2%). In antagonist protocol, MII oocyte rate was significantly lowest in group of T1 (86.6%). The fertilization rate was significantly highest in group of T3 (84.7%). The implantation and clinical pregnancy rates did not differ in flare-up and antagonist protocols.

Conclusions: Oocyte retrieval should not be performed before 35h after hCG administration. The optimal time for pick up in long protocol was 35h < T2 ≤ 36h, in flare-up protocol was 35h < T2 & T3 ≤ 37h and in antagonist protocol was 36h < T3 ≤ 37h.

P034

RESULTS OF THE FIRST TRIMESTER SCREENING (FTS) AND DIAGNOSTIC TESTS FOR ANEUPLOIDY IN ICSI PREGNANCIES COMPARED TO NATURAL PREGNANCIES

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Introduction: An increased risk of chromosomal aberrations has been reported in ICSI offspring.

Objective: To compare the results of the FTS and diagnostic tests for aneuploidy between ICSI and natural pregnancies.

Methods: A retrospective cohort study was conducted on 985 singletons at My Duc Hospital from October 2012 to August 2013. Pregnancies were divided into 2 groups based on method of conception, natural or ICSI. All women had FTS using a combination of age, biochemical markers (PAPP-A, free b-hCG) at 11W and nuchal translucency at 12W. High aneuploidy risk was defined when the adjusted risk was > 1/300, using FMF software. Amniocentesis was performed on high risk patients at 18W.

Results: A total of 985 pregnancies were studied, in which 418 were ICSI and 567 natural pregnancies. Mean age and rate of high aneuploidy risk in ICSI were significantly higher than natural conception women (32 ± 4 vs. 30 ± 4, and 15.6% vs. 9.9%, p<0.01, respectively). Twenty-eight ICSI and 34 natural pregnancies had amniocentesis. One case of trisomy 21 was detected in natural pregnancy group (2.9%).

Conclusion: There was an increase rate of high aneuploidy risk in ICSI pregnancies; however, no case of aneuploidy was detected in this group.

P035

THE ADDITION OF LOW DOSE HCG TO CORIFOLLITROPIN ALFA FOR CONTROLLED OVARIAN STIMULATION (COS) IN IVF-ET

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Introduction: One injection of corifollitropin alfa (Elonva, MSD) replaces 7 daily injections of gonadotropin during COS. hCG could be used as a top-up gonadotropin in the final days of COS.

Objective: To investigate the efficacy of using hCG as a top-up gonadotropin after Elonva injection during COS.

Methods: This was a case-series. COS was performed by using GnRH antagonist protocol. Patients, who weighed < 60kg, received an injection of Elonva 100 µg on day 2 of menstrual cycles. GnRH antagonist 0.25mg was injected from day 5 of stimulation. On day 8 of stimulation, if patients required daily gonadotropin, hCG instead of rFSH was injected 200 IU/day until there were at least 2 follicles of ≥ 17mm in both ovaries. hCG 5000IU was administrated for triggering maturation.

Results: Eight patients had the addition of hCG to Elonva during COS. Mean number of oocytes retrieved was 11.25 ± 3.32 . The implantation and clinical pregnancy rates were 16.6% and 37.5%, respectively. None of the patients developed any form of OHSS.

Conclusions: hCG can substitute rFSH in the final days of Elonva cycles, leading to a reduction of rFSH consumption. More studies are awaited to confirm the efficacy of this protocol.

P036

ENHANCED PPARG EXPRESSION IN THE CHORIONIC VILLI TISSUE IS RELATED TO MISCARRIAGE

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Miscarriage is the most common disorder of human pregnancy. Although several important factors related to miscarriage have been identified, the pathogenic mechanisms accounting for 40% to 60% of miscarriage cases are still unclear, thus requiring the identification of novel factors contributed to the causes of miscarriage. In this study, we analyzed the expression pattern of peroxisome proliferator-activated receptors- γ (PPAR γ) in the chorionic villi tissues obtained from normal pregnant individuals and miscarriage patients during the first trimester of pregnancy. While PPAR γ expression was detected in both groups, a significant enhanced expression of PPAR γ was observed in the villi tissues from miscarriage patients. Our findings proved that the enhancement of PPAR γ expression is related to miscarriage, suggesting the potential involvement of PPAR γ signaling in the pathology of miscarriage.

P037

HOW ESSENTIAL IS SYNCHRONIZATION IN ACHIEVING SUCCESSFUL EMBRYO IMPLANTATION?

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Timing embryo transfer to coincide with the "Implantation Window" has been thought to be crucial in achieving successful implantation. The optimal duration of progesterone pretreatment before the transfer of cryopreserved-thawed embryos in estradiol/progesterone-supplemented cycles is still a topic of debate.

We report a successful IVF pregnancy despite suboptimal priming of endometrium by progesterone. A 31-year-old woman with recurrent implantation failure began her third artificial FET cycle with 4mg of Estradiol Valerate, daily. After 8 days of estrogen treatment, transvaginal ultrasound scan showed her endometrial thickness to be 9mm, with no evidence of ovulation. She was instructed to insert vaginal progesterone suppository Cyclogest 200mg twice a day but later realized that she forgot to insert the prescribed dose of vaginal Cyclogest on day 2. She was instructed to start insertion and ET was done on day 3. Successful implantation for one embryo was achieved.

Progesterone priming of endometrium is thought to be vital as evident right down to the molecular level via complex endocrine and paracrine signal transduction. [ENREF 2](#) Our case illustrates implantation with minimal progesterone pretreatment. Our unique experience suggests that this elusive 'window of implantation' may vary with the individual, perhaps even shortened in some women, allowing less time for the blastocyst to achieve implantation before the endometrium turns refractive and toxic to the embryo.

P038

COMPARISON OF SUCCESSFUL PREGNANCY RATE BETWEEN FLEXIBLE AND RIGID CATHETER IN INTRA UTERINE INSEMINATION

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Objective: To evaluate comparison of succesful pregnancy rate between flexible and rigid catheter in intra uterine insemination (IUI).

Methods: An analytic observational with cohort study design to evaluate the comparison of succesful pregnancy rate between flexible and rigid catheter in IUI were performed at the Halim Fertility Centre and Stella Maris–Women and Children Hospital in Medan from July until September 2013.

Result: Blood on the catheter more often found in groups of rigid catheter 18 (60%) compared to the flexible catheter 17 (56.7 %), but we found no significant differences by Chi-Square test with $P > 0.05$. Reflux more common in rigid catheter group 8 (26.7 %), whereas only found in 1 case using a flexible catheter (3.3 %). We found significant differences through the Chi-square test with p value < 0.05 level. IUI success rate obtained higher in flexible catheter group (26.7 %) compared with rigid catheter (20 %), but we found no significant differences by Chi-Square test whereas p value 0.542. Higher success also earned in the group without the presence of blood on the tip after insertion, but there is no significant differences between groups in the presence of blood or not.

Conclusion: There is no difference in the success rate of intra-uterine insemination using either rigid or flexible catheter. However, it is better if clinicians still consider inconvenience or uncomfortable conditions caused by one type of catheter during insertion procedure.

Key Words : Intra uterine insemination, Rigid Catheter, Flexible catheter

P039

IT'S NOT JUST ABOUT NUMBERS: YOUNG WOMEN WITH LOWER THAN EXPECTED EGG YIELDS HAVE RELATIVELY POOR EMBRYO QUALITY AND LOW SUCCESS RATES

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Introduction

At IVFAustralia cleavage stage transfers are almost all associated with low embryo numbers and, in young women, current knowledge would still predict a relatively good outcome. This study tested this hypothesis with a particular focus on AMH (ovarian reserve) and embryo quality.

Methods

IVF cycles with cleavage stage transfers (2011-12) in women < 38 yo were analysed. Optimal embryos were defined as containing 4 cells on day 2, and ≥ 6 cells on day 3. "A" grade embryos had $< 25\%$ of cell volume occupied by fragmentation, and "B" grade embryos had $\geq 25\%$ fragmentation.

Results

In 145 cycles (mean age 34.5; AMH 6; eggs collected 6.1; eggs fertilised 2.3) the clinical pregnancy rate was 19.3%. Transfer of suboptimal embryos resulted in significantly lower pregnancy rates (3.8%) compared with cycles in which at least one optimal embryo was transferred (16.7%) ($p < 0.05$). Those with AMH $< 25^{\text{th}}$ centile ($n = 43$, median 3.1, age 35.4) were

compared with those >25th (n=38, median 14.95, age 34.2) and despite higher FSH starting dose stimulation ($p<0.01$), the low AMH group had fewer eggs retrieved ($p<0.01$), fewer fertilised, and fewer embryos frozen, more morphologically suboptimal embryos transferred (37.2% v 26.3%) and lower pregnancy rates (11% v 16%).

Conclusions

Lower than expected egg numbers collected in young women undergoing IVF resulted in worse than expected outcomes. This was associated with poor embryo quality and low AMH. While the link between low AMH and poor ovarian reserve is well described, more studies are needed to exclude AMH as a marker of poor egg/embryo quality as well.

P040

DOES TRANSFERRING FROZEN-THAWED EMBRYOS ONE DAY EARLIER IN NATURAL CYCLES AFFECTS THE CLINICAL PREGNANCY RATE?

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Objective: To compare the clinical pregnancy rate (CPR) of frozen-thawed embryos (FET) done in natural cycle either in a synchronized or non-synchronous (Day -1) endometrium.

Design: Retrospective study.

Materials and methods: We analysed In Vitro fertilization (IVF) outcomes in 218 ovulatory patients who underwent a Day 2 FET between January 2010 and December 2011 at KKIVF Centre, Singapore. All embryos had been cryopreserved by the slow-controlled-freezing method following a previous ICSI cycle for male factor (82%), tubal disease (9%), idiopathic infertility (6%) and other reasons (3%). The endometrial preparation consisted in a natural cycle whereby a double FET was scheduled either 2 days (Group A, n=50) or 3 days (Group B, n=168) after the endogenous LH surge.

Results: Patients' characteristics such as age, ethnic group, parity, FSH were similar in both groups. The cycle parameters (follicular size and endometrial thickness on the day before ovulation) were also comparable between the 2 groups. The CPR in group A was 28% (14/50) and 36.9% in group B (62/168). The difference between the CPR of the 2 groups was not significant (odds ratio [OR], 0.66; 95% confidence interval [CI], 0.33-1.33).

Conclusions: To avoid cancelling natural cycles FET falling on non-working days, bringing the ET one day forward appears to be a reasonable option. However, a larger study is needed to confirm this preliminary result.

P041

A PROSPECTIVE RANDOMIZED STUDY COMPARING THE OUTCOME OF ART BETWEEN A THINNER-TIPPED NEEDLE AND A STANDARD NEEDLE FOR OOCYTE RETRIEVAL

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Introduction: Oocyte retrieval plays a key role in terms of achieving a successful pregnancy at ART. The aim of this study is to compare the outcome of ART between the thinner-tipped needle (TN) (20 gauge for the last 5cm from the tip of the needle and 17 gauge for the remaining length of the needle) and a thicker standard needle (SN) (18 gauge).

Materials and Methods: A prospective randomized study was performed at our clinic from July to October 2013. Forty cycles that consisted of 21 cycles with the TN and 19 cycles with the SN were enrolled in this study.

Results: The mean BMI index in the TN group (20.2 ± 2.6) was also similar to that in the SN group (22.1 ± 4.4). However, the instance of minimal vaginal bleeding after the oocyte retrieval in the TN group (76.2%:16/21) was significantly higher than that in the SN group (36.8%:7/19). There was no significant difference in the normal fertilization rate between the two groups (TN group; 74.4% (166/223) vs. SN group; 71.2% (126/177)). Moreover, there was no significant difference in the good-quality embryo rate on day 3 between the two groups (TN group; 61.5% (102/166) vs. SN group; 54.8% (69/126)).

Conclusions: Oocyte retrieval with the thinner-tipped needle resulted in less vaginal bleeding, compared to retrieval with a standard needle. Additionally, the outcome of ART between the thinner-tipped needle and the standard needle was almost the same. From these results, we recommend using the thinner-tipped needle in terms of safety.

P042

GENETIC AND CLINICAL PREDICTORS OF OVARIAN RESPONSE IN CONTROLLED OVARIAN STIMULATION WITH RECOMBINANT FSH

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Background:

Several factors are known to influence ovarian response to rFSH stimulation such as age, antral follicle count (AFC), and basal FSH level. Mutation of allele Ser680Asn in FSHR gene was responsible to ovarian resistency toward exogenous FSH.

Objective:

To develop a prediction model of ovarian response to COS in IVF

Design:

Prospective cohort study

Method:

One hundred and thirteen women undergoing their first cycle of IVF in Yasmin IVF Clinic Jakarta were recruited to this study. Clinical datas included were age, BMI, and AFC. Basal FSH and E2 as well as serum AMH was measured from peripheral blood taken at second day of cycle. Bsr-1 enzyme is used to identify the polymorphism in exon 10 position 680 with RFLP technique. Three genotype polymorphisms, Asn/Asn (255 bp ribbon), Asn/Ser (97 bp and 158 bp), and Ser/Ser (97 bp, 158 bp, and 255 bp)

Results:

AFC has the highest predictor for ovarian response with AUC 0.922 (CI 95% 0.833-1.000). AMH also showed high predicting value (AUC 0.843 CI 95% 0.663-1.000). The multivariate analysis revealed combination of AFC, AMH, age and basal FSH is a good model for ovarian response prediction (AUC=0.97). No significant relation between Asn/Asn, Asn/Ser, or Ser/Ser genotype FSHR polymorphism with ovarian response ($p=0.866$) and total dose of rFSH ($p=0.08$)

Conclusion:

This study showed that model combination of AFC, AMH, patient's age and basal FSH are very good to predict number of mature oocytes

Keywords

prediction model, ovarian response, in vitro fertilization

P043

THE RELATIONSHIP BETWEEN FOLLICULAR DIAMETER AND SERUM ESTRADIOL (E2) IN IN VITRO MATURATION, IN VITRO FERTILIZATION AND EMBRYO TRANSFER (IVM-IVF)

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(Objectives)

It is important to determine what factors are related to better clinical outcomes in IVM-IVF. The present study was conducted to investigate the relationship between follicular diameter and E₂ level in IVM-IVF.

(Materials and Methods)

Total of 250 IVM-IVF cycles (142 cases) under 39 years old were divided into 2 groups based on the average diameter of the largest and second largest follicles on the day of HCG administration (A: 8-11mm, B: 11-13mm). Moreover, A and B were divided into 3 subgroups based on their E₂ levels (a: >75pg/ml, b: 75-140pg/ml, c: <140pg/ml) and (d: >90pg/ml, e: 90-

200pg/ml, f: <200pg/ml). The number of oocytes retrieved, maturation rate, fertilization rate, implantation rate and cancellation rate of transfer were analyzed.

(Results)

The number of oocytes retrieved in Group A-b (13.2 ± 9.9) was significantly higher than either Group A-a (8.7 ± 5.7) or Group A-c (8.7 ± 6.2). Implantation rate was significantly higher in Group B-e (44.4%) than either Group B-d (0%) or Group B-f (5.6%). Transfer cancellation rate of Group A-b (10.5%) was significantly lower than either Group A-a (38.7%) or Group A-c (47.1%). Transfer cancellation rate of Group B-d (13.0%) was significantly lower than Group B-f (47.1%). No significant differences were confirmed in other parameters.

(Conclusions)

Better clinical outcomes are expected with follicular diameter of 8-11mm and E2 level of 75-140 pg/ml or follicular diameter of 11-13mm and E2 level of 90-200 pg/m in IVF-IVF.

P044

DOES SERUM ESTRADIOL ON THE DAY OF HCG AFFECT IVF SUCCESS?

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Aim: To assess the role of serum estradiol on the day of injection HCG, on clinical pregnancy rate and oocyte/embryo quality

Materials and Methods: A retrospective review of three hundred and forty two in vitro fertilization cycle with normal ovarian reserve who underwent long GnRH agonist protocol were included. Outcomes assessed are number of oocytes retrieved(OR), fertilization rate(FR), number of cryo-preserved embryos (CPE) and clinical pregnancy rate(CPR).

Results: A positive correlation was seen between Estradiol/follicle (E2/fol) ratio and oocytes retrieved ($r=.334$, $pvalue=.0001$), no. of mature oocytes ($r=.335$, $pvalue=.0001$), no. of oocytes fertilized ($r=.222$, $pvalue=.002$) and number of cryopreserved embryos ($r=.289$, $pvalue=.0001$). Increased clinical pregnancy rate (CPR) was seen in Group C (E2/fol= 200-299.99) compared to Group A, B&D ($pvalue=.033$). With Estradiol/Oocyte (E2/O) ratio negative correlation was seen between E2/O and oocytes retrieved ($r=-.281$, $pvalue=.002$), mature oocytes ($r=-.296$, $pvalue=.008$), oocytes fertilized ($r=-.220$, $pvalue=.003$), embryos cleaved ($r=-.211$, $pvalue=.004$), Grade 1 embryo ($r=-.216$, $pvalue=.001$), cryo-preserved embryos ($r=-.206$, $pvalue=.005$). No difference in fertilization rate, cleavage rate or clinical pregnancy rate was seen. Total serum estradiol did not affect clinical pregnancy rate.

Conclusion: In conclusion serum estradiol is an important determinate of IVF success. While total serum estradiol does not exert any positive or negative influence on IVF outcome, estradiol per mature follicle and retrieved oocytes does have an impact. Pregnancy rate is better when E2/fol is between 200-299.99 pg/ml. Also increasing serum E2/fol positively correlates with better oocytes and embryo quality. In contrast E2/O negatively correlates with oocytes and embryo quality parameters.

P045

A RANDOMISED CONTROLLED TRIAL ON THE EFFECT OF DEHYDROEPIANDROSTERONE (DHEA) TREATMENT ON POOR RESPONDERS TO IVF TREATMENT: STUDY DESIGN.

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Objective: To evaluate the effects of Dehydroepiandrosterone (DHEA) supplementation on the outcome of ICSI treatment in poor ovarian responders and to investigate its action on ovarian steroidogenesis, on biochemical and ultrasonographic markers of ovarian reserves.

Design: Prospective Randomized controlled trial.

Materials and methods: We are conducting a RCT in women with a previous poor response to ovarian stimulation in an IVF cycle. Patients are recruited upon entry into IVF treatment where a previous cycle had yielded fewer than 4 oocytes (Templeton and Morris 1998), and randomised into the treatment group where patients are treated with 25mg of DHEA three times a day, over four months or no treatment.

The primary end-point is the clinical pregnancy rates and the secondary endpoints include the number and quality of oocytes and embryos generated, markers of ovarian reserves (AMH, FSH, AFC), and ovarian follicular steroidogenesis (estradiol, testosterone, sDHEA and IgF-1).

Results: 58 patients were screened, 34 enrolled. 17 patients have completed the study while 11 are ongoing.

Conclusions: The results from those first 28 patients will be presented in Aspire 2014.

Support: This study is funded by SingHealth Foundation (SHF/CTG034/2010)

P046

ULTRALONG GNRH AGONIST TREATMENT BEFORE THAWED EMBRYO TRANSFER INCREASES CLINICAL PREGNANCY RATE IN THE PATIENTS WITH ADENOMYOSIS.

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Objective: Adenomyosis decreases pregnancy rate in the IVF. Classically ultralong GnRH agonist protocol has been used in the IVF patients with endometriosis or adenomyosis by reducing the size of the tumor. However, there has been rebound enlargement of adenomyosis during COS. The aim of this study was to examine the effect of ultralong GnRH agonist - TET on the pregnancy outcome in the patients with adenomyosis.

Material and Methods:Design: Retrospective observational study- case series

Nine patients with adenomyosis were included in this study. The mean age of the patients was 35.9. 8 patients were treated with Decapeptyl depo 3.75mg every 4 weeks for 2 cycles, and a patient was treated for 3 cycles. After the last dose of Decapeptyl depo, estradiol valerate 4~6mg per day was started. Once the endometrial thickness reached 8mm, vaginal or intramuscular progesterone was started according to the patient's preference. Estradiol and progesterone support was continued for 8 to 10th gestational week according to the state of vaginal bleeding. Clinical pregnancy rate, miscarriage rate, risk of ectopic pregnancy was observed.

Results: 88.9 % (8/9) of the patients showed positive for pregnancy tests. Clinical pregnancy rate was 66.7 % (7/9), and there were a biochemical pregnancy and a miscarriage in the male factor with husband's Sertoli cell only syndrome.

Conclusion: Ultralong GnRH agonist treatment before thawed embryo transfer shows very high implantation rate and good clinical pregnancy rate in the patients with adenomyosis. Future randomized study with large number of patients is needed in the future.

P047

HOW MANY BLASTOCYSTS DO WE NEED? A NEW APPROACH TO THE AGE-SPECIFIC PROBABILITY OF LIVE BIRTH PER BLASTOCYST.

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Advanced maternal age is the most significant risk factor for embryo chromosome aneuploidy, and it is also the main reason to failed implantation and miscarriage. Based on recent researches, transferring good blastocysts is more promising to achieve successful live births than transferring cleavage stage embryos, but seldom studies provided available data to answer the most appropriate number of transferred blastocysts for IVF cases with different ages. Therefore, our aim of this study was to evaluate the probability of every blastocyst transferred to reach the final goal, to have a baby.

There were 827 ET cycles included, 1749 blastocysts transferred, and resulting in 488 newborns, since January 2010 to September 2012.

The pregnancy rate (PR) and live birth rate (LBR) per blastocyst showed a progressively descending behavior with the ascent of individual age: LBR was 34.1% in the cases aged <35 (PR=45.3%), 24.1% in the cases aged 36-38 (PR=34.0%), and 17.8% in those aged 39-40 (PR=27.0%), respectively. While the cases were older than 41 (PR=23.7%), LBR declined to only 14.8%. The p-value to each comparison was <0.0001. In the oocyte recipient group (average recipient age=41.8, average donor age=26.3), the PR and LBR per blastocyst were 51.2% and 36.4%, respectively.

This result is in concert with the fact that the risk of having aneuploid eggs is greatly increased with maternal age. The generated probabilities can be used to evaluate each patient's opportunity to achieve a successful live birth, and helps the IVF centers to decide the number of transferred blastocysts.

P048

IMPROVED OUTCOMES WITH LUTEINIZING HORMONE (LH) SUPPLEMENTATION IN WOMEN ABOVE 35 YEARS OLD UNDERGOING LONG DOWN-REGULATION STIMULATION FOR IVF.

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Introduction: The need for LH supplementation in controlled ovarian stimulation has been debated and remains controversial although LH is believed to play a role in follicle maturation.

Objective: Our primary objective is to determine if oocyte maturation rate (MR) was improved when LH is added during ovarian stimulation for IVF. Secondary outcomes were fertilization rate (FR), clinical pregnancy rate (CPR) and live birth rate (LBR). Patients in recombinant FSH (rFSH) group and rFSH + LH group were further stratified according to age (<35 and ≥35 years) to determine if there was a difference in outcome.

Methods: This is a retrospective review of 324 IVF /ICSI cycles done from year 2009 until 2012. 130 patients were stimulated only with rFSH and 194 patients were stimulated with rFSH+ LH.

Results: Overall, MR was significantly higher in the rFSH+LH group compared to the rFSH only group (79.3% vs 73.3% respectively, $p<0.0001$). There was no difference in FR in both groups.

In patients ≥35 years old, improved outcomes were found in the rFSH+LH versus rFSH alone for CPR (44.6% vs 34.3% $p=0.08$) and LBR (38.5% vs 25.7% $p=0.26$).

In patients <35 years old, better outcomes were found in the rFSH alone vs rFSH+LH group. CPR (51.9% vs 44.3% $p=0.4$) and LBR (43% vs 40% $p=0.7$).

Conclusions: Our data showed an improved oocyte MR when LH was supplemented. CPR and LBR were improved in patients ≥35 years old but not in patients <35 years old indicating that perhaps not all patients require LH supplementation.

P049

PREGNANCY AND PROGESTERONE LEVEL DURING HCG TRIGGER IN EARLY SHORT ANTAGONIST CYCLES

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The early short antagonist (ESA) protocol is being used in majority of women allocated to antagonist protocol in our institution. This retrospective study of patients undergoing controlled ovarian hyperstimulation from December 2012 to May 2013 aims to discover if elevated progesterone levels on the day or day before hCG administration in an ESA cycle will negatively affect pregnancy outcome. 52 women were included. Baseline characteristics were similar between pregnant and non-pregnant. Progesterone levels between pregnant (mean 2.4, SD 0.7584) and non-pregnant patients (mean 2.39, SD 1.013) were similar and not statistically significant. The distribution of progesterone values was almost identical, hence, no suitable cut-off level could be determined to predict a decrease in pregnancy rates after a certain level. Also, there was no progesterone value more than 4.7 nmol/L. Type of gonadotropin used did not show a statistically significant difference in progesterone levels: 2.5 vs 2.3 nmol/L for hMG and rFSH respectively, but the number of patients that received hMG was small. The data shows that there is a positive correlation between progesterone and number of follicles and estradiol level on the day or day before hCG administration. This study suggests that the ESA protocol may have lower progesterone levels than classic antagonist protocols or agonist protocol. A larger sample population can give a more robust conclusion. Progesterone levels did not reach the previously defined threshold value (≥ 4.77 nmol/L) in this group of patients so conclusions regarding the effect of progesterone level on pregnancy cannot be made.

P050

ASSOCIATION OF NUMBER OF CGG REPEATS ON THE FMR1 GENE WITH IVF FAILURE

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Abstract

Infertility is a global problem, and about one out of every seven couples in the world are suffering from infertility. IVF is one of the common ways in which sometimes is unsuccessful. IVF failure have several reason that one of them is genetic factors and the immune system .FMR1 Gene (Xq27.3) with CGG repeat sequence, through effect on immune system one of the genetic factors in IVF failure. The number of iterations is 5 to 54 normal subjects with normal ovarian function of the number

of CGG repeats in the FMR1 26 gene number is 34. This is a normal occurrence of the three genotypes; homozygous, heterozygous makes heterozygous genotype has two modes: repeat one of the two alleles of greater than 34 and lower than 26 sub-genotypes and sub-genotypes het-norm/high created het-norm/low will. The aim of this study was to investigate whether CGG repeats on the IVF failure. In this case-control study 100 women referring to the Sarem women's Hospital during March 2011- March 2012 were divided into 2 groups. Peripheral blood samples of groups analyzed with long PCR method. Case group have a history of at least 3 times IVF failure, normal karyotype and MTHFR. For determine the CGG repeat number used Total Lab. Data were analyzed by spss17. FMR1 genotypes were predictive of pregnancy with IVF ($P = 0/002$), het-norm/low most significantly and with decreasing chance in comparison to norm genotypes.

Key words: CGG repeat, IVF failure, FMR1 Gene, Autoimmunity

P051

A PROSPECTIVE RANDOMISED CONTROLLED STUDY COMPARING A LOW-COST ANTAGONIST PROTOCOL USING ORAL OVULATION INDUCING AGENTS IN IVF-ICSI CYCLES WITH A STANDARD AGONIST LONG PROTOCOL

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Objective: The objective of this prospective randomised controlled study was to compare the cost-effectiveness of a low cost antagonist protocol using oral ovulation induction agents (Clomiphene Citrate or Aromatase Inhibitors) with a standard long GnRH agonist protocol in IVF-ICSI cycles.

Design: IVF-ICSI patients in the study (June 2006 - June 2013) underwent a long GnRH-Agonist or an Antagonist protocol after randomisation using a computer generated list. The antagonist protocol patients were given only oral ovulogens (Clomiphene Citrate or Aromatase Inhibitors) for the first five days of cycle, followed by gonadotropins.

Materials and Methods: A total of 372 IVF-ICSI cycles were prospectively studied in patients less than 40 years of age. All patients received low-dose oral contraceptive pills in the preceding cycle. They were randomised into two groups using a computer generated list. The agonist group underwent the standard long GnRH analogue protocol. The antagonist group received oral ovulation inducing drugs (Clomiphene Citrate / Aromatase Inhibitor) for the first five days of cycle followed by gonadotropins and antagonist.

Results: There was a significant difference in the gonadotropin usage between the two groups. As a result, the cost of the oral ovulation inducing drugs (Clomiphene Citrate / Aromatase Inhibitor) - gonadotropin - antagonist cycle was significantly lower than the long GnRH agonist - gonadotropin protocol. Pregnancy rate per transfer was similar in both groups.

Conclusions: Usage of GnRH antagonist in combination with oral ovulation inducing drugs offers the advantage of an economical method of stimulation in IVF-ICSI cycles, as compared to the agonist protocol, with a similar pregnancy rate.

P052

DIFFERENCES IN ASIAN AND CAUCASIAN CLINICAL OUTCOMES IN IVF: A REVIEW

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Background: It has been reported that Asian women undergoing IVF have poorer outcome compared to Caucasians. However no review has been made to assess outcome differences between the two ethnicities.

Objective: To present and assess current data in ART outcomes between Asian and Caucasian women and to present possible explanations for any disparity.

Design: Best evidence review

Materials and methods: Published studies showing comparisons of ART outcomes between Asians and Caucasians were searched through Pubmed

Results: Thirteen studies were included. There was a consistent finding of lower clinical pregnancy and live birth among Asian women compared to Caucasians in fresh IVF cycles although there was no difference in one egg donor study. Two studies reported higher estradiol levels on the day of HCG trigger among Asians during ovarian stimulation. Similarly, Asians in one study had a higher chance of second or third trimester pregnancy loss and had a higher chance of moderate to severe

growth restriction among singleton births in another. Differences in egg retrieval, embryo development, embryo transfer, and miscarriage were either not significant or had conflicting results. Various theories such as FSH receptor polymorphism, accelerated ovarian aging, educational/ economic differences, and nutritional habits have been proposed to explain these differences.

Conclusions: Asian women have lower clinical pregnancy and live birth rates after IVF compared to Caucasian women, but the differences in other parameters need further investigation. More studies are needed to confirm the current findings and to discover ways to properly manage any disparity in ART outcome.

P053

THE MONOCHORIONIC TWIN PREGNANCY CAN BE DISTINGUISHED FROM SINGLETON PREGNANCY BY EARLY SERUM β -HCG LEVELS IN FRESH IVF-ET?

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Objective: To investigate whether the early serum β -hCG levels in monochorionic twin pregnancies are different from those of singleton pregnancy in fresh IVF-ET cycles.

Materials and methods: We retrospectively reviewed medical records of fresh IVF-ET cycles such as early serum β -hCG levels, and their outcomes especially focused on number of gestational sacs and fetal heart beat. 17 monochorionic twin pregnancies which resulted in ongoing pregnancy were included. Randomized selected 36 dichorionic diamnionic (DCDA) pregnancies and 36 singleton pregnancies which resulted in ongoing were included as control. The serum β -hCG levels at 12 and 14 days after ovum retrieval were compared between singleton, DCDA and monochorionic twin pregnancy group. The degree of increase of serum β -hCG levels in two days were also compared.

Results: The mean serum β -hCG levels checked at 12 days after ovum retrieval in singleton, DCDA and MCDA pregnancies were 53.1 ± 26.3 , 108.5 ± 52.2 , 63.5 ± 30.5 mIU/mL each. The mean serum β -hCG levels checked at 12 days after ovum retrieval in monochorionic twin and singleton pregnancies were significantly lower than that of DCDA pregnancies ($p < 0.05$). The serum β -hCG levels at 12 days and 14 days after ovum retrieval have a tendency to high in monochorionic pregnancies than in singleton pregnancies, but the difference was not significant ($p = 0.715$).

Conclusion: The early serum β -hCG levels seems to have no value in prediction of monochorionic twin pregnancy in fresh IVF-ET program.

P054

RELATIONSHIP BETWEEN MILD OVARIAN STIMULATION AND IVF OUTCOMES IN NORMO-RESPONDER GROUP

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Objective: Controlled ovarian hyperstimulation (COH) could make several side effects in IVF cycles. COH could also influence adverse effects on implantation by supraphysiological hormone profile. Therefore mild ovarian stimulation by reducing dose of stimulation drugs has been an important issue in recent IVF program. The purpose of this study is to assess the effects of mild ovarian stimulation by reducing starting dose to pregnancy rates.

Design: A retrospective study

Materials and Methods: We retrospectively reviewed 267 patients undergoing IVF using GnRH agonist and antagonist protocols from June 2010 to December 2011. Our inclusion criteria was $3 < \text{serum anti-mullerian hormone (AMH)} < 6$ ng/ml. Two groups of patients were formed based on starting dose of rFSH (A - conventional IVF group : 150 IU start vs. B - mild stimulation IVF group : 112.5 IU start).

Results: Total rFSH dose of group A was higher than that of group B [1410 IU vs. 1202 IU, $P < 0.01$]. Number of oocytes, 2PN, transferred embryo were similar between the two groups. Group B had significantly higher pregnancy rates than group A [52.7%(29/55) vs. 45.8%(97/212), $P < 0.05$].

Conclusions: In normo-responder group, higher pregnancy rates were achieved by reducing starting dose. Mild ovarian stimulation is important in order to decrease several side effects. In addition, mild ovarian stimulation by reducing starting dose has significant effects on pregnancy rates in normo-responder group. Therefore, clinician should make an effort to reduce dose of stimulation drugs.

P055

DEFINING THE IMPACT OF ASSISTED REPRODUCTIVE TECHNOLOGIES (ART) ON THE PLACENTAL EPIGENOME

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Introduction

Over the past three decades, some studies have suggested an association between ART and an increased risk of rare imprinting disorders, such as Beckwith-Wiedemann syndrome. The effect of ART on the placental epigenome has been the topic of lively debate in recent years, with evidence that ART may be associated with altered epigenetic status in the placenta. These changes could potentially disrupt placental function, impacting on the health of the offspring. Most placental epigenetic studies to date have focused on DNA methylation. Other types of epigenetic marks include histone modifications, which have a number of functions including the regulation of gene expression.

Methods

In this study, we are using chromatin immunoprecipitation (ChIP) combined with ultra-high throughput next generation sequencing technology to explore the potential impact of ART on histone modifications in term placentae. These data will be integrated with transcriptome profiling to determine any correlation with allele-specific expression e.g. imprinting.

Results

In our initial ChIP assays we have found that histone modifications in placentae stored at 4°C after delivery are stable for at least 24 hrs. There was no significant difference in histone modification levels at targeted loci when analysing different samples from the same placenta. Our first analysis of placental gene expression focussed on known imprinted genes. There was no significant difference in allele-specific expression at these loci in ART versus control placentae. Sample collection and data analyses are ongoing.

Conclusion

Our preliminary analyses have not identified any effect of ART on allele-specific expression in the placenta.

P056

DELAYED HUMAN CHORIONIC GONADOTROPINE (HCG) ADMINISTRATION RESULTS IN LOWER CLINICAL PREGNANCY RATE (CPR) IN GnRH AGONIST CYCLE

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The administration of hCG in GnRH agonist stimulated cycles may sometimes be postponed in order to maximize the number of competent oocytes collected. In this retrospective study, we aim to assess the effect of delayed hCG trigger on the number of mature oocytes collected and CPR in GnRH agonist cycles.

One hundred GnRH agonist cycles managed solely by one clinician in 6 months time were analyzed. The follicle sizes were monitored via transvaginal ultrasound scan, and the timing of hCG administration was accordingly adjusted to maximize the number of mature oocytes collected. Final oocyte maturation was triggered with 10,000IU recombinant-hCG. The hCG trigger is considered as delayed if it was not administered on the first day when the standard trigger criterion (3 follicles ≥ 18 mm) was met. Patients were separated into 3 groups according to the day of hCG administration; Group A= on the same day/one day after, Group B= two days after, and Group C= three or more days after the trigger criterion was met. The mean age and mean number of embryos transferred for all groups were similar.

Result: The CPR were comparable for Group A (62.2%) and Group B (58.1%), but significantly lower (25%, $P=0.02$) in Group C. The mean number of mature oocytes retrieved did not differ between group A (10.8), group B (13.0) and group C (14.5).

Conclusion: More than 2 days delayed in hCG administration does not improve the number of mature oocytes retrieved in GnRH agonist cycles but is associated with significantly lower CPR.

P057

FEMALE FACTORS ASSOCIATED WITH BLASTOCYST FORMATION IN WOMEN UNDERGOING IVF

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Background: Blastocyst transfer has been known to increase implantation rate. Several clinical factors have been associated with good quality blastocyst, such as patient's age, oocytes quality, sperm quality and etiology of infertility.

Aim: The aim of this study is to determine female factors correlated with blastocyst formation in IVF. The factors are female age, AMH level, and female cause of infertility.

Method: This is a cross sectional study on patients underwent IVF in Yasmin Clinic, Dr. Cipto Mangunkusumo General Hospital, Jakarta from January 2012 to June 2013. Data on blastocyst formation on day 5th, female age, serum AMH, and female cause of infertility were collected. Both of variables were categorized as follow: ≥ 30 years vs < 30 years for age, and $1.2 \text{ ng/ml} - 4.13 \text{ ng/ml}$ vs $< 1.2 \text{ ng/ml}$ & $> 4.13 \text{ ng/ml}$ for serum AMH. Patients with male factor were excluded from this study. Data analysis was performed with SPSS version 11.0.

Results: We obtained 52 patients with a mean age of 33.1 ± 4.2 years. Thirty-three patients (63.5%) developed blastocysts and 19 (36.5%) did not. Serum AMH has significant association with blastocyst formation ($p=0.001$). Female age was not associated with blastocyst formation ($p=0.331$). Bivariate analysis of female cause of infertility revealed that poor responders ($p=0.044$) and diminished ovarian reserve ($p=0.004$) were significantly associated with lower blastocyst formation.

Conclusion: Serum AMH, poor responders and diminished ovarian reserve significantly associated with blastocyst formation. Female age was not associated with blastocyst formation.

Keywords: blastocyst, serum AMH, patient's age, female infertility

P058

DOES DAY 3 EMBRYO CELL NUMBER PREDICT ANEUPLOIDY?

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Despite significant advances in assisted reproduction, only a small number of embryos that are selected for transfer will result in a viable pregnancy and progress to delivery, particularly in women of advanced maternal age or with reduced ovarian reserve. The use of preimplantation genetic screening significantly alters the methods by which embryos are selected for transfer compared to traditional assessment which incorporates cell number and morphology. This study assessed whether embryo cell number on day 3 was predictive of aneuploidy following preimplantation genetic screening.

Data was retrospectively analysed for women undergoing IVF with ICSI and preimplantation genetic screening. Day 3 cell number was correlated with final screening results to determine if day 3 embryo development is predictive of aneuploidy. Results were also analysed based on maternal age to determine if this altered the outcomes seen.

A total of 773 embryos were analysed to compare cell number on day 3 with aneuploidy status following biopsy and vitrification. Day 3 embryo cell number was not different between embryos that were found to be euploid versus aneuploid (8.5 ± 0.15 and 8.3 ± 0.07 respectively). Furthermore, maternal age did not alter the results seen.

Assessment of day 3 embryo cell number is not indicative of ploidy status. The results from this study may be useful in adapting cleavage stage embryo grading systems which downgrade embryos based on selected developmental criteria. It is possible that other embryo morphological criteria such as fragmentation or uneven cell division may be indicative of embryo ploidy, however this remains to be investigated.

P059

DOES SPERM QUALITY AFFECT BLASTOCYST FORMATION IN IVF ?

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Background: The role of paternal factors in embryonic development remains unclear. Some studies show that sperm quality did not affect blastocyst development whereas others did.

Aim : To determine sperm quality in blastocyst formation as the outcome of IVF success.

Method: We conducted a cross sectional study in Yasmin IVF Clinic-Dr. CiptoMangunkusumo General Hospital from January 2012 to June 2013. Female factors associated with lower oocyte quality were excluded from this study. Data on sperm concentration, motility, DNA integrity and blastocyst formation on day 5 were collected. Sperm quality was classified into low vs normal concentration (<15 million/mL vs ≥15 million/mL), immotile vs motile sperm (<40% vs ≥40%) and poor vs good DNA Fragmentation Index (DFI) (≥30% vs <30%). Data was analyzed with SPSS version 11.0.

Result: The data obtained from 56 patients showed mean age of 36 ± 6 years old. The mean values for sperm concentration was 39 ± 52.7 million/mL and 42.8% ± 24.7% for sperm motility and 27.3% ± 19.8% for DFI. Blastocyst formation appeared higher with normal sperm concentrations (56% vs 44%) and motile sperm (52% vs 48%). However, analytical study showed that there were no significant associations between sperm concentration (p= 0.931), motility (p=0.23) and DFI (p=0.200) with blastocyst formation.

Conclusion: Sperm DNA integrity as one of the parameter of sperm quality did not appear to influence blastocyst formation, although blastocyst formation was higher in groups with normal sperm concentration and good sperm motility.

Keywords: *Blastocyst, Sperm concentration, Sperm motility, DNA Fragmentation Index*

P060

ANALYSIS OF CYCLE OUTCOMES, BY DAY OF TRANSFER AND EMBRYO GRADING IN IVF/ICSI CYCLES WITH ONLY ONE FERTILISED OOCYTE.

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Objective

To compare cycle outcomes including clinical pregnancy, pregnancy loss and live birth rates in patients with only 1 fertilised oocyte.

Methods

A retrospective analysis of pregnancy outcomes for patients with a single fertilised oocyte were compared to determine if there was any benefit in extended culture between cleavage and blastocyst groups. Developmental outcomes were assessed in relation to embryo quality and embryos were split into two prognostic groups identified as poor and good graded embryos. These were respectively for day2 (n = 88; n= 98) day3 (n= 219; n= 285) and day5 (n=43; n=26). Two sided t tests were used to determine statistical significance.

Results

The timing of embryo transfer did not produce significant differences in CP outcomes between the poor prognosis (Day2: 4%, Day3: 6%, Day5: 0%) and good prognosis group (Day2: 22%, Day3: 20%, Day5: 28%). In 26 cases, embryos that were considered to have good development on day3 and cultured to day 5 there was a trend in higher CP and live birth outcomes as well as a lower rate of fetal loss in the good prognosis day5 blastocyst group from CP to live birth ((day2: 25%); (day3: 8%) (day5: 14%)).

Conclusion

Pregnancy outcomes in patients with one fertilized embryo were not significantly different between transfer groups for both developmental demographics. Importantly patients with good quality embryos appeared to have a lower miscarriage rate when embryo transfer occurred on day 5 compared to a day 2 or 3 transfer with minimal risk of failed transfer.

P061

LOW MOLECULAR WEIGHT HEPARIN DOES NOT AFFECT THE FORMATION OF PLACENTAL GROWTH FACTOR

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Introduction

Administration of LMWH is associated with the increase of *Placental Growth Factor* (PIGF) that promotes trophoblast invasion, enhances the placentation, and decreases pregnancy complication risk, such as pre-eclampsia or IUGR. The aim of this study is to evaluate the effect of LMWH on the formation of PIGF in IVF patients.

Methods

A cross-sectional study was performed at Yasmin Reproductive Clinic, dr.Cipto Mangunkusumo General Hospital. A total of 69 IVF patients receive subcutaneous LMWH 0.4 mg daily, for 2 weeks after embryo transfer. There were 23 pregnant patients and PIGF level was measured from pregnant patients.

Results and Discussion

Distribution of PIGF value from pregnant patients ranged between 9.5 to 16.6 pg/ml within 4th-5th week of gestation. This value is not significantly different compared to PIGF reference value (18-48 pg/ml, 63 ± 145 pg/ml and <50 pg/ml).

Conclusion

Administration of LMWH does not significantly affect the level of PIGF.

Keywords: LMWH, PIGF, IVF

P062

IVF OUTCOME IN PRE-STIMULATION SHORT TERM TESTOSTERONE PATCH APPLICATION IN UNEXPLAINED POOR RESPONDERS

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Introduction: Treatment of unexplained poor responders appears to be a challenging task since this group constitutes women undergoing IVF with apparently normal ovarian reserve tests but show poor ovarian response upon stimulation. This series of case studies report the usefulness of testosterone patch with long protocol in women who showed a poor ovarian response despite adequate gonadotropin stimulation in the previous cycle.

Material and methods: 9 consecutive women with unexplained poor response in the previously failed IVF cycle were treated between Nov 2010 to May 2011, with long protocol along with application of trans-dermal testosterone patch 5 mg for 12 hours from day 1 to 5 of starting of menstruation followed by standard gonadotropin stimulation and subsequent IVF-ICSI.

Results: Mean age and basal FSH of the 9 patients in the study was 34.4 ± 3.97 and 10.56 ± 4.35 respectively. Oocytes were retrieved in only 5 out of 9 patients in the previous cycle whereas all 9 patients had oocytes retrieved in the treatment cycle. The mean number of oocytes retrieved per stimulation in the present cycle (4.22 vs 2.0; $P=0.102$) was twice as higher than that of the previous cycle. An overall pregnancy rate of 44.4% per oocyte retrieval with 33% live birth rate was achieved after embryo transfer in the testosterone treated cycles.

Conclusion: Testosterone patch application as pre-treatment to ovarian stimulation appears to improve the number of oocytes retrieved and embryos available for transfer and a reduced cycle cancellation with a reasonably good pregnancy rate in unexplained poor responders.

P063

DOES DURATION OF E2 PRIMING AFTER GONADOTROPHIN-RELEASING HORMONE AGONIST (GnRHa) SUPPRESSION IN A FROZEN EMBRYO TRANSFER (FET) CYCLE AFFECT THE OUTCOME?

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Introduction: After downregulation using GnRHa, induction of endometrial receptivity can be achieved using estrogen and progesterone (P) supplementation. Endometrial proliferation and induction of P receptors can be stimulated by E₂ supplementation while subsequent P stimulation induces endometrial receptivity.

Objective: Our primary objective was to evaluate if there is a difference in clinical pregnancy rate per embryo transfer (CPR) and live birth rate per embryo transfer (LBR) in relation to the length (days) of estradiol (E₂) priming.

Methods: This is a retrospective review of 142 FET cycles done from 2012 until June 2013. Oral E₂ for endometrial priming was given for 16-26 days and vaginal P was started 3-4 days before ET. All embryos were vitrified at cleavage stage and transferred after warming. All patients included had an endometrial thickness of >6 mm before transfer.

Results: For analysis, data were grouped into 4 groups according to duration of E₂ priming before transfer. E₂ priming was carried out for: <18 days (Group A, n=6), 18-20 days (Group B, n=71), 21-23 days (Group C, n=60), ≥ 24 days (Group D, n=5).

The CPR and LBR in all groups were (0%, 39.4%, 55%, 0%) and (0%, 32.3%, 45%, 0%) respectively. There were no statistical significance between groups.

Conclusions: Although there was no statistical significance, we observed an increased CPR and LBR when E₂ priming was carried out for 21-23 days before ET perhaps because the optimal duration of estrogen stimulation was achieved before progesterone was added to prime the endometrium for implantation.

P064

INCIDENCE AND ASSOCIATED FACTORS INFLUENCING OVARIAN HYPERSTIMULATION SYNDROME (OHSS) IN DR. CIPTO MANGUNKUSUMO HOSPITAL, INDONESIA FROM 2006 TO 2013

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Introduction: Ovarian hyperstimulation syndrome (OHSS) is one of the complications of in vitro fertilization (IVF). The risk factors involved are still not very well-understood and clinical studies are still performed to find out about them.

Objective: To evaluate the profile of OHSS among patients who are treated with IVF in Yasmin Clinic, Kencana Dr. Cipto Mangunkusumo Hospital from 2006 to 2013.

Methods: The study is a descriptive study of 1088 ART cycles with several factors observed based on previous studies

Results:

Incidence of OHSS in Yasmin Clinic is 0.46% (5 patients). Factors associated with OHSS are: patient's age (mean 31.4 years old, SD 2.47), husband's age (mean 34.4 years old, SD 4.67), BMI (27.34 - 21.48 kg/m²), etiology (4 case of primary infertility and 1 case of male factor), duration (7.4 years), basal FSH (median 5.1 mIU/mL (4.5 - 6.5 mIU/mL)), basal AMH (median 6.1 mIU/mL (5.3 - 11 mIU/mL)), basal LH (median 7.5 mIU/mL (4.1 - 16.6 mIU/mL)) and E₂ (median 27 pg/mL (5.3 - 76 pg/mL)), treatment protocol (4 long and 1 short protocols), rFSH (median 2,250 IU (2.025 - 2.625 IU)), rLH (median 0 (0 - 450 IU)), GnRH agonist (median 2.25 IU (0 - 3 IU)), and mature oocyte (median 30 (5 - 36))

Conclusion:

The incidence of OHSS among the population is similar to reported incidence from previous studies (0.5% - 5%). Factors associated with OHSS are age, BMI, infertility's etiology, basal levels of FSH, AMH, LH and E₂, total rFSH, rLH and GnRH agonist and mature oocyte harvested

Keywords: OHSS, incidence, profile, factors

P065

FRESH EMBRYO TRANSFER AS A RISK FACTOR FOR ECTOPIC PREGNANCY

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Background: Patients undergoing ART appear to be at increased risk of ectopic pregnancy compared to women conceiving spontaneously. Some evidence suggests that the increased risk is related to ovarian stimulation, and ectopic pregnancy rates are reduced in women having a frozen embryo transfer compared to a fresh embryo transfer.

Aim: To investigate the incidence of ectopic pregnancy in fresh embryo transfers vs frozen embryo transfers

Methods: 7,600 pregnancies at a single IVF provider over a 10-year period are analysed for ectopic pregnancy. Fresh transfer versus frozen transfer, female age, total oocytes retrieved and total FSH dosage are also recorded where appropriate.

Discussion: This retrospective observational study assesses pregnancies at a single IVF provider over a 10-year period. The risk of ectopic pregnancy is further analysed according to level of stimulation as defined by total FSH dose and oocytes retrieved.

P066

CLINICAL PREGNANCY FOLLOWING AMNION TRANSPLANTATION ON THIN ENDOMETRIUM PATIENT IN IVF PROGRAM : A CASE REPORT

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Human amnion membrane has been widely used as biologic dressings for ophthalmology, plastic surgery, dermatology and gynecology procedures. Human amnion membrane displays anti-microbial, anti-tumorigenic, anti-fibrosis and anti-angiogenic properties. In vitro studies have demonstrated that cells derived from amnion membrane do not trigger immuno-allogenic or xenogenic responses and actively suppress the proliferation of T lymphocytes. These cells express the non-classic, less polymorphic HLA-G molecules and lacks of high polymorphic antigens HLA-ABC and HLA-DR.

Moreover, primary human amnion epithelial cells (hAEC) isolated from term placenta have stem cell-like properties including the expression of pluripotent human embryonic stem (hES) cells markers Sox-2, Oct-4 and Nanog, clonogenicity and differentiate into cell lineages *in vitro* and *in vivo*.

Endometrial receptivity plays an important role in the success of in vitro fertilization (IVF) program. Impairment on endometrial receptivity will prevent the transferred embryos from implantation. Endometrial thickness is one of the indicator for predicting appropriate endometrial receptivity. Endometrial thickness less than 7 mm or 'thin endometrium' correlates with lower pregnancy rate in IVF program. It has been hypothesized that thin endometrium may caused by insufficient function / numbers of endometrial stem cells which responsible for endometrial regeneration capacity. There is no effective treatment for improving endometrial thickness on patients with thin endometrium.

In this study we reported a case of clinical pregnancy following intra uterine amnion transplantation for patient with thin endometrium in a short protocol IVF program.

P067

COMPARATIVE STUDY OF VAGINAL CABERGOLINE VERSUS INTRAVENOUS ALBUMIN FOR EARLY ONSET SEVERE OHSS PREVENTION IN ART CYCLE

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Objective : To compare the efficacy of vaginal cabergoline versus intravenous albumin in severe early onset OHSS prevention.

Methods : There were 60 cases of high risk OHSS divided into two groups ,30 cases cabergoline group and 30 cases of albumin group.The inclusion criteria were patient with number of preovulatory follicle \geq 15, metaphase II oocyte retrieved \geq 15 ,estradiol \geq 4000 pg/ml The characteristic of age ,BMI ,duration of infertility were compared between both groups. Stimulation and laboratory data ,total preovulatory follicle of hCG day ,Estradiol level on hCG day ,number of oocyte retrieved ,number of good embryo and number of embryo transferred were compared between both groups. Vaginal Cabergoline 0.5 mg daily for 8 days was given starting from oocyte retrieval day in cabergoline group and 100 ml intravenous albumin 20% has been given on oocyte retrieval day in albumin group .Early severe OHSS incidence ,hospital admission ,clinical pregnancy rate ,miscarriage rate were compared between both groups.

Results:There were no characteristic ,stimulation and laboratory data differences in two groups .There was a reduction in incidence of early onset severe OHSS in cabergoline group compare to albumin group ($p < 0.05$) .Admission rate was also reduce in cabergoline group (< 0.05). Clinical pregnancy rate and miscarriage rate were similar in both group ($p > 0.05$).

Conclusion: Vaginal cabergoline is an effective and safe medication for prevention early onset severe OHSS.

Keyword : vaginal cabergoline ,OHSS

P068

CLINICAL AND ECONOMICAL EFFECTIVENESS OF MILD OVARIAN STIMULATION COMBINING CLOMIPHENE CITRATE AND MID TO LATE FOLLICULAR GONADOTROPIN FOR ART

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Objective: The conventional ovarian stimulation is expensive, and associated with significant morbidity and stress. Interest and demand for patient-friendly stimulation protocols is increasing. Effectiveness of mild ovarian stimulation was studied.

Stud design: Retrospective analysis of ART clinical data at a single institution

Material & Method: 41 conventional GnRH antagonist cycles (c-ANT) commencing gonadotropin at early follicular phase and 63 mild-stimulation GnRH antagonist cycles (m-ANT) combining early follicular clomiphene citrate and mid to late follicular gonadotropin were compared. GnRH antagonist was flexibly used in both groups. All were less than 40 year old and were first ART attempt. IRB approved the study.

Results:

Study group	Female age	AMH (pM)	Total gonadotropin dose (unit)**	E2 before trigger (pg/ml)*	Retrieved eggs**	Cleavage stage embryos**	G1-2 embryos*	G1-2 per cleavage stage(%)*
c-ANT(n=41)	36.2+/-3.2	21.7+/-16.3	1161+/-506	969+/-379	7.3+/-5.2	4.5+/-2.9	2.1+/-1.9	45.7+/-33.5
m-ANT(n=63)	35.7+/-3.4	20.5+/-17.4	502+/-197	795+/-427	3.5+/-2.5	2.1+/-1.5	1.3+/-1.2	63.2+/-39.1

+/-:S.D., *:p<0.05, **:p<0.005

Comparing c-ANT and m-ANT, number of transferred embryos (average 1.2 vs. 1.0) was similar. But clinical pregnancy rate (18.8 vs. 29.2%) and embryo implantation rate (17.9 vs. 29.2%) showed higher trend in m-ANT. Clinical pregnancy per oocyte retrieval was not statistically different (39.0 vs. 44.4%, p=0.64). No significant morbidity was reported in the both groups.

Conclusions: Compared to conventional antagonist stimulation, our mild stimulation protocol reduced cost of treatment but did not compromise clinical outcomes of ART. Mild stimulation yielded significantly higher fraction of good quality embryos. This approach can be the first choice of ovarian stimulation for patients with good ovarian reserve.

P069

THE EFFECT OF USING HYALURONAN-ENRICHED TRANSFER MEDIA IN HUMAN EMBRYO TRANSFERS

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Aim:

To evaluate the impact of hyaluronan-enriched transfer media (HETM) on implantation rates (IR) for human embryos in a clinical setting after fresh or frozen-thawed embryo transfers (FET). Secondary to this, we also investigated the effect of HETM in subgroups of cleavage and blastocyst stage embryos.

Method:

We retrospectively analysed 293 embryo transfer cycles undertaken between January 2012 and July 2013 using a binomial model and logistic regression. A total of 182 embryo transfer cycles using HETM (UTM, Origio) were compared with 111 embryo transfer cycles undertaken immediately prior to the introduction of HETM (control group). These groups were then divided into two subgroups based on whether fresh or frozen-thawed embryos were transferred. Within each of these

subgroups further comparison of IR was undertaken based on the stage of embryo development, namely cleavage stage or blastocyst stage.

Results:

When comparing fresh and FET cycles, we found a significant interaction between the transfer type and HETM ($p < 0.05$), resulting in increased IR for FET using HETM. When comparing cleavage stage versus blastocyst stage transfers, we found no statistically significant difference in IR between the HETM and the control group, for fresh or frozen cycles.

Conclusion:

The use of HETM resulted in a significantly higher IR in the FET subgroup only. Overall this data showed a difference in IR when using HETM; however this was evident only in certain subgroups.

P070

IS GNRH ANTAGONIST PROTOCOL BETTER IN PCOS? A META-ANALYSIS OF RCT STUDIES

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Abstract

Objective

To review published RCT studies evaluating the IVF/ICSI outcome of utilization of gonadotrophin-releasing hormone antagonists (GnRH-ant) for ovarian stimulation in PCOS compared with classic luteal long agonist protocol.

Design

A meta-analysis of prospective randomized trials published in English between 2002 and 2013.

Patient(s) and interventions

Nine RCT studies of PCOS undergoing IVF/ICSI including 588 women performed long agonist protocol and 554 women with GnRH antagonist protocol.

Main Outcome Measure(s)

Clinical pregnancy rates (CPR), ongoing pregnancy rate and OHSS rate.

Result(s)

Nine randomized clinical trials were included in this analysis. Clinical pregnancy rate per embryo transfer was similar in two groups (RR: 0.97, 95% CI: 0.85–1.10). Non-significant estimates comparing two protocols were found for age, BMI, total dose of gonadotropin consumed, stimulation days and number of oocytes retrieved. Meta-analysis for 4 RCTs concluded that GnRH antagonist protocol is better than agonist protocol on reduction of severe OHSS rate (OR 1.56; 95% CI 0.29–8.51).

Conclusion(s)

At the point of CPR, GnRH antagonist protocol is similar to GnRH long agonist protocol. However, for the severe OHSS, GnRH antagonist protocol is obviously better in PCOS.

P071

APLASIA CUTIS CONGENITA IN MALE BABY , PRODUCT OF POST ICSI TRIPLETS REDUCED TO TWINS AT 11 WEEKS,BORN TO 40F AT 33WEEKS

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Aplasia cutis congenita is a heterogeneous disorder with incidence of 1 in 10000. Most cases are sporadic but familial cases occur with autosomal dominant or recessive patterns. It has been described in Twin pregnancy where one twin died in early pregnancy. We describe here a case of male baby born with bilateral symmetrical absence of skin on trunk with no other anomaly to a 40 yr old female at 33 wks. The pregnancy was an outcome of post ICSI triplets with egg donation. Fetal reduction was done at 11 weeks with Potassium chloride. Co-twin was a female who had no congenital anomaly. Family history of biological father and mother was unsuggestive. The baby was under care of Plastic surgeon who managed conservatively by repeated dressings. Reported cases have been in twins with fused placentae supporting the vascular

disruption pathogenesis behind it. After fetal reduction for multiplicity no case has been reported. Since there was no other anomaly it may be a sporadic occurrence not related to the procedure of fetal reduction. More studies are needed to establish any correlation

P072

COMPARISON OF OPTIMAL TIME FOR HUMAN EMBRYO TRANSFER BETWEEN CULTURE DAY 3 AND DAY 4

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During ART, the decision of culture day for embryo transfer has been considered as an important factor. Several researches have suggested eight cell-stage ET (day 3) and blastocyst-stage ET (day 5) could get favorable pregnancy outcomes than other cell-stage such as sixteen cell-stage ET (day 4). The aim of this study was to compare the implantation rates of day 3 and 4 embryo transfers in randomly infertility patients.

This study was retrospective case study. Total 580 patients treated by IVF/ICSI at the department of clinical laboratory were included for January to August 2013. For embryo transfer (ET), good quality embryos were defined as having 8 blastomeres (day 3)/ 16 blastomeres (day 4) of normal size, a maximum of 20 % of anucleated fragments, and no multinucleated blastomeres. To assess of implantation rate, serum β -hCG concentrations were measured 14 day after ET, with a ≥ 10 mIU/mL increase in serum β -hCG regarded as positive for implantation. Overall, the culture day 4 ET showed higher implantation rate than culture day 3 ET, but there was not significant differences in the implantation rate between culture day 3 ET and culture day 4 ET. The rate of serum β -hCG positive was 29.3 % in culture day 3 and 38.8 % in culture day 4, respectively. As a result, culture day 3 and 4 ET had similar implantation rate in this study. However, the present study was small in case and will need more specific investigation such as clinical, ongoing pregnancy rate and miscarriage rate.

P073

THE COMPARISON OF EMBRYO DEVELOPMENT BETWEEN TIME LAPSE INCUBATOR AND STANDARD INCUBATOR USING SIBLING OOCYTES

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Objectives

Using of the time lapse incubator can analysis useful information of the embryonic developmental events morphologically by the automatic capture of images. However, the safety of the embryos cultured in time lapse incubator has not proven clearly. The aim of this study was to evaluate the safety of the culture in the time lapse incubator by comparing pronuclear formation and development of embryos between ES (EmbryoScope, Unisense FertiliTech, Denmark) and SI (Standard Incubator) culture by using sibling oocytes.

Methods

The prospective study was drawn from a total of 344 oocytes from which 276 embryos were generated in 24 cycles from June to October 2013. The sibling oocytes were randomized to analyze the pattern of the embryos development depending on the incubator (ES vs. SI) and all the embryos were derived ICSI.

Results

No differences ($P>0.05$) were found in the rates of 1PN (4.6% vs. 2.7%) and 3PN (2.1% vs. 6.0%) between ES and SI. However, 2PN rates were 82.1% in ES and 77.9% in SI, higher in ES compared to SI, statistically ($p=0.048$). Contrast of high quality cleavages rate day 3 for ES and SI were 38.1% and 34.5%, respectively and did not differ significantly ($p=0.108$). The proportion of good blastocyst at day 5, 6 were 11.3% in ES and 10.3% in SI, there was no statistically significant difference, $p=0.372$.

Conclusions

The study has shown that culture in the time lapse incubator supports embryonic development equally to the standard incubator and may therefore be safely utilized in clinical practice.

P074

THE COMPARED EMBRYO CULTURE CONDITION IN EMBRYOSCOPE OR STANDARD INCUBATOR BY CLINICAL OUTCOME

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Object:

Time-lapse monitoring gives useful information for embryologist to capture images of embryo at defined interval time in an incubator. Even through time-lapse monitoring system has a benefit to culture embryo, the safety of the cultured embryo in Time-lapse monitoring system has been controversial because embryos were periodically exposed to light. In the current study, it was investigated culturing conditions in EmbryoScope by fertilization, embryo development, and pregnancy.

Methods:

In this study, we analyzed 59 patients who came to Maria Hospital undergoing IVF from July to October 2013. Oocytes were performed to ICSI and cultured in Cook media. They were randomly divided into two groups, one was cultured in EmbryoScope™ (ES, 31 cycles) and the other was cultured in standard incubator (SI, 28 cycles). Embryo selection criteria were based on a morphology basis. Transferred embryos from ES were excluded multinucleated or irregular divided embryos.

Results:

Patient's characteristics were not different between them. Retrieved oocytes and fertilized embryo development, such as cleavage and day 3 good quality embryos, were similar except blastocysts formation. It was significantly higher in ES than in SI (38% vs 25%, $p=0.002$). The number of transferred embryos were significantly low in ES. Clinical pregnancy and implantation rates of ES were 57% and 44% tended to be higher than them of SI (39% and 31%), although not statistically significant.

Conclusion:

Even though the number is limited, it is useful system because it provides stable culture conditions and morphokinetic parameters to select embryo to get increasing clinical outcome.

P075

THE EFFICACY OF LONG ACTING FSH FOR CONTROLLED OVARIAN HYPERSTIMULATION: COMPARISON WITH SHORT ACTING FSH

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Objective

To compare the efficacy of long acting type FSH and short acting type FSH in GnRH antagonist cycles for IVF ET program.

Materials and Methods

We reviewed the our medical records retrospectively. 53 infertility patients who undergone fresh IVF-ET cycles with GnRH antagonist protocol by using long acting gonadotropin were included as study population. Age matched 106 patients who undergone IVF-ET cycles by using short acting gonadotropin were included as control. IVF outcomes such as rates of oocyte maturation, implantation, biochemical and clinical pregnancy, spontaneous abortion and incidence of OHSS were compared between two groups.

Results

Among the clinical characteristics of each patient, the serum estradiol level of 5 to 7 days after stimulation was significantly higher in long acting FSH used group (590.8 ± 40.6 pg/ml vs 985.5 ± 92.5 pg/ml, $p=0.00$). Duration of COH, rate of cancellation, incidence of OHSS, oocyte maturation and fertilization were similar in both groups. Notably, implantation rate in long acting FSH used group was significantly higher than that of short acting FSH used group (11.6% vs 22.8%, $p=0.03$). But, the biochemical pregnancy rate (6.9% vs 16.7%, $p=0.081$) and clinical pregnancy rate (27.6% vs 42.9%, $p=0.064$) showed tendency to higher in long acting FSH group. The clinical abortion rate was not significantly different between two groups.

Conclusions

Long acting FSH seemed to be as effective as short acting FSH for COH in antagonist cycles. But, despite of absence of statistically significant differences, tendency to increased early pregnancy loss rate should be clearly defined in long acting FSH used group.

P076

TWIN PREGNANCY AND SUCCESSFUL DELIVERY AFTER TRANSMYOMETRIAL EMBRYO TRANSFER.

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• Objective

To report an unusual case of congenital cervical atresia and primary infertility, achieving twin pregnancy through transmyometrial embryo transfer (ET) resulting in the delivery of healthy twins.

• Design

Case report and literature review.

• Setting

Private infertility center at Saudi Arabia.

• Intervention(s)

Controlled ovarian hyperstimulation, oocyte retrieval, in vitro fertilization, transmyometrial ET.

• Patient

26 yrs old lady para 0, she had history of cyclical abdominal pain since menarche, diagnose as case of congenital malformation of the genital organs, she had laparotomy for drainage of haematometra/haematocolpos, ovarian cystectomy and cervico- plasty at other hospital. Also she had history of failed IVF cycle and difficult embryo transfer.

• Main Outcome Measure(s)

Successful delivery after transmyometrial ET.

• Result(s)

A IVF cycles and transmyometrial ET were performed, a twin pregnancy was achieved. Cervical cerclage was done at 13 weeks of pregnancy, two healthy babies delivered at 35+4 weeks gestation.

• Conclusion(s)

With careful selection of patients and procedure, successful pregnancy is possible in similar patients.

P077

NUMBER OF OOCYTES RETRIEVED DOES NOT AFFECT CLINICAL PREGNANCY RATE PER EMBRYO TRANSFER (CPR/ET) AND LIVE BIRTH/ONGOING PREGNANCY RATE (LB/OGPR) IN NON-DONOR IVF CYCLES

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Design: Retrospective analysis of all non-donor IVF cases performed from 1st January 2012 to 31st June 2013. Patients with 11-20 oocytes retrieved (Group 1, n=52) were considered as having optimum number of oocytes based on the outcome found in previous studies (Verberg *et al*, 2009, Ji *et al*, 2013). The CPR/ET and LB/OGPR as well as embryo transfer (ET) cancellation rate for patients who had <11 (Group 2, n=20) and >20 (Group 3, n=35) oocytes retrieved were evaluated and compared to group 1. The mean age and mean number of embryo transferred were similar among the three groups.

Results: In group 1, forty-seven patients had ET (cancellation rate, CR=9.6%) giving a 68.1% CPR/ET and 59.6% LB/OGPR. Twenty-three patients (CR=4.8%) in group 2 had ET, resulted in 14 pregnancies (60.9%), of which 12 resulted in either live birth or are still ongoing (LB/OGPR= 52.2%). And for group 3, thirty patients (CR=14.3%) proceeded to ET resulted in 63.3% CPR/ET and 56.7% LB/OGPR. There was no significant difference in the CPR/ET, LB/OGPR, and the ET cancellation rate either due to no embryo available or ovarian hyper-stimulation syndrome. The overall CPR/ET and LB/OGPR for these non-donor IVF patients is 65.0% and 57% respectively.

Conclusion: The number of oocytes retrieved is not associated with CPR/ET and LB/OGPR in non-donor IVF cycles. ET cancellation rate does not increase in patients with <11 and >20 oocytes retrieved, and they have similar chance of success as patients who had optimum number of oocytes (11-20) retrieved.

P078

THE EFFICACY OF GROWTH HORMONE CO-TREATMENT IN OVARIAN STIMULATION OF THE PATIENTS WITH DECREASED OVARIAN RESERVE TREATED BY IN-VITRO FERTILIZATION

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Purpose: To evaluate the efficacy of growth hormone co-treatment to controlled ovarian hyperstimulation for in-vitro fertilization in women with decreased ovarian reserve.

Methods and Materials: This was a retrospective study. 271 medical records were reviewed from patients with low AMH level (<2.0 ng/ml) who underwent in-vitro fertilization in Gangseo Mizmedi Hospital between January 2012 and June 2013. Study group of 113 patients were given growth hormone co-treatment. 4mg of growth hormone was injected every other day from day 3 of IVF cycle until the day of hCG. Control group of 158 patients received the same treatment except the growth hormone co-treatment. Mean count of retrieved oocytes, endometrial thickness, and clinical pregnancy rate were compared.

Results: Patients' characteristics did not differ significantly between the two groups. Mean count of retrieved oocytes was 4.69 ± 3.02 in growth hormone co-treatment group and 4.83 ± 4.05 in control group without statistical significance. Endometrial thickness did not reached statistical significance also (8.23 ± 1.52 mm vs. 8.57 ± 1.76 mm, respectively). Clinical pregnancy rate in growth hormone co-treatment group (42.56%) was significantly higher than the control group (27.35%) ($p < 0.001$).

Conclusions: Women with decreased ovarian reserve undergoing controlled ovarian hyperstimulation and co-treatment with growth hormone for IVF revealed higher clinical pregnancy rate as compared with women of the same status without growth hormone.

P079

DIRECT INTRAPERITONEAL INSEMINATION: AN INTERMEDIARY BETWEEN INTRA UTERINE INSEMINATION AND IN-VITRO FERTILIZATION

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Introduction:

Among assisted reproductive techniques (ART) worldwide, In-vitro Fertilization (IVF) offers higher success rates in comparison to the simpler Intra Uterine Insemination (IUI).

Recent evidence on the success rates of Direct Intraperitoneal Insemination (DIPI), an alternate ART, is lacking.

Our study aims to determine whether DIPI can be recommended prior to IVF in selected patients.

Method:

The pregnancy rates and complications of patients undergoing DIPI at Anu Test Tube Baby Centre, an Infertility unit in Hyderabad, India, between January-October 2013 were analysed. All patients had prior tubal evaluation.

Results:

103 patients between 20 to 39 years age had DIPI.

Indications included IUI failures, unexplained infertility, cervical stenosis and male factor.

Stimulation was with clomiphene (33%), Clomiphene & Human Menopausal Gonadotrophin (CC+HMG) (63.1%) and flare protocol (0.09%).

Of the 25 (24.2%) patients who conceived, 32% had live births, 4% ectopic, 28% miscarriages and 36% have ongoing pregnancies.

Interestingly, the conception rate was 76% in 20-30 year age group, 80% in CC+HMG protocol and 90% in the first two DIPI cycles.

Complications were low and included fever (3.8 %) and post procedural abdominal pain(8.7%).

Conclusion:

DIPI is one of the least invasive and less expensive strategies of ART.

In our study, the DIPI conception rates were higher in younger patients and those on the CC+HMG protocol. The rates were highest in the first two DIPI cycles and complications were low. These encouraging results suggest that two cycles of DIPI can be recommended in young women before more expensive and invasive ART procedures.

P080

WHAT IS THE BEST HCG ADMINISTRATION TIME BASED ON THE SIZES OF THE DOMINANT FOLLICULAR GROUP IN LONG PROTOCOL IVF/ICSI?

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Objective: To analyze the sizes of the dominant follicular group for the hCG day.

Design: Retrospective study. Patients were chosen for (1) patients were ≤ 35 y; (2) BMI $< 28 \text{ kg/m}^2$; (3) bFSH $< 12 \text{ IU/L}$; (4) NO.oocytes retrieval ≥ 4 ; (5) no endometriosis, adenomyoma or uterine fibroid.

Material and Methods: 1350 cases were in this study. All the participants took standard GnRH-agonist protocol. The groups were categorized based on the number of follicles $\geq 16 \text{ mm}$ on hCG day: GroupA (≤ 4), GroupB (5-6), GroupC (≥ 7).

Results:

1350 cycles were analyzed: GroupA (N=370), GroupB (N=391), GroupC (N=589). The cancel rates were 5.67% (21/370) in GroupA, 5.62% (22/391) in GroupB, and 20.71% (122/589) in GroupC ($P < 0.05$). Clinical characteristics of the patients who underwent embryo transfer were comparable in the three groups. As shown in Table 1. OHSS rate were highest in groupC ($P < 0.05$). Clinical pregnancy rates were 54.15%, 62.60% and 57.38% respectively. In groupB, ongoing pregnancy rate and implantation rate were the highest ($P < 0.05$). A sub-analysis was shown in Table 2.

Table.1

	GroupA	GroupB	GroupC	P
Age	29.87 \pm 2.87	29.46 \pm 2.99	29.41 \pm 2.98	0.068
BasalFSH(IU/L)	8.42 \pm 2.84	8.05 \pm 3.01	7.50 \pm 2.00	0.000 ^{a)}
AFC	13.63 \pm 5.63	15.32 \pm 6.25	17.26 \pm 6.76	0.000 ^{a)}
No.oocyte	9.85 \pm 4.27	12.29 \pm 4.60	14.69 \pm 4.77	0.000 ^{a)}
Embryos transfer	2.13 \pm 0.42	2.10 \pm 0.32	2.08 \pm 0.35	0.133
CES	25.18 \pm 4.77	26.38 \pm 4.68	26.74 \pm 4.42	0.000 ^{a)}
Implatation rate(%)	36.69%(273/744)	43.09%(334/775)	39.92%(378/971)	0.034 ^{a)}
OHSSrate(%)	0.86%(3/349)	2.98%(11/369)	4.92%(23/467)	0.028 ^{a)}
Clinical pregnancy rate(%)	54.15%(189/349)	62.60%(231/369)	57.38%(268/467)	0.067
Ongoing pregnancy rate(%)	47.27%(165/349)	57.99%(214/369)	52.03%(243/467)	0.016 ^{a)}

Table.2

Clinical pregnancy rate(%)	($\Phi \geq 20\text{mm}$)=0	($\Phi \geq 20\text{mm}$)=1	($\Phi \geq 20\text{mm}$) ≥ 2	P
GroupA	53.26%(98/184)	51.96%(61/127)	65.78%(25/38)	0.305
GroupB	62.13%(105/169)	67.20%(84/125)	56.00%(42/75)	0.281
GroupC	56.20%(77/137)	56.64%(81/143)	58.82%(110/187)	0.874

Conclusions:When the number of follicle $\geq 16\text{mm}$ is 5 or 6, clinical pregnancy rate is the best. When it is less than 4, it is good to delay hCG administration; When it is more than 7, it is needless to delay hCG administration.

P081

THE EFFECT ACUPUNCTURE THERAPY TOWARD UTERINE ARTERIES BLOOD FLOW IN IVF PATIENTS ON YASMIN CLINIC, FKUI-RSCM

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Background

One of factors influencing success rate of IVF is endometrial receptivity (31-64%) which is influenced by uterine blood flow and hormonal profile. Acupuncture influencing uterine arteries blood flow by increasing LCS endorphin thus inhibiting the sympathetic system both central and segmental. Uterine arteries blood flow improved endometrial line indirectly which is useful in implantation process.

Objectives

This study is aimed to observe the effect of acupuncture toward uterine arteries blood flow by USG Doppler Monitoring of Uterine Arteries Pulsation Index (PI) and endometrial thickness.

Methods

This study applied the Quasi experimental methods. Patients in case group had given 4 times acupuncture therapy in rFSH stimulation period but hadn't to control group. Both of the group used Long Protocol. Uterine arteries blood flow (PI) evaluated on the day of embryo transfer. The acupuncture points in front of body were used LR3, SP4, SP6, ST36, SP10, CV3, CV4, LI4, PC6, ST29, KI13, and GV20. At the back BL13, BL17, BL23, BL28, BL57. Each side applied for 20 minutes. At the same time electrostimulation added to points, ST29, KI13, BL23 and BL28 in 2 Hz.

Results

Acupuncture therapy has improved uterine blood flow and increased endometrial thickness on case group significantly ($p < 0.05$).

Conclusion

Acupuncture therapy could improved artery blood flow and increased endometrial thickness in IVF patients.

Key words

Acupuncture, IVF, PI, uterine arteries blood flow, endometrial thickness.

P082

EFFECT OF BLOOD PRESENT ON THE ET CATHETER ON THE SUCCESS RATES OF IVF/ICSI

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Objectives: To investigate the effect of the embryo transfer catheter bloody staining during embryo transfer on their clinical outcomes.

Methods: Retrospective analysis about patients who underwent long protocol in vitro fertilization (IVF) or ICSI cycle from January 2011 to December 2012 in Reproductive Center of Sun Yat-sen Memorial Hospital. These transfers were divided into two groups according to whether the blood present on the transfer catheter or not. Further analysis was done in the subgroups according to the degree of blood on the ET catheter.

Results: In 1754 cycles without bloody staining on the catheter during ET, 847 cycles were associated with bloody staining. The implantation rate (IR) and clinical pregnancy rate (CPR) in the group without bloody staining were higher than the group with bloody staining (34.8% vs. 30.7%, P value = 0.002, and 54.1% vs. 47.6%, P value = 0.013). While no significant difference was found in IR and CPR among different groups of bloody staining degree.

Conclusion: Bloody staining on the embryo transfer catheter may decrease the embryo implantation and clinical pregnancy rate. However the degree of bloody staining may not worsen the clinical outcome.

P083

MEDIA SUPPLEMENTATION WITH GM-CSF IMPROVES ONGOING PREGNANCY RATES PER CYCLE STARTED

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Granulocyte-macrophage colony-stimulating factor (GM-CSF; Embryogen) has been shown to improve embryo development and has been reported to improve ongoing pregnancy rates in women with a previous miscarriage. We evaluated the effect of GM-CSF containing media on the clinical pregnancy rate and ongoing pregnancy rate per cycle started, in women undertaking IVF in an Australian setting.

Women under 43 with a previous history of miscarriage and/or implantation failure were recruited at FertilitySA. In 43 cycles, 43 women consented to their embryos being cultured in GM-CSF media to day 3 before a single or duo embryo transfer. Clinical pregnancy rates (intrauterine pregnancy on scan) and ongoing pregnancy rates (fetal heart on 7-8 week scan) per cycle started were estimated and compared to 81 previous cycles with a blastocyst transfer in this same cohort.

In the embryogen cycles, 32% (14 of 43) cycles resulted in a clinical pregnancy and 30% (13 of 43) had a fetal heart at a 7 week scan. Previously 28% (23 of 81) of cycles resulted in a clinical pregnancy whereas 14% (12 of 81) had a fetal heart. There was no statistically significant difference in clinical pregnancy rates ($P < 0.25$) but a higher rate of ongoing pregnancies ($P < 0.05$, χ^2 test) in cycles using GM-CSF containing media.

Although GM-CSF exposure did not influence clinical pregnancy rates, it was associated with higher ongoing pregnancy rates in women with previous miscarriage or implantation failure. This is consistent with previous findings that GM-CSF acts to protect embryos from stress and improve embryo development.

P084

THE SUCCESS RATE OF IVF HAS SIGNIFICANTLY IMPROVED OVER THE LAST DECADE

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This study was carried out to demonstrate that success rates with IVF have been improving despite decreasing the number of embryos transferred.

This was a retrospective cohort study comparing live birth outcomes for women who started IVF between 2001 and 2005 with women who started between 2006 and 2010. The data were obtained from a single IVF centre, Monash IVF Geelong, Victoria. The 2001-2005 cohort comprised 233 women who started IVF between the specified dates and included a total of 1119 stimulated and frozen embryo transfer cycles. The 2006-2010 cohort consisted of 453 women who started IVF between the specified dates and included a total of 1810 stimulated and frozen embryo transfer cycles.

The main outcome measure was a live birth, defined as the delivery of a baby greater than 20 weeks gestation confirmed to be a live birth. Life table analysis was used to evaluate cumulative live birth rates.

Cumulative live birth rates demonstrated that the probability of a live birth by cycle six was 73.7% in the 2001-2005 cohort; which significantly increased to 88.4% by cycle six in the 2006-2010 cohort ($p < 0.05$). There was an average of 1.8 embryos transferred per embryo transfer in the 2001-2005 cohort; which decreased to an average of 1.3 embryos transferred per embryo transfer in the 2006-2010 cohort.

The IVF success rate has significantly improved despite the number of embryos transferred being reduced, and in particular success rates among older women have increased. This study provides further support for elective single embryo transfers.

P085

IS THERE ANY ASSOCIATION BETWEEN ESTRADIOL LEVELS ON THE DAY OF HCG AND PREGNANCY RATE IN FRESH AND FROZEN EMBRYO TRANSFER CYCLES?

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Background

The impact of estradiol levels on pregnancy rate in IVF is still in debate. Some studies inferred a potential deleterious effect of estradiol (E2) on the endometrial receptivity which could affect the IVF outcome.

Objective

To define the association between E2 levels on the day of hCG and pregnancy rate in fresh and frozen embryo transfer (FET) cycles.

Material and methods

In a case control study, data were collected from 4/2012 to 10/2013. 1709 patients with the first IVF cycle underwent stimulation with recombinant FSH in agonist or antagonist fixed protocols starting on day 6 of stimulation. Criteria of hCG was at least 3 follicles ≥ 17 mm in diameter. Embryos were transferred on day 2 or day 3. Among them, 556 patients had frozen embryos and were transferred in FET cycles. All FET cycles were hormonal therapy.

Results

Multivariate analysis using logistic regression showed there was no significant association between pregnancy rate and estradiol levels on the day of hCG in fresh and frozen embryo transferred cycles.

Conclusion

Estradiol levels on the day of hCG is not a predictor of the pregnancy rate in fresh and frozen embryo transferred cycles.

P086

WNT SIGNALLING PATHWAY MAY CHANGE IN PLACENTA UNDERGOING IVF-ET

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Wnt signaling components also play crucial roles in human placental development controlling trophoblast lineage determination, chorioallantoic fusion and placental branching morphogenesis. Assisted reproductive technology (ART), including induction of ovulation with high doses of gonadotrophin and in vitro culture, may be associated with changes in imprinting genes expression and methylation in animal models. However, the role of the Wnt signaling pathway in human placental development and differentiation with ART is only partly understood. We compared and analysis Wnt signaling pathway genes expression in first trimester placenta generated by standard IVF-ET and natural fertilizations. Twenty-one differentially expressed Wnt signaling pathway genes were identified in the IVF-ET treated first trimester placenta: 18 up-regulated (FZD8, FZD2, CAMK2D, CXXC4, JUN, WIF1, JUN, PRKACB, JUN, PRICK, LE1, CCND1, VANG2, WNT5A, FZD8, SFRP2, LEF1, SFRP2); 3 down-regulated (PORCN, WNT2, DKK1). These gene products were expressed in the placental villus tissues, either in the cytoplasm or in the membrane of syncytiotrophoblastic cells detected by immunohistochemistry. This is the first study in comparing differentially expressed genes in first trimester placenta from patients undergone IVF-ET treatment vs. those underwent natural fertilization. Abnormal profiles of critical Wnt signaling pathway genes, such as Wnt2, LEP1, FZDs and SFRPs, may be critical for activation and development of placenta. Our results also revealed Wnt signaling pathway in IVF-ET process is acting in a compensation manner in human trophoblast adhesion, invasion and differentiation.

P087

TRANSFERRIN AND IRON TRANSPORT IN IVF-ET AND NORMAL FIRST TRIMESTER PLACENTAS

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Iron (Fe) deficiency in pregnancy is associated to low birth weight and premature delivery while in adults it can results in increased blood pressure and cardiovascular disease. In our study, we compared transferrin (TF), transferrin receptor (TFRC) gene and protein expression and location in human IVF-ET and normal first trimester placentas. Placenta biopsies (n=20,) in the first trimester (7~8 weeks) from IVF-ET vanishing twin patients and medically abortion from natural fertilization were extracted. DNA microarray analysis was carried out to identify TF and TFRC mRNA expression (RT-qPCR certified microarray data) and protein expression and localization (immunohistochemistry) in IVF-ET compared to natural villous trophoblast. Transferrin (TF) gene expression levels significantly high in IVF-ET placentas compared to controls ($p<0.05$). While transferrin recept (TFRC) gene lower in IVF-ET ($p<0.05$). TF and TFRC in villous trophoblast were presented both in syncytiotrophoblast and in the cytotrophoblast, but they were predominantly present in the syncytiotrophoblast. In particular, stronger intensity of TF immunoreactivity was localized in the maternal facing syncytiotrophoblast compared to the fetal facing. In conclusion, these is the first observation about TF and TFRC in IVF-ET first trimester placenta vs controls. Thus, Fe transport could be increased in IVF-ET placenta, TF and TFRC through compensatory mechanism increase fetal growth and placental development. It is not possible to draw firm conclusion until the expression and activity of all the component of placental Fe transport system, including ferroportin, ferritin, IRPs, and DMT1, is analysis.

P088

CORRELATION BETWEEN IMPAIRED AUTOPHAGY AND TROPHOBLAST FUNCTION IN ART

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Autophagy is a process of self-degradation of cellular components in which double-membrane autophagosomes sequester organelles and fuse with lysosomes so that the contents can be digested by lysosomal enzymes. By using this system, cells can survive under starvation or stress conditions such as hypoxia or oxidative stress. The expression of autophagy-related proteins in the placenta were reported in deeply invaded EVT's in myometrium and perivascular regeion. In our study, placenta biopsies (n=20,) in the first trimester (7~8 weeks) from IVF-ET vanishing twin patients and medically abortion from natural fertilization were extracted. DNA microarray analysis was carried out to identify the specific gene expression profile and the biological pathway activated during the invasion and remodeling spiral arteries window in trophoblast. Several cytokine, such as TGFβ3, sENG, uPAR, TNFα were down-regulated in human trophoblast subjected IVF-ET compared with natural pregnancy. These down-regulated gene expression show autophagy may play a compensatory mechanism as a cellular bulk degradation system to maintain cellular homeostasis under stress. In ART, autophagy by obtaining energy may play importantly compensatory role in EVT invasion and remodeling spiral arteries under oxidative stress, ER stress and inflammatory responses. However, excessive autophagy can promote cell death, which is called autophagic cell death. These results could contribute to the understanding of mechanisms involve in the early placentation and poor parental outcome of ART singletons. Even after adjustment for maternal age and parity, significant differences of autophagy in placenta between ART and spontaneously conceived singletons persist.

P089

ABERRANT EPIGENETIC REPROGRAMMING OF PLACENTA IN ASSISTED REPRODUCTIVE TECHNOLOGY

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Assisted reproductive technology (ART) involves manipulating gametes through *in vitro* methods to achieve pregnancy when pregnancy is not possible by natural fertilization. ART is performed during critical periods of epigenetic reprogramming when epigenetic modifications, including DNA methylation and modification of histones, are being erased and re-established. We completed the first transcriptome profiling of human placental (n=8, gestational weeks 7-8) gene expression dynamic from IVF-ET to natural fertilization. We report seven paternally expressed genes and four maternally expressed genes which are significant transcriptional changed in 74 putatively imprinted genes (ANOVA, FDR $p<0.01$). TaqMan RT-qPCR analysis (n=20, gestational weeks 7-8) confirmed a significant (*t*-test, FDR $p<0.01$) differentiation of placental gene expression for IGF2, NDN, NNAT, PEG3, SFRP2, SNRPN, APC, SLC22A3, TFPI2, TP73 and RB1. Our study demonstrates imprinted genes in IVF-ET early placental can be simplistically separated into two categories: those genes that are associated to

negative parental outcome and those genes that, through a compensatory mechanism, sense the placenta and fetus is at risk and act to increase growth (positive effects).

Key words: placenta; assisted reproductive technology; epigenetic; compensatory mechanism; parental outcome;

P090

RISK FACTORS FOR ECTOPIC PREGNANCY DURING AN IN-VITRO FERTILISATION STIMULATION CYCLE.

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Objective: There are inconsistencies in the literature regarding the risk factors for having an ectopic pregnancy following IVF treatment. The purpose of our study was to evaluate the effect of type of stimulation during an IVF stimulation cycle, dose of rFSH, and day of embryo transfer in a large IVF program to determine the risk factors for developing an ectopic pregnancy.

Methods:

Retrospective database analysis using logistic regression.

Results:

We analysed 27619 cycles of IVF between the years of 2010 and 2013. The risk of ectopic pregnancy after IVF is significantly increased with increasing total dose of rFSH, OR 1.24 (CI 1.12 – 1.37) $p < 0.001$, increased numbers of eggs collected, OR 1.06 (CI 1.03 – 1.09) $p < 0.001$ and having 2 embryos transferred, OR 1. (CI 1.3 – 2.8). There was no significant difference in EP rates with age, different stimulation protocols or day of embryo transfer.

Conclusion:

Higher total dose of rFSH, greater numbers of eggs collected and increased numbers of embryos transferred were significantly associated with an increased risk of ectopic pregnancy. The mechanism of how each factor contributes to the development of ectopic pregnancy needs further investigation.

P091

FROZEN EMBRYO CYCLE HAD COMPARABLE OUTCOME WITH FRESH CYCLE IN IVF

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Background: Frozen embryo cycle has the opportunity to prime the endometrial lining properly for implantation. This study is purposed to know the outcome of frozen embryo cycle and compare it with the fresh cycle.

Method: We retrospectively analyzed 1084 fresh IVF cycles and 154 frozen embryo cycles from 1st January 2005 until 30th September 2013 in Yasmin Clinic, Cipto Mangunkusumo National Referral Hospital, Jakarta, Indonesia. We compared the outcome of clinical pregnancy and contributing factors between frozen embryo cycle and fresh cycle.

Results: There were 38 among 154 frozen embryo cycles (24.7%) and 283 among 1084 fresh cycles (26.1%) which resulted in clinical pregnancy. In 154 frozen cycles, 365 thawed embryos were replaced among 832 frozen embryos (43.8%). Age less than 33 years old, basal follicle count more than seven, eight-cells-embryo more than one were associated with clinical pregnancy in fresh cycle with OR 2.5 (95% CI 1.4-2.7), OR 2.3 (95% CI 1.1-2.7), and OR 5.2 (95% CI 3.1-8.8). In frozen embryo cycle, only eight-cells-embryo more than one was associated with clinical pregnancy with OR 3.6 (95% CI 2.2-5.9). Frozen embryo cycles which resulted in clinical pregnancy mostly were caused by male factor (39%), polycystic ovaries (13%), tubal factors (13%), unexplained factors (10.4%), endometrial polyps (5.2%), endometriosis (5.2%), and others.

Conclusion: The outcome of frozen embryo cycle was comparable with fresh cycle. Embryo quality was the strongest predictor in pregnancy

Keywords: fresh cycle, frozen embryo, eight-cells-embryo, clinical pregnancy

P092

DOES FSH SURGE THE TIME OF HCG TRIGGER IMPROVES ART OUTCOMES? A RANDOMIZED, DOUBLE-BLIND, PLACEBO-CONTROLLED STUDY.

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OBJECTIVE: To determine whether an artificially induced follicle-stimulating hormone (FSH) surge at the time of human chorionic gonadotropin (HCG) trigger can improve the ART outcomes.

DESIGN: Randomized, double-blind, placebo-controlled, clinical trial.

MATERIALS AND METHODS: During a long agonist suppression protocol, subjects were randomized to FSH bolus (6 amps) versus placebo at the time of HCG trigger (36 hours prior to oocyte retrieval). The primary outcome was clinical pregnancy rate and the secondary were the number of oocytes collected, fertilization rate, and implantation rate. Clinical pregnancy was defined by fetal heart motion at time of ultrasound. Data will be analyzed using Student t-tests and Chi-square as appropriate on an intention to treat basis.

RESULTS: To date, 663 subjects have completed cycles after randomization. There was no differences in baseline demographic characteristics between the two study groups. There were also no differences in cycle characteristics such as mean number of stimulation days, total gonadotropin dose, peak estradiol. There was no statistically significant difference in clinical pregnancy rate (53.2% vs. 56.1, P value=0.452), mean number of oocytes collected, fertilization rate, and implantation rate.

CONCLUSION: These preliminary data show that there was no effect of a bolus injection of FSH on the clinical pregnancy rate, mean number of oocytes collected, fertilization rate, and implantation rate. Data collection is ongoing and final data analysis on good quality embryo, cumulative pregnancy rate and live birth rate will be presented.

P093

OVARIAN STIMULATION DURING LUTEAL PHASE IS AN OPTIMAL STRATEGY IN POOR RESPONDERS WHO FAILED TO PRODUCE VIABLE EMBRYOS IN THE PRECEDING CYCLE

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Objective: to investigate whether ovarian stimulation in the luteal phase can be a novel and feasible strategy for poor responders who failed to produce viable embryos in the preceding cycle.

Setting: Academic IVF centre.

Patient(s): 28 women who underwent luteal phase stimulation cycles, with history of poor response or failure of oocyte retrieval in ovarian stimulation protocols initiated from follicular phase.

Intervention(s): Comparing IVF outcomes of 114 cycles of 28 poor responders, including 34 luteal phase and 80 follicular phase stimulation cycles.

Main Outcome Measure(s): Large follicles on hCG day, total Gn dosage, number of oocyte retrieved, fertilization, and top quality embryo rates. Clinical pregnancy rate per transfer was compared among three embryo origins: from luteal phase stimulation, follicular phase stimulation, and from mixed.

Result(s): Enrolled patients were aged 40.4±4.0 years with bFSH 11.7±5.8 IU/L. The differences in oocyte retrieval (2.9±1.9 vs 2.2±2.1), available embryos per cycle (1.9±1.4 vs 1.2±1.3) and top quality embryos per cycle (1.4±1.2 vs 0.8±1.0) between luteal phase and follicular phase stimulation cycles were significant ($P<0.05$). The clinical pregnancy rates per transfer with embryos from follicular phase, luteal phase or mixed were 5.88 %, 25% and 37.5% respectively ($P=0.011$).

Conclusion(s): Top quality embryos and clinical pregnancy could be obtained via luteal phase stimulation in poor ovarian responders. Luteal phase oocyte retrieval is a feasible strategy for women with impaired ovarian reserve, especially for those who have failed in preceding ovarian stimulation in follicular phase.

P094

COMPARISON OF 5% AND AMBIENT OXYGEN DURING DAYS 1-3 OF IN VITRO CULTURE OF HUMAN EMBRYOS

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Objective: To compare the effect of two oxygen concentrations used during days 1-3 of human embryo culture on embryo quality and pregnancy outcome.

Design: Retrospective analysis of the use of two culture conditions.

Setting: Fertility Centre, Hanh Phuc International Hospital

Patient(s): One hundred and eight patients undergoing IVF.

Intervention(s): Embryos were cultured in 5% CO₂ balanced (20% O₂) gas phase until day 1 then assigned to 20% or reduced (5%) oxygen concentration groups and cultured until ET.

Main Outcome Measure(s): Fertilization rate, embryo quality, pregnancy rates, and implantation rates.

Result(s): There were no differences in demographic features (age, type of infertility) between the two groups.

Fertilization rate, cleavage rate, D3 embryo rate, good D3 embryo rate did not differ between groups. Corresponding, 67% vs 69%, 99% vs 96%, 90% vs 88%, 52% vs 52%. But there are differences between the 5% and 20% oxygen concentrations in clinical pregnancy rate (53% vs. 35%) and implantation rate (31.7% vs. 20.5%).

Conclusion: The pregnancy and implantation rates in group of embryos cultured in low oxygen (5% oxygen) during cleavage stage (from day 1) are higher than those of group embryos cultured totally in 20% oxygen concentration.

P095

QUALITY OF TRANSFERRED EMBRYOS AS PREDICTOR OF IVF OUTCOMES

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Introduction: Morphology-based embryo scoring and selection are commonly used in IVF.

Objective: To investigate the association between morphology-based embryo quality and outcomes in an IVF program transferring early cleavage embryos.

Methods: This was a retrospective study of 1694 IVF cycles undergoing fresh embryo transfer on day 2. The 2010 Istanbul consensus for embryo scoring was applied. The studied cycles were divided into 2 groups: group 1 (n=1070) had at least 1 good embryo transferred; group 2 (n=624) with no good embryo transferred. Main outcomes were clinical pregnancy rate (CPR), ongoing pregnancy rate (OR).

Results: Mean age of women in group 1 and group 2 were 33.3±5.02 and 33±5.1, respectively (p= 0.828). Average number of embryos transferred (ranged 1-4) were not significantly different between group 1 and group 2 (2.89 ± 1.11 vs. 2.98 ± 1.02; p=0.3). CPR and OR of group 1 were significantly higher than group 2 (42.1 vs. 20.7%, RR 2.04, 95% CI 1.91 – 2.17, p=0.001; and 37.5% vs. 15.5%, RR 2.41, 95% CI 2.27 – 2.55, p=0.001, respectively).

Conclusions: There was a strong relationship between quality of transferred embryo and IVF outcomes. Availability of good embryo for transfer is a good predictor for pregnancy outcomes.

P096

OUTCOMES FOLLOWING THE INTER-CLINIC TRANSPORT OF CRYOPRESERVED EMBRYOS USING LN2 DRY SHIPPER

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Aim: To determine whether the inter-clinic transport of cryopreserved embryos produces the same embryo survival rates, incidence of foetal heart and birth rates as non-transported cryopreserved embryos.

Method: Multi-centre retrospective data analysis was conducted on transported cryopreserved embryos intended for frozen embryo transfer in patients under 38 years of age between 2003-2012. 160 vitrified blastocysts (n=95 cycles) and 714 slow frozen cleavage stage embryos (n=325 cycles) were transported to Queensland Fertility Group (QFG) clinics in Brisbane, Toowoomba, Cairns and Townsville using dry Liquid Nitrogen shippers. Embryos cryopreserved, thawed and transferred at QFG Brisbane were used as a control. Main outcome measurements analysed were embryo survival rate, foetal heart incidence/100 embryos transferred (FH) and births after 20 weeks/100 embryos transferred (B20+). Chi-squared statistical analysis and odds ratios with a 95% confidence interval were calculated with $p < 0.05$ significant.

Results: Vitrified blastocysts transported and thawed from 2008 to 2012 showed no impact on embryo survival rate (80.8% vs 82.5%, $p > 0.05$), FH incidence (22.8 vs 20.7, $p > 0.05$) and B20+ incidence (19.8 vs 19.0, $p > 0.05$). Slow frozen cleavage stage embryos transported and thawed during 2003 to 2012 showed no impact on embryo survival rate (78.8% vs 75.9%, $p > 0.05$), FH incidence (19.4 vs 17.3, $p > 0.05$) and B20+ incidence (17.1 vs 15.0, $p > 0.05$).

Conclusion: Greater population dynamics and access to improving technology has resulted in an increased need to transport embryos nationally and internationally. The shipment of slow or vitrified cryopreserved embryos is safe when using a dry LN2 shipper according to instructions.

P097

EVALUATION OF AN OOCYTE VITRIFICATION METHOD REPLACING DIMETHYL SULPHOXIDE WITH PROPANEDIOL IN A CLOSED SYSTEM

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Introduction: Oocyte dehydration for vitrification has previously been achieved using dimethyl sulphoxide (DMSO) at a high concentration. Although this approach produces clinical results similar to those from fresh oocytes, there are concerns regarding potential DMSO-related toxicity. The aim of this study was to compare survival following vitrification of in-vitro matured (IVM) oocytes dehydrated in either a DMSO or propanediol (PROH) based cryoprotectant solution.

Methods: Immature oocytes, unsuitable for clinical use at the time of ICSI, were matured overnight (IVM). IVM oocytes were initially dehydrated in 7.5% ethylene glycol (EG) + 7.5% DMSO at 22°C, or 7.5% EG + 8% PROH (Vitrolife) at 37°C until re-expanded, followed by further dehydration in double concentrations of each cryoprotectant plus 0.5 M sucrose. Oocytes were loaded on to a Rapid-i™, placed in a pre-cooled straw, sealed and vitrified. Oocytes were warmed rapidly and rehydrated in decreasing concentrations of sucrose.

Results: Equilibration in PROH solution at 37°C facilitated faster re-expansion (3 minutes) compared to DMSO solution at 22°C (10 minutes). Survival was comparable in both solutions; DMSO 84.3% (70/83) and PROH 88.7% (55/62).

Conclusion: Dehydration of oocytes in PROH solutions at 37°C had no detrimental impact on survival. Similar survival can be achieved with both DMSO and PROH cryoprotectant solutions for IVM oocytes in a closed vitrification system.

P098

SURVIVAL RATE OF EMBRYO FOLLOWING VITRIFICATION AND WARMING IN DIFFERENT CARRIERS

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BACKGROUND: There are currently 2 methods of vitrification. The open carrier system allows embryos to be in direct contact with liquid nitrogen (LN₂), whereas in the closed carrier, a sealed system is used to sequester embryos from the liquid nitrogen.

OBJECTIVE: To compare the survival rate of embryo between open and closed carrier. We calculated the percentage of live cells in a cleavage stage embryo following vitrification and warming using two commercially available carrier systems.

METHODOLOGY AND MATERIALS: A retrospective study looking at day 2 & 3 vitrified human embryos from July 2010 to July 2013. The open system using McGill Cryoleaf™ (n=169) was tested against CBS™ High Security Vitrification (HSV) close system (n= 107). The percentage of survived cells of human embryos were calculated. A 95% confidence level ($p < 0.05$) was used to interpret the statistical significance.

RESULTS: A total of 290 human embryos were vitrified. The percentage of survived cells of human embryos were 84.02% (142/169) and 94.39% (101/107) for open and close systems respectively and the difference was not statistically significant ($p=0.164$).

DISCUSSION/CONCLUSION: There was no significant difference between the percentage of survived embryos in open and closed system groups ($p=0.164$). The survival rate of human embryos can be maintained following warming when the procedures of vitrification and warming are performed according to strict laboratory protocols and by well-trained embryologist. A closed system is more preferable since it helps reduce the chance of cross-contamination.

P099

CLINICAL OUTCOMES FROM OOCYTES VITRIFIED IN A CLOSED SYSTEM.

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Introduction: High survival and outcomes similar to fresh oocytes have been observed with an open oocyte vitrification system using donor oocytes. It has been suggested that only extremely fast cooling rates in open systems are suitable for human oocytes. However, our previous research using IVM oocytes indicated that the Rapid-i™ closed vitrification tool can achieve an appropriate cooling rate for successful oocyte vitrification. This retrospective study looks at clinical outcomes using oocytes vitrified in this closed system from June 2012 to present.

Methods: Human oocytes were vitrified in 15% ethylene glycol (EG) and 15% dimethyl sulphoxide (DMSO) together with 0.5M sucrose, as previously described, before being loaded onto the Rapid-i™ (Vitrolife) which was then sealed in a pre-cooled sleeve. Oocytes were stored under liquid nitrogen before being subsequently warmed and rehydrated via serial dilutions of sucrose.

Results: In 10 patients a total of 81 oocytes were warmed with an 84% survival rate (68/81). Fertilisation using ICSI achieved a fertilisation rate of 53% (36/68) with the majority undergoing cleavage: 92.5% (33/36). Fourteen embryos were transferred resulting in four pregnancies with an implantation rate of 29% (4/14).

Conclusion: These preliminary results indicate that the Rapid-i™ tool can achieve survival rates similar to those reported for open systems and clinical outcomes comparable to a similar fresh control population. Although the numbers are low, the apparent lower fertilisation rate may reflect extended time prior to vitrification and/or extremely poor sperm quality.

P100

IN-VITRO MATURATION (IVM) OF OOCYTES RETRIEVED FROM SMALL FOLLICLES FOR STEADY RESPONSE IN STIMULATED IN-VITRO FERTILIZATION (IVF) CYCLES

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Objective: To evaluate the effectiveness of IVM for oocytes derived from steady-response IVF patients

Methods: Cohort study. Steady response is defined as all follicles are less than 10mm after at least 10 days of standard stimulation. Patients with received 10,000 IU hCG and oocytes were retrieved from small follicles at 36-38 hours later. Oocytes were cultured for further maturation after recovered. Maturation assessment were done at 0, 20, 25 hours after oocyte retrievals. Sperm insemination, embryo culture and day 2 embryo transfers were carried out as usual. Treatment outcomes of fresh transfers were described.

Results: From January 2012 to June 2013, there were 55 patients recruited. Mean female age was 30.5 ± 4.2 years. Mean AFC and AMH were 15.8 ± 6.0 and 8.7 ± 4.1 , respectively. Means oocytes retrieved and embryos transferred were 17.4 ± 10.7 , and 3.3 ± 1.1 , respectively. Transfer was cancelled in two cases due to unfavorable endometrium. Clinical pregnancy rate after fresh transfers was 32.1% (17/53) and implantation rate was 12.6%. Twenty-nine patients (54.7%) had extra embryos for cryopreservation after fresh transfers.

Conclusion: IVM is feasible and effective for steady-response patients during ovarian hyperstimulation in IVF treatment.

P101

A PROSPECTIVE RANDOMISED CONTROLLED STUDY OF SERUM ANTI-MULLERIAN HORMONE (AMH) AS A PREDICTIVE MARKER OF OVARIAN HYPERSTIMULATION SYNDROME (OHSS) IN IVF-ICSI CYCLES

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Objective: The objective of this prospective randomised controlled study was to determine whether serum Anti Mullerian Hormone (AMH) level can predict ovarian hyperstimulation syndrome (OHSS) prior to the selection of controlled ovarian stimulation (COS) protocols.

Design: A total of 413 IVF-ICSI cycles at our centre from 2007 to June 2013 were prospectively studied. The patients in the study underwent a long GnRH-Agonist or an Antagonist protocol after randomisation using a computer generated list.

Materials and Methods: Baseline hormone profile and AMH were determined on day-2. The ovarian response was monitored using transvaginal ultrasound and serum estradiol (E2) levels.

Results: In our study, 06% of all patients developed moderate to severe OHSS. 03.9% patients undergoing the Antagonist protocol and 8.1% patients undergoing the long GnRH Agonist protocol developed OHSS. Only 31% patients with OHSS had PCOS. We found that PCOS patients had high serum AMH levels. The mean serum AMH level for patients with OHSS in our study was 5.6ng/ml. Thus, the basal serum AMH level appeared to be a more efficient predictor of OHSS than the patients Age, BMI and AFC.

Conclusions: High serum AMH levels can be utilised to predict OHSS in IVF-ICSI cycles in a better way than than Age, BMI and AFC and mild stimulation protocols may be applied in such patients. This study indicates that a single AMH assay may be used to individualise treatment strategies. Based on this evidence, AMH may be a useful marker to predict OHSS, in addition to being an appropriate marker for ovarian reserve.

P102

INSERTING HUMAN BLASTOCYST IN THE LOOP OF GAMA SLEEVED CRYO-DEVICE BASED ON MOUSE EMBRYO VITRIFICATION STUDY

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Abstract

Background: the need to cryopreserve embryos has increased. Study was conducted to provide Gama sleeved cryoloop as a suitable procedures for vitrification of mouse blastocyst, then applied clinically at Permata Hati clinic.

Objectives: to examine 0.1 µl Gama sleeved cryoloop for promotion vitrification.

Methods: mouse blastocyst were vitrified with ethylene glycol (EG) and dimethylsulphoxide (DMSO) based cryoprotectants. Equilibrium solution: (7.5% EG (v/v); 7.5% DMSO (v/v)) for 2 minutes, followed by vitrification solution (15% EG (v/v); 15% DMSO v/v; 10 mg/ml Ficoll; 0.65 M Sucrosa) for 30 seconds at room temperature before inserted in 0.1, 1, 3µl Gama sleeved, Cryologic® and Hampton Research® loop, then plunge into liquid nitrogen. Embryos was warmed in 0.25M (2 min) and 0.125M sucrosa (3 min). Handling media used was MOPS buffered. Embryo survival was assessed by in-vitro development and mitochondria activity.

Results: 0.1µl Gama sleeved cryoloop was not significantly different in retrieval, survival and hatching rate compare to other devices ($p>0.05$). However 3µl Gama sleeved and Hampton Research® loop gave less retrieval rate. Mitochondria activity of post warmed blastocyst was not significantly differ among the devices used ($p>0.05$). Upon informed consent to the patients undergoing IVF who had supernumerary embryos, blastocyst were vitrified using 0.1µl Gama sleeved cryoloop. Survival was assessed by reexpanding to its previous morphology. From 39 patients, eleven became pregnant (28.2%). Two women delivered healthy babies, 5 ongoing pregnancies, 4 ended with early pregnancy lost.

Conclusion: human blastocyst could be vitrified used 0.1 µl Gama sleeved cryoloop.

Keywords: cryoloop, blastocyst, vitrification

P103

COMPARISON OF SLOW FREEZING AND VITRIFICATION OF CLEAVAGE STAGE EMBRYOS

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Background: In recent years, there has been a worldwide move towards cryopreserving embryos by vitrification rather than the traditional slow freezing methods. However each year, many births occur following the thawing and transfer of slow frozen cleavage stage embryos.

Aim: To compare the outcomes from thawed slow frozen cleavage stage embryos with those vitrified at the cleavage stage.

Method: A retrospective data analysis was conducted on all frozen embryo transfers undertaken between June 2012 and July 2013 within two laboratories thawing or warming both slow frozen and vitrified cryopreserved cleavage stage embryos (104 FETs for vitrified embryos and 970 FETs for slow frozen embryos). 2064 slow frozen and 140 vitrified cleavage stage embryos were thawed/warmed in the reviewed FET cycles. Embryo survival, cleavage post-thawing/warming, cycles reaching transfer and cycle outcomes were reviewed.

Results: Warmed vitrified cleavage stage embryos showed a significantly higher intact survival rate (84% vs 74%; $P = 0.0065$) and post-warming cleavage rate (88% vs 68%; $P = 0.0001$) than slow frozen embryos. Moreover, there was a trend towards more of the vitrified cycles reaching transfer (95% vs 89%; $P = 0.09$). The percentage of cycles resulting in a clinical pregnancy was higher in the slow frozen group than in the vitrified group (29% vs 25%), although this was not statistically significant ($P = 0.5$).

Conclusion: Warmed vitrified cleavage stage embryos show higher survival and cleavage rates than slow frozen embryos with similar clinical pregnancy rates. Vitrification could be a faster, more convenient approach to cryopreservation than slow-freezing.

P104

DNA DAMAGE AND VACUOLES: CAN THESE BE IMPROVED BY A DENSITY GRADIENT (DG)?

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Aim:

Sperm head vacuoles and their size are associated with sperm DNA damage ⁽¹⁾, and decreased pregnancy rates ⁽²⁾. Our aim was to ascertain if DNA Fragmentation Index (DFI) and vacuoles are reduced using DG sperm preparation.

Method:

136 sperm samples had DFI and vacuoles assessed on both the raw ejaculate and post DG preparation (*SpermGrad*). Vacuoles were measured at x7026 magnification using IMSI Strict (*Hamilton Thorne*). Vacuole size was grouped as – 0-4%, 5-10%, >10% .

DFI was assessed using SCSA. DFI ranges ⁽³⁾ were Low= $\leq 15\%$, Mid= $15-29\%$, High= $\geq 30\%$.

Results:

Mean DFI reduced significantly after DG

DFI range	n	Mean Ejaculate DFI %	Mean Post DG DFI %	T-Test
Low	96	7.9	4.1	p=<0.001
Mid	34	21.3	11.1	p=<0.001
High	6	37.3	22.0	p=0.01

The % of sperm with >10% vacuoles was only reduced following DG in the higher DNA damage group.

		Ejaculate (raw)			Post DG			
		Vacuoles size (%)			Vacuole size (%)			
DFI range	n	0-4%	5-10%	>10%	0-4%	5-10%	>10%	X ²
Low	96	24523/38400 (63.9%)	11423/38400 (29.7%)	2454/38400 (6.4%)(c)	26400/38400 (68.7%)	9639/38400 (25.1%)	2361/38400 (6.1%)(c)	(c) NS
Mid + High	40	9301/16000 (58.1%)	4990/16000 (31.2%)	1709/16000 (10.7%)(d)	10251/16000 (64.1%)	4577/16000 (28.6%)	1172/16000 (7.3%)(d)	(d) p=<0.01

Conclusion:

(a) DG significantly reduces the mean values of DFI damaged sperm.

(b) DG effectively reduces, but does not eliminate, vacuole affected sperm in patients with an elevated DFI.

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P105

CAN THE DEGREE OF BLASTOCYST EXPANSION AT WARMING AND TRANSFER PREDICT PREGNANCY OUTCOME?

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Aim:

To determine whether the % expansion of day 5 and day 6 vitrified blastocysts at warming and transfer, will predict clinical pregnancy.

Method:

All surplus embryos at PIVET Medical Centre are cultured routinely to Day 6. Blastocysts with a grade 3BB or better (Gardner's Scale) on Day 5 or Day 6 are cryopreserved using the Cryotop method of vitrification.

Subsequent FET cycles are always SET under a HRT regiment and involve warming of the blastocysts about 1 hour prior to transfer.

The % expansion of the blastocoele cavity at warming and just before embryo transfer was observed and recorded.

From 2012 to August 2013, Day 5 and Day 6 blastocysts had blastocoele expansion measured and recorded at warming (n=208) and at the time of transfer (n=209)

Results:

Blastocysts that were >50% expanded at warming were more likely to result in clinical pregnancy (49% vs 32%).

Blastocysts that had expanded to >50% by the time of transfer were twice as likely to result in clinical pregnancy (49% vs 18%).

Conclusion:

Measuring the % expansion of the blastocoele cavity at the time of warming, but more importantly at the time of embryo transfer, may predict clinical pregnancy outcome.

P106

EMBRYO MONITORING USING TIME-LAPSE VIDEO MONITORING: REPORT OF 3PN FERTILIZED OOCYTE DEVELOPING INTO 8-CELL EMBRYO

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Background: Embryo morphology and pronuclei assessment are included in the criteria for embryo selection before embryo transfer. However, when using conventional methods, pronuclei assessment may sometimes be missed. The use of time-lapse video monitoring may help retrace the development of embryo, including the pronuclei and cleavage process.

Objective: To observe the development of a tripronuclear (3PN) fertilized oocyte using a time-lapse video monitoring.

Method: We performed intracytoplasmic sperm injection (ICSI) in five oocytes. A fertilization check was conducted 12 hours after ICSI to determine the number of pronucleus. Fertilized oocytes were then monitored using a time-lapse microscope in a monitoring incubator. Image-capture was performed every 20 minutes.

Result: We found one tripronuclear (3PN) fertilized oocyte which we observed under a time-lapse video monitor. It developed into a two-cell, three-cell, four-cell, and eight-cell stage 24 hours, 37 hours, 38 hours, and 64 hours after ICSI, respectively.

Conclusion: Embryo morphology alone is not sufficient as a criteria for embryo selection before embryo transfer, as a 3PN fertilized oocyte may develop into a 8-cell embryo with normal cleavage times. Time-lapse video monitoring may serve as an important tool to assist monitoring of embryo development.

Keywords: 3PN, embryo monitoring, time-lapse video monitoring

P107

EXTENDED CULTURE EMBRYO DEVELOPMENT IS PREDICTIVE OF IMPLANTATION.

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Recently time lapse imaging technology has used embryo development over time (morphokinetics) to assess embryo competence. These studies suggest that the initiation of compaction through to blastocyst development is essential for implantation. Embryos that develop slowly have a high risk of aneuploidy and are thought not to result in an ongoing pregnancy.

The aim of this study was to determine if the on time development was critical for embryo implantation and additionally to evaluate if this was impacted by maternal age.

This retrospective study of 777 IVF/ICSI single embryo transfer cycles used data collected between 2010-2012. Transfers occurred 4 or 5 days after oocyte collection. Cycles were grouped according to age, embryo development and day of transfer.

The day 4 implantation rate for slow embryos (cleavage/morula) did not differ significantly when compared to on time embryos (compacted morula/early blastocyst) for women ≤ 38 . On time embryos from women ≥ 39 did demonstrate a higher implantation rate when compared to slow embryos.

Implantation rates for slow and on time embryos were significantly different when embryos were transferred on day5 for women in both ≤ 38 and ≥ 39 age group categories.

On time development was not essential for implantation in all age groups. However, developmental delay had a much higher impact when embryos were transferred on day 5 and in women age ≥ 39 . Clinics should not be deterred to transfer developmentally slow embryos.

P108

ADVANCED SPERM SELECTION FOR ICSI: SYSTEMATIC REVIEW AND META-ANALYSIS

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Background: Advanced sperm selection techniques have developed as a means of improving ART outcomes. It is thought that selection of a structurally intact and mature sperm with high DNA integrity may improve fertilisation and pregnancy outcomes. Techniques described include selection of sperm bound to hyaluronic acid, sperm with normal ultramorphology, high nuclear birefringence, electronegatively charged, and non-apoptotic sperm.

Aim: To evaluate the impact of advanced sperm selection techniques on ART outcomes

Methods: Systematic review and meta-analysis of prospective randomised controlled studies concerning advanced sperm selection techniques for ICSI

Results: This study presents a systematic review and meta-analysis of prospective randomised controlled studies of advanced sperm selection techniques for ICSI.

P109

THE RELATIONSHIP BETWEEN THE SIZE OF LEADING FOLLICLES AND IVF TREATMENT OUTCOMES IN GNRH AGONIST (GNRHA) STIMULATED CYCLE

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In human IVF programmes, follicular size is commonly used as indicators of oocyte maturity. Follicle measuring $\leq 14\text{mm}$ are generally accepted to contain competent oocytes. However, there is no consensus on the maximum size of follicles. The effect of "over-sized" follicles on overall oocytes competency and endometrium receptivity is unclear.

In this study, we attempt to evaluate the effect of leading follicular size on treatment outcomes in 102 GnRHa cycles (IVF \pm ICSI) performed in TMC Fertility Centre. Follicle sizes were determined using transvaginal ultrasound scan and final oocyte maturation was triggered by the administration of 10,000IU recombinant-hCG. Patients were separated into 3 groups according to the size of three leading follicles during the time of hCG administration: Group 1 ($\geq 20\text{mm}$), Group 2 ($\geq 22\text{mm}$) and Group 3 ($\geq 24\text{mm}$). The fertilization rate, embryo utilization rate and clinical pregnancy rate (CPR) were analyzed. The mean age and number of oocytes retrieved were similar.

Outcome: The fertilization rate was 65.9%, 69.4% and 66.3% respectively for Group 1, 2 and 3. The embryo utilization rate is also similar among the 3 groups (Group1=66.8%, Group2=62.3% and Group3=66.2%). There CPR appeared to decrease when the size of the leading follicles increased; from 60.8% in Group 1, to 54.5% in Group 2 and 40% in Group3, but the difference is not significant.

Conclusion: The size of leading follicles does not affect fertilization rate and embryo utilization rate in GnRH agonist cycles. However, there is a trend of lower CPR when the size of the leading follicles exceeds 24mm.

P110

IS PREMIXING OF SERUM SAMPLES NECESSARY TO OBTAIN REPRODUCIBLE AMH RESULTS USING THE ANSHLAB ASSAY

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Introduction The widely used Beckman Gen II assay for AMH has recently been modified by introducing a premixing step with buffer, possibly to reduce interference from complement causing inconsistent results, particularly after sample storage. In previous studies we have found that results obtained from pre-mixed samples are significantly higher than those using the original method. A second commercial company, Anshlab, has produced new AMH assay with a different primary antibody. We investigated the stability and reproducibility of AMH measurement using this method and also investigated whether pre-mixing has a similar effect as seen with the Beckman Gen II assay. Method Fresh and stored samples from a group of pre-menopausal women were assayed using both assays with and without premixing with assay buffer prior to plating. Results Measured AMH levels were on average 2-fold higher using the Anshlab method compared with the Beckman assay, and similar to those using the premix Beckman assay. The Anshlab assay produced consistent AMH measurement when samples were stored up to 48 hours at room temperature or at -20°C but inconsistent results when samples were stored for longer periods. Premixing samples with assay buffer before assay using Anshlab kit also produced on average

40% higher but consistent AMH values regardless of the storage conditions. Conclusion Measured AMH levels seemed to be more consistent using the Anshlab kit at least for samples stored up to 48 hours. However, a similar premixing effect was also observed with Anshlab kit which suggests the likely presence of serum interference in the assay.

P111

WHICH GRADE OF HOST IS THE BEST FOR ICSI?

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WHICH GRADE OF HOST IS THE BEST FOR ICSI

Objective: Evaluate the results of ICSI in combination with different swelling grades of HOST and select HOST grades for ICSI efficiency optimally.

Design: Cross-sectional Description.

Subjects: The couples was made ICSI with 100% non-motile sperm from 01/06/2011 – 31/05/2013. Total 17 cases:

- 2 cycle of fresh sperm ejaculation
- 5 cycles of fresh sperm after microsurgery
- 10 cycles stored frozen sperm after microsurgery

Results:

Table 2: Comparison fertility rate between three grades.

Overall comparison					
X = 46.09%; γ = 0.263; p = 0.109					
Compare grade 1 and 2		Compare grade 1 and 3		Compare grade 2 and 3	
37.5 ± 4.7	62.80 ± 8.2	37.48 ± 4.7	49.7 ± 11.15	62.80 ± 8.2	49.7 ± 11.15
T-test, p = 0.013		T-test, p = 0.298		T-test, p = 0.345	

Table 3: Comparison good embryo rate between three grades.

Overall comparison					
X = 54.99%; γ = 0.58; p = 0.006					
Compare grade 1 and 2		Compare grade 1 and 3		Compare grade 2 and 3	
36.67±8.17	61.94±9.42	36.67±8.17	71.43±14.87	61.94±9.42	71.43±14.87
T-test, p = 0.055		T-test, p = 0.039		T-test, p = 0.578	

Conclusion: The selection of sperm swelling grades affects the ICSI outcome. Namely our initial study, swelling grade (2) and (3) improve significantly fertility and good embryo rate after ICSI. Since this is likely the optimal swelling grades when selecting sperm for ICSI after HOS. However, there are studies with larger sample sizes to have an accurate conclusion.

P112

THE INTRODUCTION OF VITRIFICATION AS THE CRYOPRESERVATION METHOD FOR CLEAVAGE STAGE EMBRYOS IN A SMALL REGIONAL ART FACILITY - A RETROSPECTIVE ANALYSIS OF THE RESULTS.

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In October 2010, Vitriification was introduced into our laboratory as the new cryopreservation method for cleavage stage embryos. Previously these were slow frozen. Since then, 343 vitrified and 136 slow frozen cleavage stage embryos have been warmed in 191 and 83 FET cycles respectively. We present a retrospective analysis of the survival rates of the warmed embryos during this period and demonstrate the initial learning curve of a new technique introduced into a small regional ART facility.

A logistic regression analysis was applied to the data to estimate the probability of an embryo surviving a warming procedure. Whilst the probability of an embryo surviving a warming procedure was less for vitrified embryos as compared to slow frozen embryos when the technique was first introduced in 2010, by 2012, the probability of an embryo surviving the warming procedure was the same. The survival probability of vitrified embryos has continued to increase over time.

Comparing these to the industry standard KPI's reported by the ALPHA consensus meeting, we were pleased that one year after introducing the procedure we were demonstrating 'competent' survival rates and now after 3 years of performing vitrification we are approaching 'benchmark' survival rates. Our slow frozen survival rates have been at benchmark rates for the entire period analysed.

Whilst an initial learning curve exists when introducing any new technique into a laboratory, clinics should not be discouraged, as over time we have demonstrated survival rates to be comparable. These may even show to be superior in the future.

P113

PREGNANCY RATE AFTER INTRAUTERINE INSEMINATION USING VARIOUS NUMBER OF SPERM CENTRIFUGATION FOLLOWED BY SWIM UP METHOD

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Sperm preparation is an important step in intrauterine insemination (IUI) program to select good quality sperm. The goal of this research is to observe the pregnancy rate after IUI using swim up method sperm preparation with various number of centrifugation. Fifty-one patients of Melinda Fertility Center, Bandung, Indonesia, who had not undergone IVF cycle and normospermia characteristics were participated as samples of this research. Samples were distributed to 3 groups, (i) without centrifugation (n=17); (ii) single centrifugation (n=17); and (iii) double centrifugation (n=17). Centrifugation done before placing medium above sperm layer (swim up method). After incubation time, sperm with good quality were harvested for IUI. Then, the concentration, volume, percentage of progressive motility, and number of harvested sperm were observed. The results of these research showed that: 1) There is no significant different (Kruskal-Wallis Test, $p < 0.05$) on motility and the number of harvested sperm among 3 groups; 2) The pregnancy rate after IUI using double centrifugation followed by swim up method (24%) higher than single centrifugation (18%) and without centrifugation (18%). It could be concluded that centrifugation did not influence the quality of sperm after preparation but can improving the pregnancy rate.

P114

ASSESSMENT OF HUMAN OOCYTE QUALITY BY MEIOTIC SPINDLE SIZE

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OBJECTIVE: Recent studies suggest that the meiotic spindle may be an indicator of oocyte quality. This study aimed to investigate the relationship between the meiotic spindle size in human oocytes and embryonic developmental potential after intracytoplasmic sperm injection (ICSI).

MATERIALS AND METHODS: We analyzed 1013 oocytes with a visible meiotic spindle from 222 patients. Meiotic spindle size was determined using a PolScope before ICSI. Oocytes were classified into four groups according to the size of the spindle area: group A, < 80 μm^2 ; group B, 80–100 μm^2 ; group C, 100–120 μm^2 ; group D, >120 μm^2 .

RESULTS: Among the 1013 oocytes with a visible spindle, 100 (10%) were classed as group A, 298 (29%) as group B, 351 (35%) as group C, and 264 (26%) as group D. Fertilization rate in group A (71%) was significantly lower ($p < 0.05$) than in groups B, C and D (82%, 82%, and 77%, respectively). The oocytes in groups B and C progressed to a significantly higher number of blastocysts ($p < 0.05$) than in groups A and D (44% and 51% vs. 25% and 29%, respectively). The percentage of clinical pregnancies in groups B and C was higher than in groups A and D (28% and 28% vs. 9% and 12%, respectively).

CONCLUSIONS: Oocytes with a spindle area of 80–120 μm^2 showed high blastocyst and pregnancy rates. These results suggest that quantitative measurement of the meiotic spindle area provides an indication of oocyte quality.

P115

LOW-INTENSITY PULSED ULTRASOUND-MEDIATED STIMULATION OF MOUSE SPERMATOGONIAL STEM CELL PROLIFERATION AND COLONIZATION

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Background: Spermatogonial stem cells (SSCs) are the foundation of spermatogenesis. New procedure such as sound wave especially low intensity ultrasound (LIUS) can be effective on increasing the number of cells. In this study we investigated the effect of LIUS stimulation on mouse SSCs.

Materials and Methods: In the first phase, temperature controlled by LIUS stimulation of plate containing culture medium and in the next phase, SSCs stimulated by LIPUS with 200 mW/cm² with 20% and 40% Duty Cycle for 5 day and SSCs proliferation and colonization assessed at 10th day.

Results: Average of Proliferation rate in 20%, 40% Duty Cycle and control group were 1.86, 2.36 and 1.66, respectively. Average number of colonies in 20%, 40% Duty Cycle and control group were 74.67, 112.67 and 61.00, respectively. Average diameters of colonies in 20%, 40% Duty Cycle and control group were 196.6×178.0, 194.3×184.6 and 196.0×184.6, respectively. Our results showed that there was significant increase in Proliferation Rate and number of colonies in experimental groups compared to control group ($P \leq 0.05$).

Conclusion: These results suggested that LIPUS treatment is be an efficient and cost-effective method to improve proliferation and colonization of SSCs during in vitro culture.

Key words: colonization, mouse, Stem Cell, Ultrasound

P116

THE EFFECT OF LOW INTENSITY ULTRASOUND STIMULATION ON MOUSE SPERMATOGONIAL STEM CELL PROLIFERATION AND COLONIZATION

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Background: Spermatogonial stem cells (SSCs) are the foundation of spermatogenesis. New procedure such as sound wave especially low intensity ultrasound (LIUS) can be effective on increasing the number of cells.

Materials and Methods: SSCs stimulated by LIUS with 3 different Intensity dose (100, 200 and 300 mW/cm²) for 5 day and SSCs proliferation and colonization assessed at 7th day.

Results: Average of Proliferation rate in 100, 200, 300 mW/cm² and control group were 1.93, 2.26, 1.73 and 1.66, respectively. Average number of colonies in 100, 200, 300 mW/cm² and control group were 25.67, 33.33, 20.67 and 20.67, respectively. Average diameters of colonies in 100, 200, 300 mW/cm² and control group were 230 × 180, 226 × 183, 215 × 169 and 228 × 175, respectively. Our results showed that there was significant increase in Proliferation Rate and number of colonies in experimental groups compared to control group ($P \leq 0.05$), whereas there were not significant differences between groups regarding to diameter of colonies.

Conclusion: These results suggested that LIUS treatment is be an efficient and cost-effective method to improve proliferation and colonization of SSCs during in vitro culture.

Key words: colonization, mouse, Stem Cell, Ultrasound

P117

EVALUATION OF IMMUNOGENICITY AND CONTRACEPTIVE EFFICACY OF GAMETE SPECIFIC RECOMBINANT PROTEINS FOR THE DEVELOPMENT OF CONTRACEPTIVE VACCINE

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There is an urgent need for the management of stray dogs' populations as they act as carrier of deadly rabies virus. A contraceptive vaccine which would inhibit fertilization by targeting sperm or egg or both the gametes simultaneously provides an exciting option. A fusion protein encompassing promiscuous T cell epitope of tetanus toxoid (TT) and dog zona pellucida glycoprotein-3 (TT-KK-ZP3) without any affinity tag has been expressed and purified from *E. coli*. Female FvB/J mice immunized with TT-KK-ZP3 showed dose dependent increase in antibody titer which correlated well with reduction in fertility. Antibodies showed binding with mouse and dog ZP matrix in indirect immunofluorescence. Additionally, a chimeric polypeptide comprising TT, C- terminus fragment of dog ZP3 (ZP3_{C-term}) and a fragment of dog spermatozoa specific protein, Izumo was expressed and purified from *E. coli* (TT-KK-ZP3-GGG-Iz). Female mice immunized with TT-KK-ZP3-GGG-Iz showed specific antibody response against ZP3_{C-term} as well as Izumo in ELISA. Antibodies reacted with mouse and dog ZP matrix and acrosome reacted mouse and dog spermatozoa. A physical mixture of individually expressed ZP3_{C-term} and Izumo also elicited similar kind of immune response but antibody titer was low as compared to chimeric protein. Groups of mice were also immunized with Izumo and ZP3_{C-term} individually. All the groups showed varying degree of curtailment in fertility as compared to control. It is likely that these endeavours may help us to propose a candidate vaccine for the management of street dogs' population that may eventually lower the burden of rabies infection.

P118

SPERM SPECIFIC PROTEIN FROM MARINE MOLLUSC-A POTENTIAL CANDIDATE MOLECULE FOR FERTILITY CONTROL

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Background: Proteins are the ideal candidate molecule in the evaluation of newer and safer spermicides. Marine mollusc (*Telescopium telescopium*) is a rich source of sperm protein isolated from the spermatheca gland. **Methods and Findings:** Proteins were isolated from the spermatheca gland homogenate by different protein isolation procedures. Gelfiltration and SDS-PAGE revealed that the SF-50 was a mixture of proteins with molecular weights ranging from 23-115 kD. SF-50 showed sperm immobilizing and contraceptive efficacy in laboratory mammals. The MEC of SF-50 that caused instant immobilization of rat spermatozoa in-vitro was 25µg/ml. Hypo-osmotic swelling test of rat sperm with SF-50 revealed disintegration of sperm plasma membrane and dissolution of acrosomal cap, which is the cause leading to sperm agglutination and finally death. We thus infer that SF-50 possesses significant sperm agglutination potential, which may be explored further for its possible future promise as an effector constituent vaginal contraceptive. The mechanism of spermicidal action of SF-50 was studied by measuring LPO, protein carbonyl content as an index of oxidative damage and assessed DNA fragmentation by DNA diffusion assay. The plausible findings are that the sperm death occurs due to excess generation of reactive oxygen species, which in-turn causes lipid peroxidation of sperm plasma membrane. The membrane damage is probably as a result of DNA damage caused by apoptosis brought in by the protein SF-50. **Conclusions.** These results support further clinical development of this first spermicidal protein for evaluation of in vivo spermicidal efficacy in human for development of a safer vaginal contraceptive.

P119

EFFECTS OF THYMOQUINONE SUPPLEMENTATION ON CYCLOPHOSPHAMIDE TOXICITY OF MOUSE EMBRYO IN VITRO

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Thymoquinone is the major active component derived from the traditional medicinal plant *Nigella sativa*, which has been shown to exhibit antioxidant property through different mechanisms in animal models. This study evaluates the prophylactic effect of thymoquinone supplementation on culture medium to ameliorate cyclophosphamide-induced alterations in cellular differentiation and proliferation during embryo development *in vitro*. Male and female mice were exposed to

cyclophosphamide via a single intraperitoneal (i.p.) injection at 200 mg/kg. Sperms and oocytes were collected at day 33 and day 10 respectively, for insemination and fertilization in medium supplemented with thymoquinone (1 μ M, 10 μ M and 100 μ M). The stages of fertilization, embryo division, morphological effects and fragmentation were examined and compared between groups, 24 hours post-fertilization. Thymoquinone supplementation improved fertilization rates, significantly reduced the percentage of defects blastomeres of Type C ($p < 0.001$) and significantly decreased the percentage of embryo fragmentation Grade IV ($> 50\%$, $p < 0.05$) following paternal and maternal exposure to cyclophosphamide. The good quality embryos of Type A and Grade I fragmentation were not observed in the group without thymoquinone supplementation. The findings of this study showed that thymoquinone is a suitable exogenous antioxidant for preserving fair-quality embryos which can result into full term pregnancy.

Keywords: Cyclophosphamide, Thymoquinone, *In-Vitro* Fertilization, Blastomeres, Embryo, Fragmentation

P120

GPR30 MEDIATES THE FAST EFFECT OF ESTROGEN ON MOUSE BLASTOCYST AND ITS ROLE IN IMPLANTATION

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It was reported that there was a fast action of estrogen on the target cells. Recent researches have suggested that G protein-coupled receptor 30 (GPR30) is involved in the rapid effects of estrogen. However, it remains unknown whether GPR30 mediates the fast action of estrogen in mouse blastocysts and its role in implantation. Applying immunocytochemistry, we proved that GPR30 expressed on the mouse blastocyst cells and their locations were mostly consistent with the binding sites of estrogen on the plasma membrane. Confocal microscopy was used to examine the dynamic change of intracellular calcium which was labelled by Fluo-3/AM, we found that E₂, E₂-BSA as well as GPR30-selective agonist, G-1 could induce their rapid increase of [Ca²⁺]_i, while GPR30-selective antagonist, G15 could inhibit the effect of E₂-BSA. Furthermore, Applying immunocytochemistry, we proved that E₂, E₂-BSA and G1 could induce the clustering of integrin α v, β 1 and β 3, while G15 may block the clustering induced by E₂-BSA. Utilizing FN binding assay, we revealed that E₂, E₂-BSA and G1 could increase the FN binding activity of integrins in blastocyst cells, while G15 may block this effect of E₂-BSA on it. Using embryo co-culture with endometrial epithelial cells and embryo transfer, we further found that blastocysts treated by G15 had lower adhesion rate and implantation rate compared with control. Our work provide that GPR30 expressing in mouse blastocysts mediates the rapid action of estrogen and may play an important role in blastocysts adhesion and implantation via increase the affinity of integrins with their ligands.

P121

TRANSPORTATION OF VITRIFIED EMBRYOS USING DRY SHIPPERS - DOES IT AFFECT ART OUTCOMES?

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Introduction:

Transportation of vitrified embryos has become a necessary aspect of ART with patients changing location and enabling operation of satellite IVF programs. Data must be examined to determine whether transportation is detrimental to embryo viability.

Aim:

To determine the effects of transporting vitrified embryos using a dry shipper upon thaw survival and pregnancy rates.

Methods:

Data from Monash IVF satellite clinics (shipped group) and main site (non-shipped group) between January-August 2013 were evaluated retrospectively using Fishers exact test with $p < 0.05$ considered significant.

Results:

A total of 2032 blastocysts were thawed for transfer. There was no significant difference in thaw survival rates between non shipped and shipped embryos (90.2% vs.88.6%) nor pregnancy rate per transfer with 37.8% for non shipped embryos compared to 35.1% shipped group. Similarly, A grade embryos showed no significant difference in thaw rate (91% non shipped, 95.9% shipped) or pregnancy rate per transfer (42.9% non-shipped, 48.4% shipped). B grade embryos did not show differences in thaw rate or pregnancy rate. C grade embryos showed no difference in pregnancy rate, however, the thaw rate was significantly lower in shipped embryos (89.8% vs 80.7%, $p = 0.024$). This difference may be attributed to sample size of this group ($n = 83$, 4% of total embryos).

Conclusion:

The shipment of vitrified embryos using a dry shipper does not appear to have a detrimental effect on thaw rate and implantation potential of embryos. Ongoing data collection to increase sample size is needed to determine whether there is an effect on poorer quality embryos.

P122

ICSI CYCLE OUTCOME IN RELATION TO THE TIMING OF INJECTION

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Objective: To evaluate the effect of ICSI timing on the fertilization rate (FR), clinical pregnancy rate (CPR), live birth rate (LBR) and implantation rate (IR).

Methods:

This is a retrospective analysis of 528 ICSI cycles performed in our centre from 2009 to 2012. A total of 5169 oocytes were injected. Oocyte pick up was done at 37 hours (h) post- human chorionic gonadotrophin (hCG) injection. ICSI timing was recorded when the first oocyte was injected. In our laboratory, injection time was influenced by our workload. Data analysis was done after dividing the ICSI timing into 4 groups as follows; group A: 38h, group B: 39-40h, group C: 41-42h and group D: ≥43h post-hCG.

Results:

There were 3, 415, 107 and 3 cases in each group respectively. FR was similar among the 4 groups (62.96%, 69.29%, 71.78%, 62.50% respectively). The CPR, LBR and IR for group A, B and C was (33.33%, 45.13%, 39.18%; 33.33%, 35.64%, 36.08%; 28.57%, 25.05%, 25.44% respectively). No pregnancy was resulted in group D.

Conclusion:

In our centre, acceptable ICSI outcome was achieved when ICSI is done between 39-42h. No pregnancy was achieved when ICSI was done after 43h indicating that although preincubation before ICSI is necessary, delayed injection produced a poorer outcome might due to postmaturity in oocyte. Therefore, planning should be done properly to ensure that ICSI can be performed on time especially on busy days.

P123

IS VITRIFY ALL A BETTER OPTION?

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Study question

Is there a significant difference in clinical outcome between fresh embryo transfer and frozen embryo transfer (FET)?

Summary answer

Clinical pregnancy rate in FET cycle is significantly higher compared to fresh cycle. Miscarriage rate is lower in FET cycle compared to fresh cycle.

What is already known

Implantation rate is improved in a non-controlled ovarian stimulation (COH) cycle due to no deleterious effects of COH in embryo-endometrium synchrony.

Study design, size, duration

This is a retrospective study where we included all patients below the age of 42 (n=190) with fresh embryo transfer and had surplus embryos vitrified in 2012. We compared the clinical outcome of the fresh embryo transfer (ET) and the frozen embryo transfer (FET) of this group.

Main results and the role of chance

Result: See Table 1

	Fresh	FET
# of ET	190	107
#of biochemical	97	61
# of clinical	70	54
#of miscarried	27	7
biochemical PR	51.05%	57.01%
clinical PR	36.84%	50.47%
Miscarriage rate	27.84%	11.48%

Using analysis clinical pregnancy rate is higher and miscarriage rate is lower in the FET group $p < 0.05$.

Wider Implication

From the analysis it is clear that FET gives higher clinical pregnancy and lower miscarriage rate therefore moving forward there is a role to vitrify all embryos in order to achieve higher cumulative clinical pregnancy rates.

P124

RELATIONSHIP OF EARLY CLEAVAGE WITH EMBRYO DEVELOPMENTAL POTENTIAL AND IVF±ICSI CLINICAL PREGNANCY RATE (CPR)

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Objective: To evaluate the developmental potential of early-cleavage (EC) and non-EC embryos and their pregnancy outcome in IVF±ICSI patients.

Methods: 44 IVF±ICSI patients in Tropicana Medical Centre who had at least 4 oocytes fertilized were recruited into this prospective study. Embryo assessment was first performed at 25-27hours post insemination/injection. Embryos displaying 2 cells at inspection were designated as EC embryos and were cultured separately from non-EC embryos. Embryo quality was again assessed on the day of transfer and scored from grade 1 to 4, with grade 1 being the best grade and grade 4 being the worst. EC embryos were preferentially transferred in all cases. Patients were categorized into 3 groups according to the embryos selected for transferred: Group A= EC embryos only (n=20); Group B= combination of EC and non EC embryos (n=17); Group C =non-EC embryos only (n=7) and the CPR was evaluated. Mean age (33.0) and mean number of embryos transferred (2.0 ± 1) does not differ between the 3 groups.

Results: The proportion of good embryos (grade 1 and 2) on day of transfer was significantly higher in EC group compared to non-EC group (74.6% vs 56.5%, $P=0.0001$). There is a trend of higher CPR following the transfer of only EC embryos (Group A=75%), compared to group B (58.8%) and group C(57.1%). However, there difference was not statistically significant.

Conclusion: Early cleavage is associated with better embryo quality and the selective transfer of EC embryos only shows a trend of higher CPR in IVF±ICSI treatment cycle.

P125

THE RELATIONSHIP BETWEEN BLASTOMERE REGULARITY, EARLY CLEAVAGE AND MULTINUCLEATION

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Introduction

Embryos showing uneven cell division have significantly lower early cleavage, pregnancy and implantation rates than evenly cleaved embryos as well as higher levels of aneuploidy and multinucleation (Hardarson et al, 2001). In this study, we have reexamined the relationship between uneven embryo cleavage, early cleavage, multinucleation and pregnancy rates.

Method

Normally fertilized oocytes from 720 IVF/ICSI cycles were examined for early cleavage at 23-25 hours post insemination. On day 2, embryos were scored for cell number, multinucleation and regularity of cleavage. The latter was divided into four categories: even, slightly uneven, uneven and very uneven. Only cycles in which embryos of the same cleavage group were transferred were analysed.

Results

3973 zygotes were cultured to day 2. 30.4% divided evenly, 28.1% were slightly uneven, 26.3% uneven and 15.3% had very uneven cleavage. Cleavage irregularity was significantly correlated with early cleavage to 2-cells ($P = 0.01$) and the incidence of multinuclei ($P = 0.04$), with very uneven embryos having fewest 2-cell embryos (10.4%) and the highest multinucleation rate (35.4%), compared to even embryos (22% and 10.1% respectively). When only even or slightly uneven cleaved embryos were transferred, pregnancy and implantation rates (28.8% and 23.0%) were higher than uneven and very unevenly cleaved embryos (24.4% and 17.5%).

Conclusion

This study confirms that embryos showing irregular cleavage on day 2 have slower early cleavage and increased multinucleation rates as well as lower pregnancy and implantation rates. Delayed early cleavage may produce embryos with higher aneuploidy and/or multinucleation levels and associated cleavage anomalies.

P126

SUCCESSFUL DEVELOPMENT OF A MOUSE ZYGOTE VITRIFICATION PROTOCOL UTILISING AN AUTOMATED INSTRUMENT.

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The aim of this study was to develop a mouse zygote vitrification protocol utilising an automated vitrification instrument (AVI). As part of this protocol development the impact of vitrification solution exposure times on the survival and development of mouse zygotes were assessed.

F1 (C57BL/6J X CBA) zygotes were vitrified with the Kitazato Cryotop® (CT) or AVI, with 60 (T60) or 90 (T90) second exposure to the final vitrification solution. All zygotes were cultured using sequential media.

Zygotes were placed on the AVI in a custom-designed pod. Cryopreservation solutions containing increasing concentrations of Ethylene Glycol, dimethylsulfoxide and trehalose were robotically aspirated and dispensed for equilibration and vitrification. Finally the pod was sealed and vitrified.

The recovery rate achieved by the instrument was 120/124 (97%). Initial survival rates of recovered embryos were significantly reduced for the AVI groups (CT: 67/68 (98.5%), T60: 51/58 (87.9% $p=0.02$) and T90: 54/62 (87.1% $p = 0.01$)). However, there was no significant difference in day 5 blastocyst outcomes calculated from all recovered zygotes (T60: 44/58 (75.9%), fresh: 45/63 (71.4%), CT: 54/68 (79.4%)). The T90 group had significantly lower (37/62 (59.7% $p=0.049$)) blastocyst development.

These results show it is possible to develop a mouse zygote protocol for the automated vitrification instrument that achieves equivalent blastocyst development rates to a commonly used manual vitrification system. The AVI automates and standardises vitrification protocols which increases the repeatability of outcomes and allows variables, such as solution exposure times, to be further defined and tailored to the requirements of the embryo.

P127

DYNAMIC ANALYSIS OF RELATIONSHIP BETWEEN TIMING OF SYNGAMY AND HUMAN EMBRYONIC DEVELOPMENT USING TIME-LAPSE CINEMATOGRAPHY

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Objective

In 2011, Istanbul consensus workshop has proposed that the assessment of syngamy is very sensitive predictors for embryonic quality. However, the value as independent predictor of outcome is still unclear. In this study, we analyzed the relationship between timing of syngamy and human embryonic development using time-lapse cinematography (TLC).

Materials

and

Methods

We have developed a system of TLC, which culture temperature was maintained at $37.0 \pm 0.2^\circ\text{C}$ and pH at 7.37 ± 0.05 . After c-IVF or ICSI, donated oocytes (n=203) were analyzed for TLC. In c-IVF, the cumulus cells were gently removed and transferred into culture media for TLC. In ICSI, oocytes were commenced TLC after the procedure.

Result

Of 203 oocytes, normally fertilized embryos were 128 (c-IVF: 50, ICSI: 78) and the rest were abnormal fertilization (n=13), undetectable (n=35) and unfertilized (n=27). In normal fertilized embryos, the time required from the extrusion of 2nd polar body to syngamy in c-IVF and ICSI were 21.2 ± 3.6 hours and 20.8 ± 3.6 hours, respectively. The time required for syngamy in good quality embryos (GQE) was 20.0 ± 3.3 hours and in poor quality embryos (PQE) was 22.1 ± 4.3 hours, respectively and the difference was significant ($P < 0.05$).

Conclusion

Although there was no difference in the time required for syngamy by the insemination procedures (IVF or ICSI), the time required for syngamy in GQE was significantly shorter than that in PQE, suggesting that timing of syngamy would be a useful parameter to assess the embryo quality in human embryos.

P128

ANALYSIS OF COMPACTION INITIATION IN HUMAN EMBRYOS USING TIME-LAPSE CINEMATOGRAPHY

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Objective

To analyze the initiation of compaction in human embryos *in vitro* during the compaction process, using a time-lapse cinematography (TLC) technique, with a goal of clarifying the exact details of the events in human embryogenesis, and both the precise timing of compaction and the underlying mechanisms controlling the process.

Materials and Methods

One hundred and fifteen embryos donated by couples with no further need for embryo-transfer were used in this study. Donated embryos were thawed and processed for TLC observation for 5 days. The morphological behavior was dynamically analyzed by the observation of the process of initiation of compaction in human embryos via TLC technique.

Results

Although initiation of compaction occurred throughout the period from the 4-cell to 16-cell stages, 86.1% of embryos initiated compaction at the 8-cell stage or later, with 8-cell stage initiation being most frequent (22.6%). Of these, 49.5% developed into good-quality blastocysts. In contrast, compaction initiation prior to the 8-cell stage was observed in 13.9%, of which only 18.8% developed into good-quality blastocysts. Embryos that initiated compaction before the 8-cell stage showed significantly higher numbers of multinucleated blastomeres, due to asynchronism in nuclear division at the 3rd mitotic division resulting from cytokinetic failure.

Conclusions

This study indicated that the initiation of compaction occurs at or after the third cell division in human embryos. Furthermore, embryos which initiated compaction before the 8-cell stage were associated with aberrant embryonic development (i.e., cytokinetic failure accompanied by karyokinesis).

P129

THE EFFECT OF CULTURE MEDIA PH ON MEIOTIC SPINDLE ASSEMBLY IN MOUSE OOCYTES.

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The aim of this study was to determine the effect of pH on the meiotic spindle morphology of mouse oocytes. The pH of bicarbonate-buffered fertilisation medium (G-IVF) was adjusted by exposure to standard incubator levels of CO₂ (6%) and the pH of MOPS buffered handling medium (G-MOPS) was pre-adjusted. Using polarized light microscopy to measure spindle retardance, images of metaphase II stage (MII) oocytes (n=76) were captured after exposure in G-IVF to 6% CO₂ for 15, 30, 45, 60, 75, 90 and 105 min. In the second experiment, MII oocytes (n=69) exposed to pre-equilibrated G-IVF were subsequently cultured in an ungassed incubator for 15, 30, 45, 60 and 75 min. Mean spindle retardance changed with time in both experiments ($P < 0.001$), reaching a peak corresponding to pH 7.4 to 7.5. A third experiment examined MII oocytes (n=68) held in standard G-MOPS (pH 7.2) and pre-adjusted G-MOPS (pH 7.4 and 7.5). Mean spindle retardance increased when oocytes were transferred from standard G-MOPS to the pre-adjusted G-MOPS, and decreased when they were returned to the standard G-MOPS ($P < 0.001$). These results show that meiotic spindle morphology is sensitive to changes in media pH, and that spindle retardance is greatest after exposure to media of pH 7.4 to 7.5. Therefore, culturing oocytes in a slightly alkaline medium during in vitro fertilisation (IVF) treatments may optimise meiotic spindle assembly. This finding has important implications for human IVF treatments, such as intracytoplasmic sperm injection, and warrants further investigation.

P130

NUMBER OF FOLLICLE FLUSHING AND RE-ASPIRATION OOCYTE AFFECT ON OOCYTE QUALITY AND PREGNANCY RATE

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OBJECTIVE

Ovarian follicle flushing during OPU has been suggested to improve oocyte recovery rate. In this study, we assessed prospectively the effect of flushing and re-aspiration of follicles on oocyte quality and pregnancy rates.

MATERIALS AND METHODS

During OPU, sequential follicular flushing were performed at low pressure using syringes filled with heparinized flushing media. Data were collected according to the average number of flushing and re-aspiration follicle divided by the total follicle retrieved. Then, its divided into five groups (one to up to five times flushes).

RESULTS

A total of 424 cycles were collected. Oocyte recovery rate was 67, 72, 72, 66 and 55% for group one, two, three, four, and five times flushes respectively.

Immature and abnormal oocyte recovery rates were increased with increasing number of flushing and re-aspiration.

No significant different in fertilization rate for one to up to four times flushes, but for five times flushes was highest.

Although no difference in the embryo quality was observed in each group, there were significant differences in pregnancy rate (40, 35, 36, 28, and 27% for group one, two, three, four and five times flushes respectively). The pregnancy rate was decreased by increasing number of follicle flushing and re-aspiration.

CONCLUSION

The number of flushing and re-aspiration doesn't improves the oocyte retrieval rate but on the contrary, increasing the immature and abnormal oocyte rates. Although no difference for fertilization and embryo quality in each group, our studies indicated that increasing re-aspiration follicle has negative effect on pregnancy rate.

P131

HIGH SURVIVAL AND ONGOING PREGNANCY RATES USING A NEW GENERATION OF SLOW FREEZING MEDIA

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The aim of this study was to evaluate the efficacy of slow freezing at pronuclear-stage (2PN) as a safe alternative in cases of elective freezing of all embryos (FAE). The outcome of 502 frozen embryo transfer cycles (FET) in 352 patients at Westmead Fertility Centre was retrospectively analysed (Jan 2012 – Sep 2013). Elective FAE was performed in cycles where; i) serum progesterone ≥ 5 nmol/L on the day of hCG (246 cycles), and ii) risk of ovarian hyperstimulation syndrome (136 cycles) iii) uterine pathologies and other (120 cycles). 2PN's were frozen and thawed using new generation cryo solutions. Thawed 2PN's were cultured for two or four days before being transferred.

A total of 1753 out of 1877 2PN's survived and underwent cleavage (93.4% per 2PN's; 94.8% per patient). 158 ongoing pregnancies were achieved in 1.4 FET cycles per patient with 1.2 embryos transferred (31.5% ongoing pregnancies per FET cycle; 44.5% per patient). 111 poor prognosis patients in 118 cycles with three or less embryos available had FET on day three achieving 18 ongoing pregnancies (15.3% per FET cycle; 16.2% per patient). 241 patients in 384 cycles had three and four embryos available and had FET at the blastocyst stage. 140 of these patients had an ongoing pregnancy (36.5% per FET cycle; 58.1% per patient). These results show that freezing of 2PN's is a safe and successful option for FAE cycles, allowing for high survival and pregnancy success, but most importantly assuring that all the patients embryos are indeed frozen.

P132

MELATONIN PROMOTES THE IN VITRO DEVELOPMENT OF MICROINJECTED PRONUCLEAR MOUSE EMBRYOS AND INCREASES THE EFFICIENCY OF BLASTOCYST IMPLANTATION THROUGH ANTI-OXIDATIVE AND ANTI-APOPTOTIC EFFECT

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Microinjection is the main methods to produce transgenic animals. However, the low developmental rate of embryo due to microinjection limits its application and this common problem remains to be solved. In the current study, we observed that melatonin at its physiological concentration (10^{-7} M) significantly promoted the *in vitro* development of murine microinjected pronuclear embryos. This was indicated by the increased blastocyst rate, hatching blastocyst rate and blastocyst cell number with melatonin treatment. In addition, when these blastocysts were implanted into female recipient mice, the pregnancy rates (91.7% vs. control 60.0%) and farrowing rates (30.5% vs. control 19.9%) were significantly improved compared to their non-melatonin treated counterparts. Mechanistic studies revealed that melatonin treatment reduces ROS production and cellular apoptosis during *in vitro* embryo development and improves the quality of blastocysts. The implantation of blastocysts with higher quality leads to high pregnancy rates and increased farrowing rates.

P133

CLINICAL OUTCOMES USING CRYOTEC WARMING METHOD FOR EMBRYOS VITRIFIED BY MEDICULT VITRIFICATION MEDIA

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Introduction:

We had previously used Medicult for vitrifying embryos at Alpha International Fertility Centre but since July this year, we had started using the Cryotec Method (Cryotech, Japan) in our frozen-thawed cycles. This study describes the outcomes of embryos vitrified by Medicult, but thawed using the Cryotec Method.

Methods:

20 patients who had their embryos frozen with Medicult were thawed with Cryotec from July to October 2013. The mean age of patients is 33.2 years. From these 20 patients, 16 have embryos frozen on Day 3 (Group A), 2 have blastocysts frozen on Day 5 (Group B) and 2 have blastocysts frozen on Day 6 (Group C). The mean number of embryos transferred was 2.2. Clinical pregnancy and number of gestational sacs were determined using ultrasound.

Results:

Post-thawed survival rates for Group A, B and C were 88.5%, 100.0% and 100.0% respectively. Clinical pregnancy rate per embryo transfer for Group A was 60.0%, Group B was 50.0% and Group C was 100.0%. The implantation rates for Group A, B and C were 47.1%, 50.0% and 100.0% respectively. The overall clinical pregnancy and implantation rates per embryo transfer were 63.2% and 51.2% respectively.

Conclusion:

This preliminary study shows that embryos at different developmental stages which were previously vitrified using Medicult vitrification media can be successfully thawed using Cryotec Warming Method with excellent survival rate, and results in high implantation and clinical pregnancy rates.

P134

ENDOMETRIOSIS DECREASED THE NUMBER OF GOOD QUALITY EMBRYOS

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Abstract

Oxidative stress and trace elements in the oocytes environment is explored in endometriosis and impact on in vitro fertilization (IVF) outcome assessed. Embryos are subdivided according to the morphology of compaction to predict embryo's potential. Grade 1 embryos represented optimal quality, while grade 2 (exclusion of fragments) was characterized by a loss of cytoplasm.

AIM:

Using cleavage stage embryo in in-vitro fertilization as an ideal model, we sought to compare quality of embryo in endometriosis-associated infertility versus male factors-associated infertility.

METHODS:

276 consecutive embryos cycles from 84 patients from April 2011 until June 2013 were retrospectively evaluated and analyzed to determine if either embryo resulted from endometriosis patients (n=114 embryos) and male factors patients (n=162 embryos) at cleavage stage (day 3). The primary outcomes measured grade 1 (good) or grade 2 (poor) of embryo on day 3 embryo status.

RESULTS:

Average age of endometriosis and male factors patients are 34±4 years old, 33±5 years old, respectively. Average embryos of endometriosis and male factors patients are 2,7±1,8, 3,8±2,6, respectively. Endometriosis resulted lower embryo quality (40% [46/114]) compared to male factors (41% [66/162], P>0.05), but not significant. In total, good quality embryo resulted from endometriosis and male factors 41% (68/164) and 59% (96/164), respectively.

CONCLUSION:

Endometriosis decreased the number of good quality embryos compared to male factors.

P135

LOW OXYGEN TENSION INCREASE ANTIAPOPTOTIC EFFECT BY EXPRESSION OF OXYGEN AND ANTIOXIDANT RELATED GENES IN MOUSE BLASTOCYST CULTURED IN VITRO.

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Objective: O₂ concentration is a crucial factors on embryo culture. It is range from 2% to 8% in the oviduct and uterus of mammalian species. However, the effects of low oxygen tension in embryogenesis are still unclear. The aim of our study is to evaluate embryonic growth and apoptosis in normoxia and hypoxia. Moreover, the expression of hypoxia inducible factors (HIF-1α and HIF-2α), the key regulators in hypoxia, its target genes (GLUT-3 and VEGF) and antioxidant genes (MnSOD and PRDX5) were evaluated in mouse embryo cultured under hypoxic and normoxic conditions.

Methods:The 2-cell embryos were cultured to blastocyst stage under 3% or 20% O₂ tension. Real-time PCR, Immunofluorescence and terminal deoxynucleotidyl transferase-mediated dUTP nick-end labeling (TUNEL) assay were performed.

Results:The blastocyst formation rate was not affected in either 3% O₂ or 20% O₂ group. However the hatching rate was significantly increased in 3% O₂ group compared to 20% O₂ group ($P < 0.05$). Expression of GLUT-3 and VEGF was increased 4.14 and 7.99-fold, respectively in 3% O₂ group ($P < 0.05$), although HIF-1 α and HIF-2 α mRNA level was similar in two groups. Immunofluorescence staining showed the intensity of MnSOD was higher and HIF-2 α was observed in the nucleus in 3% O₂ group. Apoptotic index was significant increase in 20% O₂, compared with 3% O₂ group ($P < 0.05$).

Conclusions:This study indicates that low O₂ tension culture not only provides a more conducive environment by antiapoptotic effect, but also promotes embryonic glycolysis and angiogenesis during early implantation and hatching, these effect may regulated by HIF-2 α , act as key mediators, in hypoxia.

P136

RBPJ MODULATES THE CHORIOALLANTOIC ATTACHMENT BY TARGETING VCAM-1

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Placenta forms the interface between the maternal and fetal circulation, facilitating metabolic and gas exchange as well as fetal waste disposal. The development of a functional mouse placenta is a dynamic phenomenon and regulated by many pathways, including Notch signaling pathway. However, the molecular processes by which members of the Notch pathway exert their roles remain not well understood, and the roles of the transcription factor *Rbpj* in the placentation is undefined. *Rbpj* was deleted systemically after *ZP3-Cre* and *Prm-Cre* mice mated with *Rbpj loxp/loxp* mice. At E8.5, the allantois failed to fuse with the chorion in *Rbpj* null embryos. The tetraploid complementation assay showed that the dysfunction of allantois in *Rbpj* deficiency embryos was the major factor for the failure of chorioallantoic attachment. It is known that chorioallantoic attachment is dependent on the interaction between the cell adhesion molecule *Vcam-1*, which is expressed on the allantois, and its ligand *Integrin- α 4*, which is expressed by the chorionic mesothelium. *In situ* and immunostaining analysis discovered that the level of *Vcam-1* declined significantly while the level of *Integrin- α 4* did not change. And the dual-luciferase reporter assay showed that *Rbpj* regulates the *Vcam-1* promoter. In summary, these findings reveal that *Rbpj* may modulate the placentation through regulating the chorioallantoic attachment by targeting *Vcam-1*.

P137

INTRAVAGINAL DANAZOL EFFECTIVENESS ON MENSTRUAL PERIODS AND BLEEDING VOLUME IN HEAVY MENSTRUAL BLEEDING PATIENTS IN MEDAN-INDONESIA, GRADED ON A PICTORIAL SCALE

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ABSTRACT

Objective : To determine the effect of intravaginal Danazol use on duration and bleeding volume in Heavy Menstrual Bleeding (menorrhagic) patients.

Study Design: This interventional clinical pre-post test trial study was performed at the outpatient clinics of Adam Malik General Hospital, Dr. Pirngadi General Hospital, and Halim Fertility Centre from January 2011 to June 2011 or until a minimum number of samples were obtained.

Material and Methods: For one menstrual period prior to treatment, all samples were not administered with Danazol, after which bleeding volume and duration were assessed, followed by administering **intravaginal Danazol** for 10 days during the next 2 consecutive menstrual cycles. Menstrual bleeding duration and volume were self measured, assessed and recorded using the Pictorial Blood Loss Assessment Chart (PBAC) by each patient. Data was statistically analysed after which results were presented in frequency distribution tables.

Results : Results showed that mean menstrual lengths prior to, 1 month and 2 months after danazol intravaginal use were 8.83 ± 1.11 , 6.63 ± 1.08 and 5.95 ± 1.22 days, respectively. Mean PBAC scores prior to, 1 month and 2 months after danazol

intravaginal use were 201.4 ± 57.7 , 103.1 ± 33.23 and 73.7 ± 34.34 points, respectively, indicating that this method is highly effective. Side effects (weight gain, digestive disturbances, headaches, facial hair, and soariness) were minimal.

Conclusion: Intravaginal Danazol is an effective alternative treatment in cases of menorrhagia to reduce menstrual bleeding duration and volume with minimal side effects.

Key words: Intravaginal Danazol, Heavy Menstrual Bleeding, Pictorial Blood loss Assessment Chart (PBAC).

P138

THE EFFECTS OF SLEEP DEPRIVATION ON REPRODUCTIVE FUNCTIONS OF FEMALE RATS

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Objective: It has been well documented that sleep deprivation (SD) is one of the health problems. However, the roles of SD on ovarian function were controversial. Using this animal model, we want to elucidate the effects of total SD and neurotransmitter-serotonin action on ovarian functional regulation.

Methods: Rats were subjected to a schedule of total SD for 1 to 4 days. Ovarian corpus luteums were counted. Serum levels of LH, E2, 5-HT were measured with EIA. Large follicles were extracted total protein. Serotonin receptor, steroidogenic acute regulatory (StAR) protein, and LH receptor were examined with Western blotting. Granulosa cells line were cultured and treated with 5-HT (0, 1, 10, 100, 1000 ng/ml) for 24 h to evaluate the StAR protein expression.

Results: SD for 4 days decreased the corpus luteum number ($P < 0.01$). SD led to significant decreases serum E2 levels and StAR protein expression, while 5-HT levels were significantly elevated ($P < 0.05$). The level of E2 and ovarian StAR protein were significantly lower on the rats for 4 days total SD. 5-HT decreased hCG-induced StAR expression for 30%, which appeared to be dependent on 5-HT₂ receptor activation. The reduction of the proteins could be reversed by 5-HT antagonist treatment.

Conclusion: Ovulation was disturbed in total SD rats. Decreased E2 concentration in total SD rats may be associated with decreased StAR protein expression and elevated 5-HT concentration. The 5-HT acts on the 5-HT receptor and reduces the StAR protein expression, subsequently, the StAR protein level were decreased and diminishes E2 secretion.

P139

NORETHISTERONE CONTROLLED VERSUS SPONTANEOUS MENSES IN IVF AGONIST CYCLES

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Objective:

This is a retrospective analysis comparing outcome of IVF patients on agonist protocol who had Norethisterone (NE) to adjust the timing of the next period (Group A) versus those who had natural menses (Group B).

Materials and Methods:

57 patients in Group A and 106 in Group B aged 36 from slow were analyzed from July 2011 to June 2013 in Alpha International Fertility Centre. Oocyte donation and PGD cases were excluded. The mean age of patients for Group A vs Group B were 31.9 vs 32.4 ($p > 0.05$). The indications for IVF and the mean number of embryos transferred were similar in both groups. In Group A, patients were given 10mg NE 3 times per day until they are ready to start their IVF cycle. Buserelin injections were started about 1 week before menses in Group A, and on day 21 or 22 in Group B. Puregon or Gonal-F was used. Final maturation of oocytes was triggered with Ovidrel 36.5 hours before the planned OPU time. Clinical pregnancy and number of gestational sacs were determined by ultrasound.

Results:

Clinical pregnancy rates per embryo transferred in Group A and Group B were 75.9% and 64.4% respectively ($p > 0.05$). Implantation rate for Group A was statistically higher (44.1% versus 32.9%) than Group B ($p = 0.03$).

Conclusion:

This study shows that the use of NE in IVF agonist cycles did not adversely affect clinical pregnancy and implantation rates. Unexpectedly, NE cycles had better implantation rates compared to those who had menstrual cycles.

P140

AMG LEVEL IS A PECULIAR INDICATOR OF PHYSIOLOGICAL FUNCTIONING OF OVARIES AT GIRLS OF TEENAGE AGE

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Level antimullerian hormone (AMH) in blood serum at "healthy" girls and patients with a syndrome of polycystic ovary syndrome (PCOS) at the age of 15 – 17 years

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Research objective — to study level AMH in blood serum at "healthy" teenage girls at the age of 15 – 17 years and patients with PCOS.

Materials and research methods: 11 patients participated in research with PCOS and 68 healthy girls. At all girls except standard inspections, determined the AMH level in blood serum by Hoffman — La Rosh test system the IFA method.

Results of research. At healthy girls average concentration of AMG in serum of blood made 4.7 ± 0.32 $\mu\text{U/ml}$ ($\sigma = 2.65$), with fluctuations from 0.23 to 14.9 $\mu\text{U/ml}$ and a median 4.0. At girls with PCOS the average level of AMH in serum was almost twice higher in comparison with healthy girls and was equal 8.5 ± 1.96 $\mu\text{U/ml}$ ($\sigma = 6.5$), with fluctuations from 3.5 to 27.3 $\mu\text{U/ml}$ and a median 7.1. When comparing the AMH level of healthy girls and patients with PCOS a reliability of the received distinctions, as is established when carrying out the one-factorial dispersive analysis ($p=0.001$).

Thus, the AMG level is a peculiar indicator of physiological functioning of ovaries at girls of teenage age and it is possible, can play a role of a preclinical marker at girls with a being formed PCOS.

P141

A STUDY OF BMI AND MENSTRUAL CYCLE IN ADOLESCENTS, JAKARTA, INDONESIA

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Introduction: Menstrual cycle is used as a sign of women's health. The length and menstrual regularity are caused by many factors, one of which is BMI. Some of studies said underweight or overweight is associated with short or long cycles in menstruation

Objective: The study aims to determine the relationship between BMI and menstrual cycle in adolescents in Jakarta, Indonesia.

Methods: This was a cross-sectional study, including 372 female adolescents, aged range 12-18. They filled a questionnaire to detect the menstrual pattern and BMI. The data was analyzed with Chi-square, using SPSS version 17. The BMI was divided into 2 groups, normal and underweight. Each category was associated with menstrual cycle durations.

Results: The median age of participants, menstrual cycle length, and BMI were 15 years, 21-35 days, and 19.1 kg/m². Most of subjects had normal BMI ($n=259$, 69.6%) and underweight ($n=113$, 30.4%). The menstrual cycle lengths, 15.6% ($n=58$) had short cycle (< 21 days) and 4.8% ($n=18$) had long cycle (>35 days). There was no significant association between normal or underweight BMI and menstrual cycle durations ($p>0.05$, $p= 0.4$). In normal BMI, 80.3% of subjects had normal menstrual cycle, 15.8% short cycle, and 3.9% long cycle. In underweight category, 77.9% of participants had normal cycle, 15% short cycle, and 8% long cycle.

Conclusion: The majority of adolescents in Jakarta had a normal menstrual cycle. Even though, there was no difference in menstrual cycle between 2 groups, normal and underweight BMI.

Keywords: BMI, menstrual cycle, adolescent

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THE MENSTRUAL PATTERNS AMONG ADOLESCENTS IN JAKARTA

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Background: A variety of problems can arise during menstruation period, such as dysmenorrhea, menstrual irregularity, and menorrhagia. Many adolescents complained about these gynecological problems. The abnormality of menstrual patterns might be a sign of women's health problems, such as endometriosis, and it needs a proper treatment.

Objective: The study aims to describe generally about menstrual patterns in female adolescents in Jakarta.

Method: This was a cross-sectional study, using questionnaire distributed to 5 schools in Jakarta, Indonesia. Subjects of this study were junior and high school students, aged range 12-18, who already got menstruation period.

Result: The mean age of 402 participants was 15 (32.3%). The age of menarche between 11-13 years, 84.1% (n=338). The median of menstrual duration was 3-7 days, 69.2% (n=278). 313 of female adolescents (79%) had normal duration of menstrual cycle, 21-35 days. Number of changing pads every menstruation was 3 times a day, 55.2% (n=222). 62.4% participants (n=251) got menstrual pain in every menstruation. The menstrual pain was felt the most during menstruation (n=238, 59.2%) with the median pain scale was mild pain (scale 1-4), 66.4% (n=267). The majority of them didn't consume any drugs, 87.1% (n=350).

Conclusion: The menstrual patterns of adolescents in Jakarta, classified as normal by the age of menarche, 11-13 years, the menstrual duration between 3-7 days, menstrual cycle 21-35 days, and 3 times changing pads everyday during menstruation.

Keywords

Menarche, menstrual duration, menstrual cycle, changing menstrual pad, dysmenorrhea

P143

CORRELATION BETWEEN OBESITY, DYSMENORRHEA AND PREMENSTRUAL SYNDROME AMONG INDONESIAN ADOLESCENCE

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Introduction: There is no study explaining menstrual pattern among Indonesian adolescence yet. Obesity is thought to be one of risk factor for dysmenorrhea and premenstrual syndrome; however larger study is needed. The aim of the study was to develop base line character of menstruation pattern of Indonesian adolescence population besides to confirm whether obesity is linked with dysmenorrhea and premenstrual syndrome or not.

Methods: We used guided questionnaire to 448 girls, 12-18 year old (mean 15 years old). We did descriptive analysis and bivariat analysis using chi square. The determined p value was <0.05.

Result: Majority of the population had their menarche at 11-13 year old (83.3%). The major menstrual duration was 3-7 days (69%) with major periods 21-35 days (75.7%), and most of subjects change pad 3 times daily (53.3%). Prevalence of dysmenorrhea in this study was 57.1% with 78.9% had Visual Analog Scale (VAS) 1-3, and 65% accompanied by only one premenstrual symptom. Most subjects had BMI <18 (42,4%). The correlation between BMI and dysmenorrhea, between

BMI and VAS score, and between BMI and number of premenstrual syndrome symptom were not statistically significant (p value >0.05).

Conclusion: This study proved that there was no correlation between obesity, dysmenorrheal, and premenstrual syndrome.

Keyword: obesity, dysmenorrhea, premenstrual syndrome, menstruation, Indonesia

P144

IVF OUTCOME IN SELF STIMULATED PATIENTS WITH LOW VERSUS NORMAL AMH LEVELS

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Objective: To determine whether low AMH level can give satisfactory oocytes and good IVF outcome compared with normal AMH level patients.

Design: A total of 300 IVF-ICSI cycles at our centre from February 2012 to February 2013 irrespective of antagonist or agonist protocols were prospectively studied with computer generated database. Amongst them 150 patients were having AMH level between 0.3 to 2 ng/ml and 150 patients were having more than 2 ng/ml.

Material and Method: 300 patients with the age group between 25 -40 years in two groups based on reduced (0.3-2 ng/ml) versus normal (more than 2 ng/ml) AMH levels were studied for total HMG dose requirement, total number of oocytes retrieved, total numbers of M2s and embryos and their IVF outcome.

Results: Though low AMH level group required high doses of HMG and reduced numbers of total oocytes, M2s and embryos, clinical pregnancy rates and take home baby rates were nearly same than those of the patients with the normal AMH levels in in younger age group patients (25 to 35 years),

Conclusion: Low AMH alone does not portray grave prognosis for the IVF outcome. These patients can be reassured of good outcome with their own eggs and not necessarily subjected to donor egg cycle to begin with.

P145

ISOLATED LOW FOLLICLE STIMULATING HORMONE (FSH) IN INFERTILE MALES – A PRELIMINARY REPORT

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Objectives: High levels of follicle stimulating hormone (FSH) in infertile males received a significant attention and exploration. Studies investigating the isolated deficiency of FSH in males are few, and its real prevalence is still unknown. Therefore, the objectives of the current study was to report the prevalence of isolated low FSH in infertile males and highlight their demographics and standard sperm parameters.

Methods: Records of 3335 infertile men were retrospectively checked. Patients with isolated low FSH were retrieved. FSH levels were categorized into 3 groups based on the number of affected sperm parameter (s). Study variables were also arranged into 2 groups in relation to smoking history. A control group was included to document the changes in sperm morphology.

Results: Isolated low FSH (1.146 ± 0.219 mIU/mL) was found in 29 (0.87%) patients. All patients showed at least one abnormal sperm parameter. The abnormal parameters were present in different combinations within the same patient but with no significant correlations with the FSH levels. The FSH levels got lower as the number of the affected sperm parameters increased although the decline was insignificant. The most frequent abnormal parameter presented was sperm morphology (86.2%). Anomalous sperm morphology was highly and significantly demonstrated in the head; specifically in

acrosome. Abnormal sperm parameters were present in both smoking and nonsmoking groups but with no significant differences in between.

Conclusions: Isolated low FSH among infertile males has a low prevalence. This may be associated with abnormality in semen parameters; particularly sperm morphology. These patients are suggested to be found as a primary entity. However, an additional work-up is highly recommended to validate this hypothesis

P146

PROGESTIN INDUCES A TRANSCRIPTION FACTOR HAND2 IN HUMAN ENDOMETRIAL STROMAL CELLS.

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OBJECTIVES: A transcription factor Hand2 is known to play a key role in uterine receptivity and to be induced in the mouse uterus during decidualization. We investigated the regulation of Hand2 mRNA expression in human endometrial stromal cells (ESCs) during decidualization and the functional roles by silenced Hand2 with small interfering RNA technology.

METHODS: All human samples were obtained using a protocol for the protection of human subjects approved by the Institutional Review Board, together with informed consent from all patients. ESCs were cultured with estradiol (E), medroxyprogesterone acetate (MPA), and/or Br-cAMP. Real-time polymerase chain reaction assessed the expression levels of Hand2 and decidual specific genes, including fibulin-1 and prolactin.

RESULTS: The mRNA levels of Hand2, fibulin-1, and prolactin were significantly induced in ESCs stimulated with E+MPA after 1, 3, and 12 days of culture, respectively. MPA induced Hand2 mRNA levels in ESCs in a time- and dose-dependent manner, and this stimulatory effect was blocked by RU-486 (P receptor antagonist). Simultaneous MPA and Br-cAMP treatment synergistically enhanced Hand2 mRNA levels, whereas E or Br-cAMP alone had no effect. Silencing Hand2 expression significantly reduced fibulin-1 after 3 days of E+MPA treatment. However, the silencing of Hand2 had no effect on the levels of cell proliferation.

CONCLUSION: These results indicate that Hand2 is induced during decidualization earlier than fibulin-1 and prolactin. Progesterone-induced Hand2 contributes to fibulin-1 expression in human ESCs.

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IDENTIFICATION AND CHARACTERISATION OF PROGESTERONE-INDUCED MICRORNAS IN MOUSE ENDOMETRIAL EPITHELIAL CELLS

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It is essential for progesterone to inhibit estrogen-induced proliferation of endometrial epithelial cells (EECs) and make endometrial epithelium to convert into the secretory epithelium for the preparation of embryo implantation. But the mechanism of progesterone effect on EECs remains unclear. Recent studies have indicated that some reproductive hormones can regulate the expression of some special genes in post-transcriptional level. MicroRNAs are important post-transcriptional regulators in the target mRNA, thus they are involved in many physiological and pathological processes. We speculate that there should be progesterone-induced microRNAs which mediate the physiological effect of progesterone on EECs.

In this project, total RNAs have been respectively extracted from endometrial epithelial cells of ovariectomized mice which were treated by only estrogen or by both estrogen and progesterone. Using microRNA high-throughput sequencing combined with in silico prediction, we found 149 progesterone-induced microRNAs. KEGG pathways analysis showed those microRNA are involved in focal adhesion, endometrial cancer, positive regulation of transcription, cell cycle and so on.

Furthermore, we have validated 9 microRNAs (miR-23a-3p, mmu-miR-26a-5p, mmu-miR-1a-3p, mmu-miR-133a-3p, mmu-miR-195a-5p, mmu-miR-3473b, mmu-miR-204-5p, mmu-miR-145a-5p, mmu-miR-143-3p) with real-time PCR.

In our conclusion, progesterone can specifically regulate the expression of some miRNAs in EECs, which possibly mediate the effect of progesterone on the structure and function of endometrial epithelium for preparing for implantation through influencing some functional proteins. This project will be helpful for further exploring the molecular mechanism of progesterone's effect on EECs.

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DISORDERED LIPID FLUXES IN ENDOMETRIOSIS RELATED INFERTILITY

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Peritoneal fluid from endometriotic women reduces implantation rates in animal models, suggesting key factors in inducing endometriosis-associated infertility. However, the identity of these factors is unknown. Our aim is to clarify the role of peritoneal fluid sphingolipids in endometriosis-associated infertility.

Peritoneal fluid from 15 fertile (8 endometriosis and 7 without) and 33 infertile women (21 with endometriosis and 12 without) were collected during laparoscopic surgery.

Advanced liquid chromatography-mass spectrometric methods were deployed to measure more than 80 peritoneal fluid sphingolipids in a single assay. False-discovery rate were kept at less than 15% to identify true significant sphingolipids. Two-way ANOVA was used to test for interactions between endometriosis and infertility.

We quantified unambiguously more than 80 sphingolipids simultaneously. False-positives were controlled via adjusted p-values. We found elevated peritoneal fluid glucosylceramides in infertile endometriotic women relative to infertile controls, congruent with the mitogenic properties of glucosylceramides. There were no significantly abundant sphingolipids between fertile controls and women with endometriosis. Importantly, comparing fertile controls versus infertile endometriotic women revealed 50-130% higher levels of peritoneal fluid ceramide-1-phosphates, ceramides and lactosylceramides, indicating the interaction of endometriosis and infertility on these sphingolipids. In addition, the significance of these sphingolipids increased with severe endometriosis.

In this study we provide the first clinical evidence that the PF infertile women with endometriosis contain higher amounts of sphingolipids that may be embryotoxic or causes defective implantation through stromal dysfunction, and in turn, contributing to infertility. Understanding the underlying altered sphingolipid metabolism and managing it may significantly improve endometriosis associated infertility.

P149

THE LAPAROSCOPIC TREATMENT OUTCOME OF FALLOPIAN TUBE ACCESSORY OSTIUM

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Research background: Fallopian tube accessory ostium is a rare fallopian tube disease. It is present when an ectopic fimbria is seen at some distance from the fimbriated end. The pathogenesis of accessory ostium may be related with endometriosis and the cause of infertility may be the loss of ovum. This article is aimed to investigate the diagnosis and treatment outcome of accessory ostium

Research method: Retrospective analysis of 21 accessory ostium patients who received laparoscopy in Peking University People's Hospital from 2010.1 to 2012.5. 2 patients who had to use contraception method after myomectomy and 1 patient lost follow-up are excluded. 18 patients were proved to have endometriosis during laparoscopy including 16 early stage cases and 2 cases with endometrial cyst. Two surgical techniques were used: 1 purse-string suture and accessory ostium resection 2 accessory ostium opening and fimbrioplasty. GnRH-a were given after surgery to prevent the recurrence of endometriosis. All patients are followed up 12-19 months to analyse pregnancy rate, abortion rate and ongoing pregnancy rate

Research result: 12 patients conceived after surgery, 10 patients conceived spontaneously and 2 patients received IUI. Pregnancy rate is 66.67%(12/18). Abortion rate is 5.56%(1/18) and the ongoing pregnancy rate is 61.11%(11/18).

Research conclusion: Accessory ostium is a rare disease lacking clinical feature. A certain diagnosis is made during careful investigation of fallopian tube in surgery. Accessory ostium has a close relationship with endometriosis and infertility. The treatment is laparoscopy surgery and post-surgery medical therapy to reduce estrogen level. The treatment outcome is satisfactory

P150

THE ROLE OF ESTROGEN METABOLISM IN ENDOMETRIOSIS

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Estrogen hormone has suggested play an important role in endometriosis, since endometriosis can be considered as an estrogen dependent inflammatory disease.

This study would like to observe the role of estrogen metabolism by measuring the level of estradiol (E2), estrone (E1), estriol (E3), and the ratio between E2:E1, E1:E3 and E2:E3.

We did cross sectional study on endometriosis patients that was being treated in Dr. Cipto Mangunkusumo General Hospital, Jakarta. 27 endometriosis patients that have been proved by either laparotomy or laparoscopy and 27 patients that underwent either laparotomy or laparoscopy not due to endometriosis or other disorders that related with estrogen as a control group were recruited. Estrogen hormones were measured by ELISA technique.

The level of E2, E1 and E3 were found lower in endometriosis patient compared to control group despite no statistically significant (29 vs. 35 pg/mL, p 0.815; 54.7 vs. 73.5 pg/mL; and 1.11 vs. 1.67 pg/mL). The ratio between E2:E1 was found higher in endometriosis patients despite no statistically significant (0.51 vs. 0.38, p 0.164). The ratio between E1:E3 was found higher in endometriosis patients despite no statistically significant (58.6 vs. 50.3, p 0.684). Finally, the ratio between E2:E3 was found higher in endometriosis patients despite no statistically significant (26.5 vs. 21.1, p 0.223).

This study basically has confirmed the previous knowledge about the involvement of estrogen metabolism on endometriosis. However, there is a challenge to explain why the level of estrogen hormones in endometriosis patients was lower compared to control group.

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THE CORRELATION BETWEEN CLINICAL MANIFESTATION, BIOCHEMICAL MARKER (CA 125), DIAGNOSTIC LAPAROSCOPY AND/OR LAPAROTOMY FINDINGS AND HISTOPATHOLOGICAL FINDINGS FOR THE DIAGNOSIS OF ENDOMETRIOSIS

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OBJECTIVE

To develop the Cli-Endomet as a reliable tool in the diagnosis of endometriosis. It was performed to evaluate the association and identify the prognostic factors between the clinical manifestation and biochemical marker (CA 125) in the diagnosis of endometriosis.

METHODS

This cross sectional study was conducted from 1st October 2011 until 31st March 2013. It consisted of 176 patients who presented with pelvic pain (dysmenorrhea, dyspareunia, ovulation pain, dyschezia), and/ or infertility. All patients were examined physically and ultrasonography was performed. Blood sample for CA-125 was taken. Operation laparoscopy or laparotomy was performed and/or tissue biopsy was taken if available for histopathology examination. The staging of endometriosis was determined. The clinical criterias which were strongly associated with diagnosis of endometriosis were extracted, and were transformed for development of Cli-Endomet.

RESULTS

Among 176 patients, 103 patients were diagnosed as endometriosis, and were staged according to the revised classification of the American Fertility Society (rAFS), with stage I, 3.8%; stage II, 11.7%; stage III, 41.8%; and stage IV, 42.7%. Among the clinical manifestation and biochemical marker (CA 125), the statistically significant (p value < 0.05) criterias included : Dysmenorrhea (mild, p value : 0.006; moderate, p value : 0.030; severe, p value : 0.001), ground-glass appearance (or thick with sediments) on ultrasonography (p value : 0.001) and CA 125 (p value : 0.000).

CONCLUSION

Cli-Endomet was created as a diagnostic tool to diagnose endometriosis, with benefit of inexpensive test and avoiding patient from surgical procedure for confirmation, and the appropriate treatment could be started with certainty.

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OVARIAN RESPONSE IN IVF CYCLE AFTER LAPAROSCOPIC OR LAPAROTOMY CYSTECTOMY FOR ENDOMETRIOTIC CYST

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Residual ovarian function after ovarian excision of endometriotic ovarian cyst is still controversial. This study was carried out to investigate whether ovarian cystectomy interferes with follicular recruitment and the number of oocytes retrieved in an in-vitro fertilization (IVF) cycle. Data from IVF patients between January and September 2013 in Melati Clinic, Harapan Kita Hospital were reviewed. Patients were included who previously underwent laparoscopic or laparotomy excision of a monolateral endometriotic ovarian cyst. The operated ovary and contralateral intact ovary were compared in terms of number of basal antral follicles and the number of oocytes retrieved after stimulation in IVF cycle. A paired Student T-test was used to investigate differences between two ovaries. In total, 8 patients were identified. The mean (\pm SD) number of basal antral follicles was 4.5 ± 1.9 in the control ovary and 3 ± 2.1 in the previously operated ovary ($P < 0.002$). Meanwhile, the mean (\pm SD) number of oocytes retrieved after stimulation in IVF cycle (3.3 ± 2.0) in the control ovary and (2.1 ± 1.1) in the previously operated ovary was not statistically significant ($P < 0.166$). Excision of endometriotic ovarian cysts is associated with a significant reduction in ovarian reserve, but does not affect the ovarian response after stimulation in IVF cycle.

P153

ASSOCIATION OF THE MIF POLYMORPHISMS WITH RISK OF ENDOMETRIOSIS IN IRANIAN WOMEN

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Background

Macrophage migration inhibitory factor (*MIF*) is a key pro-inflammatory cytokine that is secreted by active macrophages in ectopic tissue of endometriosis. *MIF* is a mitogenic factor that promotes angiogenesis, as well as plays an essential role in cell proliferation by negative regulation of p53 and stimulates the synthesis of PGE₂, leads to elevate the local estradiol synthesis. We aimed to evaluate the association between *MIF* polymorphisms with endometriosis.

Methods

Polymorphisms were studied in 57 endometriosis patients and 70 healthy controls. Restriction fragment length polymorphism (RFLP) was applied to determine -173G/C polymorphism. -794 (CATT)₅₋₈ and ORF region were detected by sequencing. Statistical analysis was done by Chi-square test.

Results

In this study five polymorphisms were identified including 2 promoter SNPs, +254rs2096525 at intron one (p=0.73), +656rs2070766 at intron two (p=0.94), +624 rs35625249 at intron2 (P=0.05).

Both allele and genotype frequencies of -794 (CATT)₅₋₈ and -173G/C SNPs were not significantly different (p>0.05) between two groups. Although, haplotype frequency of -794 and -173 was significantly different in control group (p=0.001) and endometriosis patients (p=0.004). -794(CATT)₅ genotype was only observed in control group.

Conclusions

In conclusion we report for the first time that both increased number of CATT repeat in -794 position and presence of -173 C polymorphism in endometriosis patients, which can be resulted in recently reported high-expression of *MIF* in endometriosis. Therefore it seems the polymorphisms of *MIF* promoter and +624 rs35625249 SNP might be considered as an important factor in pathophysiology of endometriosis.

key words:

Endometriosis, Macrophage migration inhibitory factor, Polymorphism

P154

FOLLICULOGENESIS OF ENDOMETRIOSIS PATIENTS POLYMORPHISM VARIABLE NUMBER TANDEM REPEAT INTRON4 GENE CYP19

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The frequency of endometriosis patients with infertility are around 20-50% with primary infertility and 15% with secondary infertility. One of the cause of infertility is folliculogenesis. The growth and development of the follicle needs synergic

interaction between estrogen and gonadotropin hormone. Variations or CYP19 gene polymorphisms cause changes in expression activity of cytochrome P450 enzyme aromatase, which can lead in estradiol production.

Polymorphism variations are analyzed by Gene Scanning. The subject of this study consists of 25 individuals as a control group of endometriosis patient and 25 as a non endometriosis patient group.

From gene scanning, it was shown that lengths of fragments were 152-191 bp in accordance with (TTTA) in repeat 2-12. In the endometriosis group there are 7 polymorphism groups (1) 6/6, 6/6, 6/8, 6/11, 2/11, 7/12 and 11/12, most cases are 6/11 genotype or (TTTA)6(TTTA)11 (8 patients, 32%) and the most control groups are 6/6 genotype or (TTTA)6(TTTA)6 (8 patients, 32%). In control group, the dominant homozygote genotypes were 6/6 and 7.3/7.3 is 32 and 12%; endometriosis genotype 6/6 groups was 16%. This study found long allele polymorphism frequency ≥ 175 bp higher in patients with endometriosis with significant difference. OR=4.57; (95% CI: 1.25 – 16.69).

Conclusion: that there is a significance difference in polymorphism (TTTA)_n repeat intron 4 gene CYP19 correlate with folliculogenesis between endometriosis patient group and non endometriosis patient group.

Keywords: Endometriosis, Polymorphism gene CYP19 and Folliculogenesis

P155

ROLE OF ADIPONECTIN IN ENDOMETRIOSIS

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BACKGROUND: Endometriosis is one of the most common gynecological disorder known to be related with complain of infertility, dysmenorrhea and chronic pelvic pain. Etiopathogenesis of endometriosis is not fully understood yet. Genetic factor may contribute to the pathogenesis of endometriosis. Adiponectin is substance produced by adipocyte tissue which promote insulin sensitivity and scavenger activity which involves in endometriosis pathogenesis. There is little evidence on correlation between adiponectin receptor gene polymorphism with endometriosis.

PURPOSE: To describe correlation between adiponectin including plasma level, gene polymorphism and gene receptor polymorphism, with risk of endometriosis.

DESIGN AND METHOD: This is a case-control study. The case group consist of 30 reproductive age women with endometriosis while the control group consist of 34 women without endometriosis. Detection of adiponectin plasma level was performed using ELISA technique. Snp detection of adiponectin and adipoR2 gene were performed by PCR-RFLP technique. Genotype distribution was compared between two groups.

RESULT: Sixty-three samples were obtained, 30 cases and 34 controls. In cases group A/A genotype was found in 5 (16,7%) subjects, A/T in 18 (60%) subjects and T/T in 7 (23,3%) subjects. In control group, A/A genotype was found in 8 (23.5%) subjects, A/T in 21 (61,7%) subjects and T/T in 5 (14,7%) subjects.

CONCLUSION: No correlation between adiponectin with risk of endometriosis, in terms of plasma adiponectin level, adiponectin gene and adipoR2 gene polymorphism.

Keywords: Adiponectin, AdipoR, endometriosis, polymorphism

P156

THE ROLE OF DNMT3A IN THE PROGRESSION OF HUMAN ADENOMYOSIS

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OBJECTIVE: DNA methylation is an approach of epigenetic modification in the process of gene expression regulation. Increasing evidences have showed that the aberrant expression of DNA methyltransferases (DNMTs) was involved in the development of diverse human disorders. The aim of this study was to investigate the role(s) of DNA methyltransferase 3 alpha (DNMT3a) in the progression of human adenomyosis.

METHODS: Select the ectopic endometrial tissues from 20 cases with adenomyosis and the eutopic endometrial tissues from 20 cases without endometriosis, RT-PCR and Western blotting (WB) were used to detect the mRNA and protein expression of DNMT3a; isolating primary endometrial stromal cells (ESCs) and WB was used to detect the differential protein expression of DNMT3a, then constructed the lentiviral vector of DNMT3a, infected the ESCs to regulate the protein expression of DNMT3a, and detected the functional effects of the transgenic cells.

RESULTS: (1) Compared with controls, the mRNA and protein expression of DNMT3a in patients with adenomyosis were significantly down-regulated. (2) The protein expression of DNMT3a in the ESCs from adenomyosis was significantly lowered than controls. (3) DNMT3a lentivirus overexpression in the primary ESCs from adenomyosis exhibited lower proliferation ability and decreased cell invasion and migration ability when compared with ESCs from controls.

CONCLUSIONS: The aberrant expression of DNMT3a played crucial roles in the development of adenomyosis; however, further studies would be needed to discern the underlying mechanism.

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THE ROLE OF ABNORMAL PROMOTER METHYLATION AND GENE EXPRESSION OF 14-3-3 ζ IN THE PROGRESSION OF HUMAN ADENOMYOSIS

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OBJECTIVE: We hypothesized that DNA methylation might play certain roles in the development of human adenomyosis. The present study was to investigate the role of abnormal promoter methylation of 14-3-3 ζ in the progression of human adenomyosis.

METHODS: Select the ectopic endometrial tissues with adenomyosis, and eutopic endometrial tissues without endometriosis as control, isolate primary endometrial stromal cells (ESCs) and then Western blotting (WB) and methylation-specific PCR (MSP) were used to detect the expression of 14-3-3 ζ and the promoter methylation, respectively; constructed the lentiviral vector of DNA- methyltransferase 3 alpha (DNMT3a), infected the ESCs to obtain transgenic cells, detect the protein expression of 14-3-3 ζ and the methylation level of 14-3-3 ζ gene promoter. Subsequently, detected the functional effects of the transgenic cells.

RESULTS: Compared with control stromal cells: (1) the expression of 14-3-3 ζ protein in the stromal cells of adenomyosis was increased significantly, and 14-3-3 ζ gene promoter was demethylated. (2) DNMT3a lentiviral vector significantly down-regulated 14-3-3 ζ protein expression and increased the methylation level of 14-3-3 ζ in the promoter region. (3). The viability

of the transgenic cells was significantly lowered, and the results of transwell test showed that cell invasion and migration were significantly down-regulated.

CONCLUSIONS: Combined with previous reports, the present study supported that the aberrant methylation of 14-3-3ζ gene promoter played crucial roles in the development of adenomyosis; however, further studies would be needed to discern the underlying mechanism.

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DOWN-REGULATION OF TENSIN 1 GENE EXPRESSION IN HUMAN ENDOMETRIOTIC TISSUE FOLLOWING GNRH AGONIST TREATMENT

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Aim:

Identify a genomic biosignature of endometriosis patients with and without GnRH agonist (GnRHa) treatment in order to elucidate the molecular change based on the level of patients' gene expression profiles. We hypothesized that Tensin 1 would be transcriptionally down regulated in endometriosis patients receiving GnRHa treatment.

Methods:

Affymetrix microarray chip was used to identify genes differentially in human endometriotic tissues with and without GnRH treatment. The candidate genes were independently validated by qRT-PCR. Analysis of gene expression profile in endometriotic tissue with (n=35) and without GnRH treatment (n=34) was done. The subsequent primary culture experiments were done from eutopic and ectopic endometrial tissues and divided into two groups: with and without estradiol (E2) treatment to investigate the hormonal function related gene. Cell migration assay was done to look at the role of gene in endometriosis.

Results:

Sixty-five genes exhibited alterations in expression following GnRHa treatment. After validating all 65 genes from microarray data, the expression of Tensin 1 from tissues of patients with GnRHa treatment was significantly down-regulated compared with control (P=0.01). The expression of Tensin 1 in ectopic endometrial cells was up-regulated by E2 compared with control (P=0.03). The function of Tensin 1 is related with cell migration.

Conclusions:

The expression of Tensin 1 was down-regulated following GnRHa treatment and up-regulated following E2 treatment. Tensin 1 is an important component linking ECM, the actin cytoskeleton, signal transduction and mediates signaling for cell motility and migration. This finding broadens our understanding of pathogenesis of endometriosis related with cell migration.

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THE ROLE OF GENE PROMOTER DEMETHYLATION OF 14-3-3 ZETA, SF-1 RECEPTOR AND ER-BETA RECEPTOR IN ECTOPIC ENDOMETRIAL STROMAL CELLS OF ADENOMYOSIS

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OBJECTIVE: This aim of this study was to investigate the role of the promoter demethylation situations of 14-3-3 ζ , SF-1 receptor and ER-beta receptor in the pathogenesis of adenomyosis and to further explore the related genes that promoter demethylation in ectopic endometrial stromal cells of adenomyosis.

METHODS: Select the ectopic endometrial tissues with adenomyosis and eutopic endometrial tissues without endometriosis as control, separated into single ESCs and WB and MSP were used to detect the expression of 14-3-3 ζ , SF-1 receptor and ER-beta receptor and the promoter methylation situation. Additionally, we also detected the functional effects of the ESCs.

RESULTS: Compared with control ESCs: (1) The protein expression of 14-3-3 ζ , SF-1 receptor and ER-beta receptor were all significantly higher in the ectopic ESCs; (2) The promoter CpG islands of these three genes were all existed demethylation condition in the ectopic ESCs; (3) The viability of ectopic ESCs was enhanced, the invasion and migration of ectopic ESCs were significantly up-regulated; and the results of apoptosis showed no significant difference.

CONCLUSIONS: The gene promoter CpG islands of 14-3-3 ζ , SF-1 receptor and ER-beta receptor in the stromal cells of adenomyosis were presneted demethylation. The protein expression of these genes were up-regulated. It affected the etiological and molecular biological processes of adenomyosis. However, further studies of the underlying mechanism are necessary.

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EXPRESSION AND CLINICAL SIGNIFICANCE OF 14-3-3 PROTEIN FAMILY IN CHOCOLATE CYST OF OVARY

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OBJECTIVE: To investigate the role(s) of 14-3-3 proteins isoforms played in the development of chocolate cyst of ovary.

METHODS: To selecte 30 patients with chocolate cyst of ovary and 30 controls, respectively. RT-PCR was firstly used to examine the mRNA expressions of all seven 14-3-3 isoforms in the affected and normal endometriums; for those differentially expressed isoforms, WB was used to detect their protein expression.

RESULTS: All of the seven 14-3-3 isoforms mRNA were detected in both the chocolate cyst of ovary ectopic endometrium and normal endometrium. When compared with normal endometrial tissues, the 14-3-3 σ mRNA was significantly decreased ($P<0.05$) while the 14-3-3 ϵ and 14-3-3 ζ mRNA were significantly increased ($P<0.01$) in the chocolate cyst of ovary ectopic endometrial tissues; accordingly, the protein expression of 14-3-3 σ , 14-3-3 ϵ and 14-3-3 ζ exhibited the same expression trend as their mRNA expression.

CONCLUSIONS:(1) All of the seven 14-3-3 isoforms were expressed in both the chocolate cyst of ovary ectopic endometrium and normal endometrium. (2) When compared with normal control tissues, the mRNA and protein expressions of 14-3-3 σ decreased in chocolate cyst of ovary ectopic endometrial tissues, and the mRNA and protein expressions of 14-3-3 ϵ and 14-3-3 ζ were significantly increased. Our results suggested that 14-3-3 σ , ϵ , and ζ might be involved in the development of the chocolate cyst of ovary.

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STUDY OF COGNITIVE-BEHAVIORAL INTERVENTION MODEL ON REPRODUCTIVE HEALTH AMONG FEMALE FLOATING POPULATION IN CHINA

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Objective: Investigate the cognitive-behavioral intervention model to improve the reproductive health among female floating population in China, providing the basis of a higher reproductive health cognitive-behavioral level.

Methods: Randomly select 360 child-bearing aged female from floating population of a community in Changsha City, Hunan Province entering the group: one control group and two intervention groups, with 120 cases in each group. Intervene the objects with the "knowledge, attitude, belief" theoretical model. Intervention group one is intervened by releasing materials and providing knowledge lectures. While group two, besides the treatments used in group one, is also intervened with consultation?regular telephone or home follow-up. Control group is free from intervention.

Results: One and three months after intervention, the two intervention groups get higher reproductive health cognitive-behavioral scores and higher scores in every dimension ($p < 0.01$). In the comparison between the two intervention groups, group two gets higher score only in knowledge dimension ($p < 0.01$).

Conclusion: Intervention is effective in improving the reproductive health cognitive-behavioral level, while releasing materials, providing knowledge lectures is simple and more suitable for popularization and application in floating population.

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STUDY OF DEPRESSION, ANXIETY, HAPPINESS AND LIFE SATISFACTION IN WOMEN UNDERGOING ASSISTED REPRODUCTIVE TECHNOLOGY IN AN IRANIAN INFERTILITY TREATMENT CENTER

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Introduction:

Infertility is a biopsychosocial crisis for people who want children. Most studies showed that attention to the psychological aspects during assisted reproductive treatment is strongly advisable. our aim was to determine the levels of depression, anxiety, happiness and life satisfaction in infertile women of Kermanshah in Mo'tazedi Hospital.

Methods:

In this descriptive-analytic study, a total of 130 women that visited our center between April and December 2012 were selected by using of convenience sampling. Depressive and anxious symptoms were assessed with Beck depression inventory (BDI) and Beck anxiety inventory (BAI). For happiness and life satisfaction assessment we used Oxford happiness inventory (OHI) and satisfaction with life scale (SWLS) questionnaires.

Results:

Our results showed that 45 woman (34/6 Percentage) were not depressed and 41 woman (31/5 Percentage) had mild to sever depression. Also 20 women (15/4 Percentage) were not anxious and 74 woman (56/9 Percentage) had anxiety. Mean score of happiness and life satisfaction was 30/98 and 20/10, that these scores are significantly lower than common people scores ($t=9/59$; $p < 001$) ($t=3/94$; $p < 001$).

Conclusions:

Data showed that depressive and/or anxious symptoms in our study were worthy of note and should not be underestimated. While happiness and life satisfaction is lower than the general population. This study showed that these women are at risk of psychological problems and even improvement in their mental health could affect on their infertility treatment. So, the women with a history of infertility might benefit from psychological intervention.

Key words: infertility, depression, anxiety, happiness, life satisfaction

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ETHICS IN ASSISTED REPRODUCTIVE TECHNOLOGY

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Ethics in Assisted Reproductive Technology

Abstract

Assisted Reproductive Technology has both an applied and a research aspect to it. The tenets of ethics, namely; Beneficence/Non –Maleficence, Justice and Autonomy apply to the IVF patient, and the embryo in both applied and research scenarios. Aspects of Ethics in ART such as informed consent, proper counselling, proper Quarantine of donor gametes, health of the gestational surrogate, and parental status are all important ethical issues in the applied field of ART and will be addressed in a comprehensive manner.

The ethical issues regarding the research aspect of Assisted Reproductive Technology are of equal importance and will be discussed by addressing issues such as the moral status of the embryo; and the ethical theories that enable a biological organism to obtain moral status. The impediments caused by international and national regulation to carry out any research pertaining to Human Embryonic Stem cells will also be discussed. The ethics behind using Human Embryonic Stem cells for research purposes will be discussed. Most importantly; the recent development of Induced pluripotent Stem cells and their ability to bypass the ethical obstacles faced while using Human Embryonic Stem cells will be discussed.

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OFFICE FLEXIBLE HYSTEROSCOPY VERSUS SALINE INFUSION SONOGRAPHY FOR UTERINE CAVITY SCREENING ASSESSMENT AT INFERTILITY PATIENT

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Objective: To compare predictive values of flexible hysteroscopy (FH) and saline infusion sonography (SIS) for the detection of abnormalities in outpatients undergoing uterine cavity assessment. Materials and methods: A total of 83 women underwent uterine cavity assessment using office FH and/or SIS were included. After operative procedure (dilatation and curettage and biopsy, hysteroscopic resection or pelvicoscopic resection), histopathologic results were used as a gold standard to calculate predictive values of FH and SIS. Results: Of 83 patients, 71 and 67 underwent FH and SIS, respectively. Both procedures were done in 55 patients. Transvaginal ultrasonography findings before procedure were endometrial polyp (42.2%), thickened endometrium (31.3%), submucosal myoma (14.5%), and endometrial fluid (1.2%). Histological diagnoses were endometrial polyps (43.4%), normal range (39.8%), leiomyoma (14.5%), and adenocarcinoma (2.4%). As a screening test for uterine cavity assessments, FH had 100% sensitivity, 88.9% specificity, 93.2% positive predictive value (PPV) and 100% negative predictive value (NPV), and SIS had 90.5% sensitivity, 87.5% specificity, 92.7% PPV and 84% NPV. Office FH showed higher sensitivity and NPV than SIS, but these differences were not statistically significant. Diagnostic accuracy of FH and SIS were 89.7% and 84.8 %, respectively. Conclusion: Our results suggest that office FH and SIS both have good predictive values for uterine cavity assessment. Office FH, easily performed and provides direct intrauterine visibility, could be an optimal option for outpatient screening evaluation of uterine cavity for infertility patient.

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EFFECT OF THERMAL STRESS ON PHYSIOLOGICAL AND REPRODUCTIVE PARAMETERS IN BOS INDICUS, CROSS-BRED AND BOS TAURUS DAIRY COWS – A PRELIMINARY STUDY

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Fertility of the dairy cattle can be severely affected when exposed to elevated ambient temperatures. This study was conducted to assess physiological and reproductive changes, in relation to heat stress in different dairy cattle breeds. A total

of thirty six (Nine dairy cows from each breed of Sahiwal, Achai, Cross-bred and Pure-exotic) lactating dairy cows were selected. Physiological parameters (rectal temperature, respiratory rate and pulse rate) were monitored. The intensity of changes of all physiological parameters was higher in pure-bred. The two local breeds expressed little changes in glucose level than crossbred and pure-bred breeds ($p=0.014$). Similarly increasing temperature also has significant effect on glucose concentration with more susceptibility of pure-exotic than other breeds, probably due to non-utilization in the milk synthesis. Both breed and temperature has significant ($p<0.001$) effect on cortisol level. Cortisol level increases significantly ($p<0.001$) with ambient temperature. All the breeds showed almost similar level of cortisol at 18 °C, however as the temperature increases from 18 °C to 32 °C and 42°C, there is remarkable increase in cortisol level of cross and pure-bred cattle. As the ambient temperature increased from 18 °C to 32 °C and 42 °C, the progesterone level started to decrease in all breeds. Significant decrease was observed in progesterone level at 42 °C. Daily milk yield and progesterone level are negatively correlated with ambient temperature. Glucose and cortisol levels were positively correlated with each other and negatively with Progesterone.

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A NOVEL CHROMOSOMAL TRANSLOCATION AND HETEROMORPHISM IN A FEMALE WITH RECURRENT PREGNANCY LOSS—A CASE STUDY

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Background: Recurrent pregnancy loss is defined as three or more consecutive spontaneous miscarriages. Approximately 15–20 % of all pregnancies end up in spontaneous abortions with 50 % of them being associated with chromosomal abnormalities. The present report describes clinical, biochemical and cytogenetic analyses of a couple with reproductive failure.

Material and Methods: A couple with a history of recurrent pregnancy loss was referred to Institute of Genetics for cytogenetic evaluation. Chromosomal analysis of the phenotypically normal couple was done by GTG banding to ascertain chromosomal constitution. Advanced karyotype analysis by spectral karyotyping was also carried out in the couple and parents of the female partner, for confirming the chromosomal abnormality.

Results: Clinical and hormonal profile of the couple was found to be normal. The ultrasound scan of the uterus of the female showed normal uterus and ovaries. Chromosomal analysis of the couple revealed a normal 46, XY karyotype in the male spouse, and a unique balanced reciprocal translocation 46, XX, t(12;13) (q13;q33) + 15pstk+ in the female partner. Cytogenetic analysis was performed for the parents of the female partner to detect the origin. The analysis revealed a similar translocation between chromosomes 12 and 13 in the father and 15pstk+ in the mother. Further, corroboration of the chromosome abnormalities was carried out by spectral karyotyping.

Conclusion: The present study reports a rare, unique and novel chromosomal translocation in a female with history of recurrent pregnancy loss associated with coinheritance of balanced chromosomal translocation from father and heteromorphism from the mother.

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TVS – THE “TOOL “ FOR INFERTILITY

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INTRODUCTION:

The use of transvaginal sonography (TVS) in the management of infertility is gaining increasing popularity. The improved resolution and better tissue textural differentiation afforded by TVS makes this technique useful in monitoring ovarian follicular growth, ovulation, and corpus luteum formation, and in evaluating the normal anatomy of the uterus and the cervix and their cyclic response to ovarian steroids. Adnexal and cul-de-sac abnormalities related to infertility can also be identified. TVS-guided procedures can be accurately and safely performed; transvaginal follicular aspiration, gamete or embryo transfer to the Fallopian tube, and fetal reduction are just a few of the procedures recently introduced to reproductive medicine.

We have evaluated 5000 cases of TVS diagnosis with Laparoscopic and Hysteroscopic Co-relation in last 3 years.

Results:-

TVS diagnosis co-related well with almost 95% of Laparoscopic and Hysteroscopic findings and we conclude that:

1. Only diagnostic Laparoscopy and Hysteroscopy as a first investigation tool is not cost effective.
2. TVS accurately diagnosis 95% of infertility problems.
3. TVS guides the endoscopic surgeon for accurate planning with operative Laparoscopic and Hysteroscopic procedures.
4. TVS is safe, quick accurate, reproducible and cost effective first step investigation in all cases of female infertility.

To practice infertility without TVS is like walking in the dark without a torch.

P168

DENDRITIC CELLS AND INFERTILITY: A STUDY OF SUBPOPULATIONS IN HUMAN ENDOMETRIUM AND BLOOD

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Background:

Infertility affects 10-15% of all couples. Dendritic cells (DCs) are crucial for initiating and regulating both innate and adaptive immune responses and have important roles in immune tolerance during implantation and pregnancy. Dysregulation of DCs has been implicated in fertility complications such as recurrent miscarriage, however detailed knowledge is lacking.

Objective:

To comprehensively characterise DC subpopulations in the blood and endometrium of fertile and infertile women.

Methods:

In this ongoing study, endometrial (n=9) and blood samples (n=10) are prepared as single cell suspensions, labelled to discriminate live and dead populations and stained with a cocktail of fluorescent-conjugated DC antibodies. DCs are analysed by multi-colour flow cytometry.

Results:

Analysis of DCs shows three CD11c+ myeloid (mDC; CD1c+, CD141+, CD16+) and two CD304+ plasmacytoid (pDC; CD2+/-) subpopulations in blood and endometrium with higher proportions of mDC than pDC (mDC range in blood = 19.6-87.4% of lineage-HLADR+ cells and tissue = 17.4-64.5%; pDC range in blood = 3.7-29.4% and tissue = 0.1-53.2%). DC populations show variations during the menstrual cycle. The patterns of cyclical changes in DC populations appear to be subtly different in fertile and infertile women in preliminary results but further investigation is required.

Conclusions:

Circulating and endometrial DCs have important roles in female fertility and may be disturbed in infertile women. Cyclical DC changes indicate possible recruitment from circulation into the uterus and involvement in signalling and functional changes. This study is ongoing.

P169

TUBAL PATENCY HAS SIGNIFICANT IMPACT ON IVF-ET OUTCOMES

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Objective

Tubal function is imperative for pregnancy. On the other hand, IVF have been used for tubal factor without considering their functions. The relation between tubal patency and IVF-ET outcome has not been investigated. Therefore, the present study was conducted to investigate if tubal patency influenced outcomes of IVF-ET.

Materials and Methods

IVF outcomes of 64 patients (131 cycles) who received IVF due to bilateral tubal occlusions between 2009 and 2012 were analyzed. Tubal patency was diagnosed by hysterosalpingography (HSG). The patients with bilateral tubal occlusions were

asked to select falloposcopic tuboplasty (FT) first to recover tubal patency for conventional infertility treatment, then IVF-ET if necessary or to receive IVF-ET directly. Forty six patients (95 cycles) received IVF after FT (Group A) and 18 patients (36 cycles) received IVF directly (Group B). Pregnancy rate (PR) and other parameters were compared.

Results

Overall PR of the patients in Group A (67.4% per patient and 36.8% per cycle) was significantly ($P<0.05$) higher than in Group B (38.9% and 19.4%, respectively). PR of frozen embryo transfer showed significantly ($P<0.03$) higher in Group A (55.8%) than Group B (37.7%). The endometrium was significantly thicker in Group A ($12.0\pm1.8\text{mm}$) than B ($11.2\pm1.4\text{mm}$, $P=0.09$). However, there were no significant difference in development of embryos to blastocyst between Group A and B (46.3% and 38.9%, respectively).

Discussion

Tubal patency may improve implantation via endometrial condition, but does not affect the quality of embryos. Oviduct is not only tubal structure, but also biologically important for implantation.

P170

POTENTIAL DONOR ATTRITION IN AN OOCYTE DONATION PROGRAMME

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Background

Fewer than 10 or of every 1000 potential sperm donors are accepted as a consequence of variable semen quality or unacceptable medical, family, psychological and logistical circumstances. With oocyte donation, provided donor age is <35 years, it is assumed that oocyte quality is not a concern. Oocyte donors however have all of the other issues along with a far more formidable physical and psychological donation process. Clinics invest considerable time and money evaluating, counselling and testing potential oocyte donors.

Aim

To understand the reasons for attrition and enable earlier identification of unsuitable donors, thereby reducing costs.

Method

A telephone questionnaire conducted with past potential oocyte donors who have not proceeded to donation.

Results

21 were rejected by age, 19 being too old and 2 being too young. 13 were rejected on the basis of medical history, genetic reasons or drug use. Of the remaining 33, 13 could not accept ID release (5 donor, 4 both, 4 partner). 20 of the 33 found that the process was more involved than expected, 11 could not afford the time involved, 7 might have proceeded had there been better financial compensation and 2 could not accept the counselling process.

Conclusion

Information provided during initial contact with potential oocyte donors has been enhanced to more comprehensively detail medical requirements, the donation process, reasons behind the mandated identification release requirement, and reimbursable costs to allow earlier exit from the process if they are unsuitable or decline to continue.

P171

G-TCT REVERSES BODY, ORGAN AND FOETAL WEIGHT LOSS IN THE NICOTINE-TREATED PREGNANT MICE

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The decrease in body, reproductive organs or fetuses weights in nicotine-exposed mice before and during pregnancy has been reported. Studies have suggested that supplementation of γ -TCT, an isomer of Vitamin E, to pregnant mice reduced the impact of nicotine on pregnancy outcome. In this study, the effect of γ -TCT on pre-partum foetal and neonatal growth/weight following nicotine exposure was investigated. Ninety six female mice were divided into 12 groups and cohabited with fertile males. Pregnant mice of Groups 1, 5 and 9 were injected (sc) with 0.9% saline. Groups 2, 6 and 10 were injected with 3.0 mg/kg bw/day nicotine. Groups 3, 7 and 11 were gavaged with γ -TCT of 60 mg/kg bw/day. Groups 4, 8 and 12 received concomitant treatment of nicotine concurrently with γ -TCT from Day 1 through Day 7 pc. Body weights of the

pregnant mice were recorded. Groups 1-4 were sacrificed on Day 10 pc. Ovaries, fallopian tubes, placentae and foetuses were weighed and recorded. Groups 5-8 were laparotomized on Day 8 pc., implantation sites were counted and animals were sacrificed on Day 13 pc. Respective organs were also weighed and recorded. Groups 9-12 were laparotomized, implantation sites were counted and pregnancy outcome as well as the survival rate of the new born were evaluated until the neonates aged 14 days old. Nicotine treated animals showed significant body weight loss (~20%) followed by decreased in weights of the organs ($P < 0.05$). γ -TCT supplementation concurrently with nicotine reversed the recorded weight loss back to normal. Nicotine-induced oxidative stress was shown to reduce plasma progesterone levels in pregnancy. γ -TCT supplementation, however, brings it back to normal. Our findings therefore suggest that γ -TCT might have beneficial effect in moderate maternal smokers.

P172

A COMPARATIVE ANALYSIS OF OUTCOMES IN ASIAN AND CAUCASIAN WOMEN FOLLOWING ART.

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Awareness of the disparity in ART outcomes between different ethnic and racial groups may enable improved tailored regimes. Whether the difference is genetic or confounded by other variables is difficult to ascertain as shown by multiple conflicting reports. A large study in one institution aimed to optimise exploring this hypothesis.

The study and control group included patients managed by a specialist in a single IVF centre. Patients in their first IVF/ICSI cycle were retrospectively analysed and excluded if they represented for another cycle within the ten-year study period. 2594 cycles were evaluated (3:1 Caucasian :Asian). Logistic regression analysis was performed.

Results revealed that Asian women are older than their Caucasian counterparts at their first cycle of IVF/ICSI owing to later presentation and longer duration of infertility. Despite higher doses of gonadotrophins they achieved less oocytes; had resultant fewer embryos for transfer or cryopreservation. Notwithstanding, they achieved a lower clinical pregnancy and live birth rate than their Caucasian counterpart, following replacement of additional embryos.

This study highlights the issues of late presentation and prolonged duration of infertility being pivotal in IVF/ICSI outcomes. Educating health practitioners to relay this information to women seeking assistance for infertility may ultimately improve pregnancy rates in this population.

P173

PREVALENCE OF PERSISTENT CHLAMYDIA TRACHOMATIS INFECTION IN SUBFERTILE WOMEN FROM SERBIA

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BACKGROUND: *Chlamydia trachomatis* infection is the most common sexually transmitted bacterial infection in the world. High prevalence rates of genital *C. trachomatis* infection with drastically increased incidence in last years, are imposed as an important public health concern.

PURPOSE: The objective of our study was to determine the prevalence of *C. trachomatis* persistent infection in women with tubal factor infertility (TFI) and spontaneous miscarriage.

METHODS: Serum was collected from 94 patients (38 with TFI and 56 with spontaneous miscarriage) and analyzed for the presence of IgG and IgA antibodies against *C. trachomatis* MOMP antigen (DIA. PRO) and IgG antibodies to chlamydia heat shock protein 60 (cHSP60) antigen (Medac). Patients who are IgG and/or IgA seropositive with simultaneous cHSP60 seropositivity were defined as patients with persistent chlamydial infection.

RESULTS: In our study population, we determined high degree (56.4%) of seropositivity against chlamydial antigens. Persistent chlamydial infection was confirmed in 24/94 (25.5%), while serologic evidence of previous or recent chlamydial infection was confirmed in 29/94 (30.9%) women. Contrary, 41/94 (43.6%) patients were without serological evidence of chlamydial infection. The persistent chlamydial infection was prevalent in women with TFI (36.8%) than in women with spontaneous miscarriage (17.9%), while serological evidence of previous or recent chlamydial infection was predominant in women with spontaneous miscarriage (46.4%) than in women with TFI (7.9%).

CONCLUSION: Our study confirmed serological evidence of persistent chlamydial infection in women with TFI and in women who have experienced a spontaneous miscarriage.

P174

A COMPARATIVE STUDY OF SERUM AND FOLLICULAR FLUID CONCENTRATION OF LEPTIN IN EXPLAINED INFERTILE, UNEXPLAINED INFERTILE AND FERTILE WOMEN

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Background: The relationship between metabolism and reproduction has been always among important topics in female endocrinology. It seems that leptin is one of the involved factors in infertility. Leptin, in addition to regulating body weight has an important role in regulation of endocrine, reproductive and immune functions.

Objective: To compare serum and follicular fluid leptin concentrations among explained infertile women, unexplained infertile women and fertile women.

Method: A case-control study included, a total of 90 women were divided into three equal (n=30) groups of explained infertile, unexplained infertile (case groups) and normal fertile (control group). The three groups were matched in regard to demographic features (age:20-40 and BMI:20-25). In order to determine leptin level, blood sample and follicular fluid were taken respectively one hour prior to and at the time of follicular puncture. Serum and follicular fluid leptin levels were measured with ELISA Data were analyzed using descriptive-analytic tests Mann-Whitney and kruskal Wallis.

Results: In explained infertile and fertile groups, as opposed to unexplained infertile group, mean leptin level was lower in follicular fluid than in serum. Mean follicular fluid leptin concentration in women with unexplained infertility was higher compared to the other two groups. Women with unexplained infertility had lower level of serum leptin in comparison to the other two groups. Leptin level of follicular fluid in all subgroups of causes in explained infertile group was lower as compared to unexplained and fertile women.

Conclusion: The results suggest high leptin level of follicular fluid is one factor in infertility.

P175

UTERINE ULTRASONOGRAPHY ON WOMEN UNDEGOING EMBRYO TRANSFER WITH FROZEN EMBRYOS OR EMBRYOS DERIVED FROM DONOR OOCYTES

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Background: Is there a relation between uterine characteristics on ultrasound and the clinical pregnancy rate of women undergoing embryo transfer with frozen embryos or embryos derived from donor oocytes?

Methodology: A nested case-control study conducted from 1/2013 to 6/2013. All women undergoing embryo transfer with frozen embryos or embryos derived from donor oocytes were recruited and followed until clinical pregnancy.

Result: Investigated 116 cycles, 63 (54.31%) clinical pregnancies, 53 (45.69%) without pregnancy. Endometrium with triple-line pattern made up only 15,9% in cases and 17,0% in controls. Endometrial thickness > 10 mm (before embryo transfer) was recorded in 50,8% of the cases and 49,1% of the controls. No relationship was found among clinical and epidemiology features and clinical pregnancy rate. However, data from this study indicated the potential relation among clinical pregnancy rate and several features such as maternal age, infertility duration, history of embryo transfer, history of cycle cancel, subtraction of endometrial thickness before transfer and at the beginning of the cycle, Estrogen doses before transfer.

Conclusion: No relationship was found between these uterine characteristics on ultrasound and the clinical pregnancy. A wider-sample study with the same objective should be conducted in order to find out the real relation between the above features with the clinical pregnancy rate after embryo transfer. In the near future, studies with further design could find out the best cut off of endometrium for the best pregnancy chance after embryo transfer.

P176

A RANDOMIZED STUDY ON THE OUTCOMES OF FSH PRIMING AND NONPRIMING IN IN VITRO MATURATION OF OOCYTES IN THAI INFERTILE WOMEN WITH PCOS

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Background

FSH priming has been studied in *in vitro* matured oocytes in terms of oocyte maturation rate, embryo quality, and pregnancy rate. However, the results were not encouraging. Delayed FSH priming on day 6, which was supposed to synchronize follicle development in long cycle in PCOS, may be beneficial in oocyte development and proper endometrial preparation.

Objective

The objectives of the study were to compare the oocyte maturation rate and pregnancy rate between the non-FSH priming group (Group 1) and the FSH priming group (Group 2).

Study design

Randomized controlled study

Methods

40 patients were enrolled in this study and randomly allocated into 2 groups. The 3-day FSH priming was commenced on day 6 in Group 2. For both groups, hCG was injected on day 10. Oocytes were obtained on day 12. Estradiol supplementation was provided to patients having thin endometrium after day 10. Two day-3 embryos were transferred.

Results

The oocyte maturation rates within 24 hours were $50.2 \pm 4.07\%$ and $50.77 \pm 4.62\%$ respectively ($p > 0.05$). The pregnancy rates were 50% in Group 1 and 30% in Group 2 ($p > 0.05$). However, the embryo cleavage rate was significantly higher in Group 2 ($p = 0.025$).

Conclusion

FSH priming facilitated the embryo cleavage potential, however, was not beneficial in promoting oocyte maturation rate and fertilization rate. Delayed estradiol supplement before embryo transfer might have a role to synchronize the embryo and endometrial development to improve the pregnancy rate.

P177

CONVERSION OF IVF TO IVM TREATMENT FOR FERTILITY PRESERVATION: TWO CASE STUDIES OF SUCCESSFUL OOCYTE AND EMBRYO STORAGE PRIOR TO CHEMOTHERAPY TO AVOID CYCLE CANCELLATION.

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Aim: Two patients sought fertility preservation treatment before commencing chemo/radio therapy of nasopharyngeal and cervical carcinomas.

Method: After prompt referral both patients ceased the oral contraceptive pill and began ovarian stimulation with 125IU of FSH. After six days of stimulation ultrasound examination demonstrated in excess of 30 follicles $< 11\text{mm}$. To avoid cancellation due to ovarian hyperstimulation, the cycles were converted to IVM. FSH stimulation was ceased and without receiving a HCG trigger, patients were prepared for TVOA two days later. All immature oocytes were cultured for 24 hours in maturation medium before assessment of nuclear maturity. Both patients were advised to freeze half the mature oocytes and culture the remaining to the blastocyst stage after fertilisation due to the high number of oocytes collected and the young age of the couples.

Results: Patient one had 32 immature oocytes collected, 20 matured to the M2 stage of which eight were vitrified. The remaining 12 were inseminated by ICSI. Nine oocytes fertilised and were cultured to the blastocyst stage of which three were vitrified. Patient two had 18 GV stage oocytes collected, 11 matured of which six were vitrified. Five were inseminated and three fertilised, with two blastocysts suitable for freezing.

Conclusion: Were it not for the option of IVM in these circumstances, the patients would likely have had their IVF cycles cancelled or experienced ovarian hyperstimulation syndrome, delaying their cancer treatments. Both patients recovered well and began chemotherapy in under two weeks, with the almost certain reassurance of preserved fertility.

P178

PATTERN OF DELIVERIES DURING THE CALENDAR YEAR IN RURAL INDIA

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ABSTRACT:

Objective: to understand the delivery pattern during the calendar months in rural India.

Material and Methods: The study was carried in a retrospective manner by collecting month-wise delivery data for the past three calendar years from registration center in a rural area of Ambala District of Haryana Province(India). This included deliveries occurring after viable period of 28weeks gestation conducted in (i) a teaching Institute, (ii) government set-up hospital and (iii) domiciliary environment of that particular area.

Observations : During the study period, 43,191 deliveries were recorded with an average monthly rate of 1,199 births. Sinusoidal pattern was observed in the monthly distribution of deliveries peaking during August – October and decline in January and April-May. There was a statistically significant difference between the highest and lowest rates.

Conclusion: Results of this study may be beneficial in health system planning and in the interpretations of seasonal variations in other reproductive parameters.

P179

A LINE THAT SHOULD NOT BE CROSSED: ECTOPIC PREGNANCY AT THE CESAREAN SECTION SCAR

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What would you do if you were in a line that should not be crossed?

Nearly 95 percent of ectopic pregnancies are implanted in the various segments of the fallopian tubes and less than 1% are implanted in the cesarean scar.

Cesarean scar pregnancy is defined as ectopic pregnancy embedded in the myometrium of a previous cesarean scar. It is considered to be the rarest form of ectopic pregnancy and constitutes a life threatening condition.

This is a case of a 38 year old Gravida 2 Para 1 (1-0-0-1), cesarean section, who presented with vaginal spotting, on her 5 weeks and 6 days age of gestation. Serum β -hCG level was 5,640 mIU/ml. Transvaginal sonography revealed cesarean section scar pregnancy. Initial treatment with methotrexate 50mg intramuscularly was given. Another dose of methotrexate was contemplated, however, repeat serum β -hCG showed 18, 429 mIU/ml which is far from the goal of less than 15 % from the baseline. A repeat ultrasound revealed early fetal demise. Hence, ultrasound guided dilatation and curettage was done.

Repeat serum β -hCG one day after the surgery showed decreased level of 5, 414 mIU/ml. She was advised with weekly monitoring of serum β -hCG until level becomes undetectable, repeat ultrasound on the second day of next menstrual cycle and oral contraceptive pills.

Methotrexate in conjunction with dilatation and curettage may avoid unnecessary hysterectomy and offers preservation of fertility in cesarean scar pregnancy patients.

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A GLIMPSE OF THE SILVER LINING: CORNUAL HETEROTOPIC PREGNANCY

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Introduction

A heterotopic pregnancy is the coexistence of an intrauterine pregnancy and an ectopic pregnancy. It is a rare form of twin pregnancy, with an estimated incidence of 1/30,000 in spontaneous pregnancies.

The occurrence of a heterotopic pregnancy with an extrauterine gestation at the cornual area is a rarer variant of twin pregnancy. Its incidence remains unknown and it poses a potentially fatal condition when diagnosed belatedly. Hence, its prompt diagnosis is of utmost importance in order to salvage the intrauterine pregnancy with an aim at preserving fertility; and, to avert potential maternal and fetal morbidity and mortality.

Case Report

A cornual heterotopic pregnancy was diagnosed at 6 weeks of amenorrhea in a 29-year-old woman complaining of persistent, gnawing hypogastric pain. The patient had a previous history of right salpingo-oophorectomy secondary to a dermoid cyst. Exploratory laparotomy was done, revealing presence of a ruptured right cornual pregnancy. Cornual wedge resection was performed. Following laparotomy, the intrauterine pregnancy was supplemented with progesterone and was brought to 37 weeks. Elective cesarean section was done and she delivered to a live healthy baby boy. Post-surgery, the course of both mother and child was uneventful.

Conclusion

The occurrence of a cornual heterotopic pregnancy heralds a very rare and potentially dangerous condition. With prompt diagnosis, its potentially catastrophic sequelae, such as fetal loss and maternal morbidity and mortality, could be averted; and, ultimately, the silver lining amidst this difficult situation could be glimpsed with the birth of a healthy baby with concurrent preservation of fertility.

P181

DETORSION AND CONSERVATIVE MANAGEMENT OF TWISTED ADNEXA: A CASE REPORT OF SUCCESSFUL OVARIAN PRESERVATION

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BACKGROUND

Torsion of the uterine adnexa accounts for approximately 2.7% of all gynecological emergencies and one of the important causes of acute abdominal pain. The traditional recommended treatment for ovarian torsion is removal of the ovary without detorsion. However, conservative therapy for preservation of ovarian function should be undertaken, because most women with adnexal torsion are of reproductive age.

CASE ILLUSTRATION

We report a case of nulliparous 28 years old lady, with acute abdominal pain due to suspected twisted ovarian cyst. A mobile cystic mass diameter 8 cm was palpated on right adnexa with tenderness. On transrectal ultrasound examination revealed a cystic mass on right adnexa size 9x5 cm without internal echo suggesting right simple ovarian cyst. The patient underwent emergency explorative laparotomy with post operative diagnosis was twisted right simple ovarian cyst. Intraoperative finding was bluish, 2 fold-twisted right adnexa, with serous simple cyst diameter 11 cm. After performing detorsion, cystectomy was performed. The patient had Doppler ultrasound evaluation the next four weeks and revealed good follicle development of the affected ovary.

CONCLUSION

Although twisted adnexa may appear to be ischemic or hemorrhagic, it can safely be revived by detorsion with preservation of fertility function. Complete arterial obstruction is not commonly encountered. Detorsion should be the first choice treatment in the management of ovarian torsion, regardless of the color or number of twists of the ovary. Even gangrenous-appearing adnexa should not be removed because it is impossible to predict the chances of the ovary reviving after detorsion.

P182

IN VIVO OPTICAL IMAGING OF IMMATURE MURINE TESTICULAR TISSUE ENGINEERING FOR MALE FERTILITY PRESERVATION

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Objective

To generate new clinical strategies for male fertility preservation in pediatric cancer patients is important.

Design

Transgenic mouse model

Material & Methods

FVB/N-Tg (PolII-luc) Ltc transgenic male mice were used as the donors. Inbred FVB/NJNarl wild-type mice were used as recipients. Before each experiment, the recipient mice underwent unilateral orchiectomy to leave scrotum as the grafted site. In experiment 1, donors from 20-week-old or 3-week-old were transplanted into scrotum, head, abdomen or back muscles of recipients, followed by tracking the testicular tissue development by bioluminescence imaging (BLI) in a longitudinal model to monitor transplanted grafts until 50 days later. In experiment 2, 3-week-old recipients were transplanted age-matched donor testicular tissue to the scrotum of orchiectomy with or without scaffold (Poly-L-lactic acid, PLLA) (Fig 1E)(n=5). BLI was utilized to measure transplanted grafts until 42 day after transplantation. Two-tailed t-test was used for statistical analysis and $P < 0.05$ was considered statistically significant.

Results

Based on the quantity of BLI analysis, the first experiment showed that scrotum is the most optimal site for testicular tissue development, so are the younger donors. The second experiment was extended from the first experiment, scaffold commonly used in tissue engineering can also increase cell regeneration compared with the control group, especially in the 14 days after transplantation ($P < 0.05$).

Conclusions

This pilot study supports the *in vivo* tissue engineering of regeneration capacity of immature testicular tissue and spermatogenesis in the most optimal site of scrotum, younger donors and scaffold application.

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EFFECT OF COLLAGENASE DISAGGREGATION OF OVARIAN TISSUE ON FOLLICLE VIABILITY

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Introduction: Follicles can be a source of oocytes for oncology patients' fertility preservation. Collagenase IV (CIV) has been used to isolate follicles from ovarian tissue, but damages the granulosa cell layers. Collagenase II (CII) disaggregation has not been examined. Follicle viability has been assessed by subjectively scoring the morphometry of DAPI-stained follicles. The application of mitochondrial CMXRos staining, or resazurin metabolism, to assess follicle viability has not been reported.

Aim: to determine follicle yield and viability after ovary disaggregation with collagenase II or IV, and to develop objective, quantitative follicle viability assessment assays using CMXRos staining and resazurin to measure NADPH dehydrogenase activity.

Method: Mouse ovaries were mechanically disaggregated without enzymes, or with 0, 0.5, 1, 2 mg/mL collagenase II or IV for 30 or 60 min before 5min mechanical, enzyme-free disaggregation, to collect follicles.

Results: The yield of follicles after CIV disaggregation was twice that of CII. Both collagenase types yielded predominantly primary follicles (7 follicles/mg tissue) with few primordial, secondary or antral follicles. The amount of CMXRos staining, and the numbers of high grade DAPI-assessed follicles, were not affected by the type of collagenase. The prototype resazurin assay was sensitive enough to detect the metabolic activity of 4 follicles per well after 18h in a simple *in vitro* culture system. CIV decreased the metabolic activity of follicles in the resazurin assay compared to CII.

Conclusion: the reduction in NADPH enzyme activity caused by collagenase IV doesn't compensate for the increased follicle yield. Optimisation of CII disaggregation continues.

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OOCYTES CRYOPRESERVATION IN YOUNG BREAST CANCER PATIENTS USING VITRIFICATION AFTER IN VITRO MATURATION (IVM)

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Oocyte cryopreservation is one option for cryopreservation. Because increase in estradiol (E2) levels during ovarian stimulation to retrieve oocytes in breast cancer patients with hormone receptor positive has potential to deteriorate primary diseases, oocyte retrieval before E2 increase followed by IVM could minimize the risk. Here, we studied the possibility of oocyte cryopreservation after IVM in those patients.

We investigated in 2 patients (33 and 29 years-old) with a total 6 stimulation cycles. In the first patient, we attempted a GnRH antagonist cycle and obtained 7 mature oocytes and vitrified 6 oocytes after IVM (E2: a total of 363 pg/ml for 7 follicles), and a GnRH agonist short cycle: 12 immature oocytes and vitrified 11 oocytes after IVM (E2: a total of 954 pg/ml for 14 follicles). In another patient, she received four GnRH agonist short cycles: 1 immature oocytes and vitrified 1 oocytes after IVM (E2: a total of 236 pg/ml for 6 follicles); 15 immature oocytes and vitrified 8 oocytes after IVM (E2: a total of 1997 pg/ml for 33 follicles); 6 immature oocytes and vitrified 1 oocytes after IVM (E2: a total of 566 pg/ml for 20 follicles); 18 immature oocytes and vitrified 12 oocytes after IVM (E2: a total of 2743 pg/ml for 24 follicles). The retrieval rate was 56.7% with average E2 at 66.0 pg/ml/follicle

We could cryopreserve mature oocytes after IVM with minimum increases in E2 levels, suggesting our approach is suitable for breast cancer patients with hormone receptor positive.

P185

PRESERVATION OF FERTILITY FOLLOWING ABNORMALLY ADHERENT PLACENTA TREATED CONSERVATIVELY

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Abstract

Introduction One of the ensuing complications of placenta accreta includes loss of fertility.

Case presentation An Asian origin Indian national patient with history of placenta accrete at the time of previous delivery and had conservative management with injection methotrexate after the failure of surgical intervention, conceives again and has uneventful antenatal period and parturition.

Conclusion Conservative strategy of leaving the excessively adherent placenta in-situ alongwith adjuvant therapy in the form of injection methotrexate, not only prevents readful complications but also retains fertility in haemodynamically stable patients desirous of future pregnancy.

P186

EFFECTS OF NT3 AND LIF ON THE DEVELOPMENT OF PRIMORDIAL FOLLICLES DURING IN VITRO CULTURE OF NEONATAL RAT OVARY

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Primordial follicles are the key population for fertility preservation. During a female reproductive lifespan, primordial follicles leave the arrested pool and undergo primordial to primary follicle transition. This study aims to explore the interacting effect of NT3 and LIF on the survival and initiation of primordial follicles, and investigate the possible mechanism underlying NT3 contributes to follicular development. Ovaries from 4 day-old rats were cultured for 14 days in M2 medium supplemented with NT3, LIF alone or combined. Non-cultured and M2 group ovaries were set as controls. The developmental stages and viability of follicles were assessed using histomorphology and TUNEL labeling. Immunohistochemistry for PCNA was performed to evaluate the mitotic activity of granulosa cells. Measurement of androstenedione and estradiol in the medium was performed and the expression of KL mRNA in the ovary was examined. After culture, follicles sustained healthy and less apoptotic cells were found in the presence of NT3 or LIF. Furthermore, the proportion of developmental follicles increased and the percentage of GCs that stained for PCNA was significantly higher. However, when combined NT3 with LIF, no obvious additional effect was seen. The treatment of NT3 had the highest expression of KL mRNA, about 3.38±0.43 times higher than M2 group. In summary, NT3, LIF alone or combined had a stimulative effect on the survival and development of primordial follicles. However, there was no obvious synergistic or additional effect between LIF and NT3. One of the mechanisms of NT3 underlying the initiation of primordial follicles might activate c-kit/KL interactions.

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EDUCATING FOR EARLY REFERRAL: THE HOLY GRAIL OF AN ONCOFERTILITY SERVICE

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EDUCATING FOR EARLY REFERRAL: THE HOLY GRAIL OF AN ONCOFERTILITY SERVICE

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Aim:

To initiate and market a viable oncofertility service to young people with newly diagnosed cancer who wish to preserve their fertility.

Method:

An educational and marketing program was initiated in November 2012 to inform oncological health providers of the logistics of early referral required for timely gamete preservation prior to chemotherapy in young cancer patients. This involved presenting to breast cancer and solid tumor groups in Flinders Medical Centre and the wider community.

We assessed the number of referrals for a 12 month period before and after the intervention.

Results :

None of the oncology patients had received chemotherapy at gamete collection. All referrals for oocyte storage came from breast cancer or solid tumor surgical groups.

Oocyte storage referral numbers increased from 1 per annum before the campaign to 10 for the next year.

Responses to stimulation, AMH, peak estradiol, oocyte retrieval numbers and AMH in these women were similar to controls.

Referral rates for semen storage increased from 3 to 21 per annum in the same period.

While 9 of these 24 men had a count or motility less than WHO criteria, all had enough spermatozoa recovered for potential Intracytoplasmic Sperm Injection (ICSI).

Patient acceptance rates were subjectively high

Conclusion:

Education and awareness of early referral remains the Holy Grail of oncofertility.

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SPERM VITRIFICATION WITHOUT CRYOPROTECTANT

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Vitrification is one of the methods for sperm cryopreservation. This procedure needs high concentration of cryoprotectant to avoid ice crystal formation which is harmful for the sperms. There are two kinds of cryoprotectant: (1) intracellular cryoprotectant which will cross the cell membrane so that it can buffer the intracellular salt, (2) extracellular cryoprotectant has an important role in cell dehydration process. Sperm is gamete cells that has intracellular matrix with high viscosity that may function as internal cryoprotectant. Moreover, sperm has a compact structure and small number of cytosol so that it doesn't need extracellular cryoprotectant to assist dehydration process. The aim of this research was to observe the cryosurvival (motility and viability) of the sperm after vitrification without cryoprotectant. Ejaculated sperm from 20 normospermia patients were used as samples and EBSS as buffer medium. Samples were centrifuged to remove seminal plasma. Pellets were mixed with buffer medium then loaded into 0.25 ml straws, equilibrated for 10 minutes before plunged into liquid nitrogen. Samples were warmed after 24 hours of sperm cryopreservation for analysis. The result showed that after vitrification process, 35 % of progressive motility and 48% viability could still be observed. In conclusion sperm vitrification can be applied without cryoprotectant

P189

LOCAL (LIDOCAINE) VERSUS GENERAL ANESTHESIA IN TESTIS BIOPSY TO EVALUATE SPERM PARAMETERS

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Abstract

Objective: to obtain sperm from azospermia men for Assisted Reproductive Technique (ART) need direct biopsy from testis. It is usually done with lidocaine injection as a local anesthesia method in the site of biopsy. The interaction between lidocaine and sperm is controversial. In this study we compare the effects of local anesthesia (lidocaine) with general anesthesia on sperm parameters after biopsy.

Methods: This single-blind randomized clinical trial study was conducted in Fatemezahra Infertility Research center at Babol (the north of Iran) in 2012-2013. 51 azospermia men with age between 20-50 yr considering our inclusion criteria entered the study. The patients randomly divided into 2 groups (25 in group A and 26 in group B). The first group was under testis biopsy with local anesthesia (by lidocaine) and the second group underwent general anesthesia. The extracted sperms were evaluated in parameters in ART laboratory at 0, 1, 2 hr after biopsy.

Results: Group B showed an increase in motility rate and normal morphology at 1, 2 hr in comparison with Group A but this difference was not significant. Sperm parameters had no reduction at 1 and 2 hr after biopsy in the group with local anesthesia. No significant difference was seen in sperm parameters at 0 hr after biopsy in both groups.

Conclusion: Local anesthesia by lidocaine for testis biopsy has no adverse effect on sperm parameters.

KEYWORDS: Azoospermia, Testis biopsy, General anesthesia, Local anesthesia

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STUDIES ON MAMMALIAN MATURATION ANTIGEN(SMA2) ANTIBODY AND THEIR ROLE IN SPERM FUNCTION

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To understand the involvement of the antigens in the event of fertility as well as the cause of the infertility of male and female, the characterization of the sperm antigens and their antibodies that can be used in blocking these events are essential. The major goat sperm maturation antigen (SMA2) is heavily glycosylated, contains hexosamine, mannose, galactose and glucose. In the present study, effect of deglycosylation of SMA2 antigen on the serological reaction and acrosome reaction was investigated. SMA2 glycoantigen showed positive immunoreactivity after treatment with sodium borohydride (NaBH₄) and this generated a 44 kDa protein band. Trifluoromethanesulfonic acid (TFMS) caused aggregation and restricted free mobility of the treated antigen on SDS-PAGE and the protein band generated by TFMS treatment also showed positive immunoreactivity. The results supported the views that the protein portion retains its immunoreactivity even after oxidation of the vicinal hydroxyl group of saccharide component of SMA2 antigen. These data suggest that immunodominant epitopes exist on the core protein and by which the SMA2 antigen retains its immunoreactivity even after disruption of the saccharide portion and the protein epitopes have a role in capacitation and acrosome reaction in presence of antibody which is raised against this protein portion of SMA2 using the negative staining of FITC-PSA (fluorescein isothiocyanate-labeled *Pisum sativum* agglutinin) probe. Altogether, the protein portion might fulfill the serological activity of the antigen as well as the protein epitopes affect the acrosome reaction. In view of this property we propose that the protein portion of SMA2 antigen might be considered as good immunogen.

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SEMEN ANTIOXIDANT STUDY IN MALE PATIENTS AT EAST COAST OF MALAYSIA

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Reactive oxygen species (ROS) is believed to be one of the factors that cause male infertility. Antioxidants in the seminal plasma play an important role in protecting sperm from ROS. The purpose of this study is to determine the antioxidants level in semen sample in male patients without antioxidant treatment at IIUM Fertility Centre, Malaysia. A total of 43 semen samples were taken and seminal fluid analysis (SFA) test was performed according to WHO 1999 guideline. Patients were

divided into two groups; normozoospermia and oligozoospermia. Semen samples were then stored in liquid nitrogen at 196 °C using standard preservation technique. The samples were thawed and centrifuged to separate the seminal plasma and the sperm. The antioxidant levels of Vitamin C and Vitamin E in seminal plasma were measured using the spectrophotometer. Descriptive statistic was done for SFA to summarize the result of sperm parameters and antioxidant levels. It is shown in this study that the level of antioxidants, both Vitamin E and Vitamin C, are higher in the normozoospermia samples compared to the oligozoospermia samples ($p < 0.05$). There is a positive correlation between the sperm concentration and the level of antioxidants.

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THE ASSOCIATION BETWEEN BODY MASS INDEX (BMI) AND SPERM QUALITY IN MALE PATIENTS AT IIUM FERTILITY CENTRE

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Obesity is known as an adverse health cause including male infertility. The increasing of body mass index (BMI) is associated with reduced sperm quality in a few studies however some of findings are oppose to this result. The aim of this study was to determine the relationship of BMI and semen quality. A total of 372 male patients were recruited at International Islamic University Malaysia Fertility Centre (IIUM FC) for semen fluid analysis (SFA) test. Sperm parameters such as sperm count, motility and semen volume were evaluated based on World Health Organization (WHO) guidelines, 1999. Height and weight were measured to determine the BMI. The BMI was categorized into four groups; underweight, normal, overweight and obese. Statistical analysis was done and our result shown that overweight and obesity have reduced the sperm count compared to sperm motility and semen volume ($p < 0.05$). In conclusion, there was an association between BMI and semen quality.

P193

ULTRASTRUCTURE OF THE APICAL POLE OF THE GLANDULAR EPITHELIUM OF RAT PROSTATE INDUCED BY TOLUENE

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Toluene is a clear, refractive liquid that has an aromatic odor similar to benzene. Applications of toluene include use as an indirect food additive, gasoline additive, extraction solvent for plant materials, and as a solvent for adhesives, rubbers, resins, paints etc.

The aim of the present study is to investigate apical pole region of the ventral lobe of rat prostate after expositions to toluene.

Adult male albino rats were used for the experiments. The study was approved by Institutional Animal Ethics Committee. The rats were divided into two groups: control and toluene exposed (each presents 18 animals). We used toluene in concentration at the level 500 mg/m³ during 60 days. After that animals were anesthetized and then killed. After standard preparation technique the prostates were embedded in epoxy-resin. Ultrathin sections were with uranyl acetate and Reynold's solution and examined with transmission electron microscope.

We found well-defined secretory elements consist of membrane-bound secretory granules that contain condensed material in the control animals. The membranes of some of the secretory granules fuse with the luminal plasma membrane before their extrusion into the lumen as part of secretion. The luminal border of the plasma membrane shows micrivilli. Other structures in the apical pole are the coated vesicles, which appear free within membrane-bound vacuoles in the form of multivesicular bodies.

In the treated animals, structural changes in cytoplasmic organelles were observed. The nuclei were indented. The organelles were atrophied and there is commencing of vacuolization. The endoplasmic reticulum was completely disturbed and dilated.

P194

ULTRASTRUCTURE OF THE INTERSTITIAL TISSUE OF THE RAT VENTRAL PROSTATE INDUCED BY TOLUENE

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Toluene is widespread in the environment owing to its use in a wide variety of commercial and household product.

The aim of the present study is to investigate the interstitial tissue of mature rat prostate after expositions to toluene.

Adult male albino rats were used for the experiments. The study was approved by Institutional Animal Ethics Committee. The rats were divided into two groups: control and toluene exposed (each presents 18 animals). We used toluene in concentration at the level 500 mg/m³ during 60 days. After that animals were anesthetized and then killed by decapitation. After standard preparation technique the prostates were embedded in epoxy-resin. Ultrathin sections were with uranyl acetate and Reynold's solution and examined with transmission electron microscope.

In the treated animals we found that smooth muscle and fibroblasts are the most numerous cells of the interstitial tissue of prostate. They are accompanied by macrophages and undifferentiated cells with low electron density. Smooth muscle cells have protrusions of cytoplasm that extend into corresponding depressions in adjacent muscle cells. Fibroblasts and smooth muscle cells are arranged in parallel between adjacent epithelial alveoli and form a sheath around alveolus. This sheath is composed of a layer of fibroblasts and smooth muscle cells.

We conclude that the influence of toluene in concentration at 500 mg/m³ does not lead to significant changes in the fine structure of the interstitial tissue of the rat ventral prostate.

P195

OUTCOME OF FRESH OR FROZEN TESTICULAR SPERMATOZOA, AND CRYO-THAWED EMBRYO TRANSFER IN COUPLES UNDERGOING ICSI-TESE TREATMENT.

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Objective

We performed this study to evaluate use of fresh and frozen testicular spermatozoa and cryo-thawed embryo transfer in azoospermia testicular sperm extraction (TESE) treatment.

Design

A cohort study was undertaken at the Dongtan Cheil Women's Clinic IVF Center in Gyeonggi-do, South Korea.

Materials and Methods

Data from Dongtan Cheil Women's Clinic that reported from January 2008 to July 2013. The timing of TESE was scheduled according to the work load of the center. Sperm retrieval was performed on the day of oocyte pick-up. A total of 35 cycles were included in the study. Group A (fresh testicular spermatozoa of ICSI-TESE) consisted of 24 cycles, Group B(frozen-thawed testicular spermatozoa of ICSI-TESE) consisted of 5 cycles, Group C (frozen-thawed embryo transfer of ICSI-TESE) consisted of 11 cycles.

Results

Groups were comparable in terms of male and female patient ages and ovarian response to stimulation as well as the number of oocytes injected. Details of the three groups were summarized in the following table1.

Table 1 Outcome of ICSI-TESE cycles in the three different groups.

Outcome	Group A	Group B	Group C
Cycle	24	5	11
Biochemical PR (hCG) per ET (%)	50	20	36.4
G-sac PR (%)	37.5	0	18.2
FHB PR (%)	37.5	0	18.2
Miscarriages (%)	16.7	100	50

Conclusions

In this study, A group has higher pregnancy rate than B and C group. But The results need to be independently confirmed in a larger case.

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THE RELATIONSHIP OF HALOSPERM TEST AND HYALURONAN BINDING ASSAY (HBA) IN SEMEN SAMPLE GROUPS CLASSIFIED WITH SPERM PARAMETERS SUCH AS COUNT, MOTILITY AND MORPHOLOGY.

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Objective: To investigate the relationship between hyaluronan binding(HA) ability and DNA integrity in semen samples arranged into groups by semen parameters.

Design: Retrospective study

Materials & Methods: We analyzed the semen from 101 patients. The DNA fragmentation of sperm was assessed by sperm chromatin dispersion (SCD) test using Halosperm kit, and hyaluronan ability of sperm was analysed by sperm-hyaluronan binding assay.

Results:

The mean male age was 38.4±5.8 years for semen analysis in this study. The DNA fragmentation rate of sperm was significantly increased when the semen samples showed subnormal-count($\leq 30.0 \times 10^6/\text{ml}$), the progressive motility(<32.0%) or SM(<4%). Also, there was tend to positive relationship between HA-binding and not sperm count($r=-0.01$) but motility($r=0.35$). Also, the HB-binding rate could be positive relative to the sperm DNA fragmentation of rate($r=0.24$). However, there was no significant difference because it might be small number of test subjects.

	No.	DNA FR (%)	Correlation analysis	No.	HBA assay	Correlation analysis
sperm count($\times 10^6/\text{ml}$)			$r=-0.40$			$r=-0.01$
≥ 30.0	70	26.6	$r=-0.34$	70	84.7	$r=0.19$
<30.0	31	36.6	$r=-0.05$	31	77.4	$r=-0.42$
Sperm motility			$r=-0.57$			$r=0.35$
$\geq 32.0\%$	42	21.1	$r=-0.41$	42	84.8	$r=0.76$
<32.0%	59	35.8	$r=-0.41$	59	78.1	$r=0.31$
Strict Morphology			$r=-0.34$			
$\geq 4\%$	10	30.9	$r=-0.39$			
<4%	62	27.5	$r=-0.04$			
HB binding	No.	DNA FR (%)				
$\geq 65\%$	24	36.7				
<65%	5	44.1				

Conclusion

The combined analysis of semen parameter, sperm DNA integrity and HA-binding ability would be help that we can more exactly solve male infertility. Also, these complex methods could give us some information for the best sperm selection in IVF.

P197

FUNCTIONAL CHARACTERIZATION THE TESTIS SPECIFIC LYSOZYME LIKE PROTEINS

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Lysozymes can damage bacterial cell walls by cleaving the glycosidic linkages in the peptidoglycan layer. They serve in antibacterial defenses thereby forming a part of the innate immune system. Lysozyme-like proteins (LYZLs) that are classified as c-type lysozymes are not well characterized in many species including the rat. Hence the present study focuses on characterization of lysozyme like proteins (LYZL 1-6) in the rat. *In silico* analysis revealed higher similarity among LYZL proteins and they are highly conserved. All the LYZL proteins showed the presence of conserved 4 disulphide bridges similar to lysozyme. Only LYZL1 and LYZL6 contained the active site residues and the three dimensional structures of LYZL proteins are very similar to that of lysozyme with helix loop helix domain. *In vivo* analyses revealed the presence of *Lyzl* gene in male reproductive tract specifically in the testis and the transcripts are developmentally regulated. Immunohistochemical staining correlated with their mRNA expression and majority of them are also localized on the sperm. These proteins were found to have no role in sperm capacitation and acrosome reaction. However, they possess potent antibacterial, muramidase, isopeptidase and free radical scavenging activities. Results of this study suggest that though LYZL proteins are specifically expressed in the testis, they may not have a role in sperm function, but could contribute to immune and stress responses.

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EXPRESSION OF GALECTIN-3 AS A TESTIS INFLAMMATORY MARKER IN VASECTOMISED MICE

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Abstract:

Vasectomy, though in some cases are being confronted with irreversibility, has been accepted as an effective contraceptive method. It is estimated that near 2-6% of vasectomised men ultimately show a tendency to restore their fertility. In some cases, vasectomy has been considered as an irreversible procedure due to many post-vasectomy complications causing this debate. The aim of present study was to investigate the pattern of expression of galectin-3, an inflammatory factor secreted by macrophages and immune cells, following the vasectomy in mice testis tissue. In this experimental study, twenty mature male Balb/c mice, aged two months, were divided into two equal groups: sham and vasectomised groups (n=10). They were sacrificed four months after vasectomy, while the pattern of galectin-3 expression was investigated using a standard immunohistochemistry technique on testicular tissues. Stereological analyses of testes parameters in vasectomised and sham-operated groups were compared by mixed model analysis. Based on observations, although galectin-3 was not expressed in sham-operated group, it was expressed in 40% of testicular tissues of vasectomised mice, like: seminiferous tubules, interstitial tissues and tunica albugina. Also, our result showed a significant alteration in number of germ and sertoli cells of testicular tissue in vasectomised group in comparison to sham-operated group. The expression of galectin-3 at different parts of testicular tissue in vasectomised group is higher than sham group. This express illustrates the increase of degenerative changes and inflammation reactions in testicular tissue, leading to chronic complications and infertility, after the vasovasostomy.

Keywords: Vasectomy, Galectin-3, Inflammation, Testis, Immunohistochemistry

P199

BORTEZOMIB DECREASES MALE FERTILITY VIA UQCRC2

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Bortezomib is a small molecule proteasome inhibitor that affects several other cellular pathways, suppress protein p53 and cell cycle regulators. To date, however there are no known effects of bortezomib on reproduction. Therefore, present study was designed to investigate the effect of bortezomib on male fertility. In this *in vitro* trial with mice spermatozoa, we utilized CASA, CTC staining, ATP assay, western blotting and IVF to measure the main study outcome. The short-term exposure of spermatozoa in bortezomib a decreases sperm motion kinematics, intracellular ATP production, capacitation, [Ca²⁺]_i, the acrosome reaction, ubiquinol-cytochrome-c reductase complex core protein 2 (UQCRC2), and tyrosine phosphorylation (TYP) of sperm proteins in a dose-dependent manner. Notably, the decreased UQCRC2 and TYP were associated with reduced sperm kinematics, ATP production, and capacitation, which ultimately led to adverse effects on male fertility such

as poor fertilization rates and embryo development. Thus, bortezomib may be considered as a potential male contraceptive agent due to its ability to decrease fertility secondary to changes in overall sperm physiology and embryonic development. However, the results of this preliminary study have to be confirmed by additional independent trial.

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GENISTEIN EFFECT ON REPRODUCTIVE SYSTEM IN MALE RATS.

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Background:

Genistein is a soya phytoestrogen that has estrogenic effects. Phytoestrogen has been introduced as one of the causes of infertility in animals. Previous studies on genistein role in male reproductive system have shown contradictory results. Since no similar study has taken place in Iran, the present study has been designed for evaluation of genistein effects on male reproductive system.

Methods:

In this experimental study, 30 male rats with 13 weeks of age and limited weight of 220 to 250 grams were selected. They were divided into six groups with different genistein doses (0, 0.5, 1, 2, 4 mg/kg) were injected subcutaneously on 14 consecutive days to male rats. After 24 hours animals were sacrificed and then their blood testosterone, FSH (Follicle-Stimulating hormone), and LH (Luteinizing Hormone) were measured via ELISA method. Sperm count and viability were measured through WHO protocols.

Findings:

There was a significant reduction in FSH plasma levels among groups that were injected low doses of genistein while by increasing the genistein dose the inhibitory effect of reducing became slower ($p < 0.05$). there were not any significant differences between other indicators.

Conclusion:

Based on our findings, genistein has an effect on FSH level of plasma and the functioning of male reproductive system.

Keywords:

genestein, phytoestrogen, testosterone, sperm count, rat.

P201

TOWARDS IDENTIFICATION OF MOLECULAR MARKERS OF MALE FACTOR INFERTILITY

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Human fertility is declining in many parts of the world, and incidence of infertility is somewhere near 15-20% in many populations. Available evidences indicate that 35% of infertility is related to male factor problems such as structural abnormalities, sperm production disorders, ejaculatory disturbances and immunologic disorders. Though spermatogenic disorders are a very common, the assessment of the same is derived largely from very routine semen analysis which relies highly on the sperm density and morphology. Recognizing the lack of available data to evaluate spermatogenic insufficiency at molecular level, we investigated the structural and functional make up of human spermatozoa by evaluating their membrane architecture and proteome profile. We observed significant differences in the bulk fluidity, lipid ordering and lateral diffusion properties of sperm membranes of infertile human males, when compared with those from fertile controls. Differential display proteomics identified the absence of some proteins in spermatozoa associated with male factor infertility. Our studies indicate that the absence of any one protein from a set of 20 critical proteins is indicative of male factor subfertility. These parameters appear to have strong relevance in prognosis, diagnosis, management and prediction of the outcome of ART in cases involving male factor subfertility.

P202

STAINED QUALITY AND SPERM MORPHOLOGY ASSESSMENT IN FIXING AND NON-FIXING PAPANICOLAOU SMEAR METHOD

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Objective: To evaluate the slides stained quality of fixing and non-fixing Papanicolaou smear and difference in sperm morphology detection.

Material and Method: This was an experimental study. Semen samples were collected from male infertile patients who attended our clinic during April 1, 2008 through March 31, 2009. All of the semen samples were analysed with standard method, then each sample was stained in two glass slides and divided into fixing (95% alcohol) and non-fixing groups. Conventional Papanicolaou staining technique was used. Dye-stained quality and sperm morphology evaluation of each slide were collected and analyzed.

Results: The total 75 men who had official semen report. The mean patients' age was 35.6±5.8 years. Eighty-five percent were referred for primary infertility. The totals of 150 sperm prepared-slides were evaluated. The slide quality of non-fixing Pap smear was not different from fixing method in terms of air-drying artifact, cell loss and cell border, p value = 0.321, 0.159 and 0.083 respectively. Acrosome staining was inferiorly in non-fixing group as well as abnormal sperm morphology evaluation p value < 0.001.

Conclusion: The dye stained quality of non-fixing Pap smear was not different from fixing method. Fixing Pap smear for abnormal sperm morphology detection was recommended.

Key words: fixing, sperm morphology, Papanicolaou smear, slide quality

P203

STATUS OF LEYDIG CELLS IN INFERTILITY

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Objective: to analyze the morphofunctional state of Leydig cells in infertility. **Materials and Methods:** we undertook a full clinical study of 30 married couples, who addressed the doctors due to the infertility. The male patients had a clinical diagnosis of secretory infertility, and they all had testicular biopsy. The age of patients varied from 18 to 40 years. The morphological study included light and electron microscopy. The state of hormonal testicular function was judged by the content of testosterone in the blood. **Results:** we established a sequence of Leydig cell damage and it included the quantitative characteristics and structural changes:

- with maintained processes of spermatogenesis, the index of Leydig cells was 4.8;
- with inhibited process of spermatogenesis at the level of spermatids - 14.4;
- with inhibited process of spermatogenesis at the level of spermatocytes - 10;
- with inhibited level of spermatogenesis at the level of spermatogones - 2;
- in the absence of processes of spermatogenesis, the index of Leydig cells was 0.

Patients with infertility experience lack of androgen function at the stage of the retention of spermatogenesis, which corresponds to the decrease of testosterone in the blood (21.6 ± 1.2 nmol/l). In cases of retention of spermatogenesis, we observed potentially and functionally active Leydig cells in the interstitium, while in cases of inhibition, the Leydig cells disappeared.

Dominance of involutive forms of Leydig cells becomes a reason of hormonal testis dysfunction, so patients require transplantation of donor Leydig cells.

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MALE INFERTILITY WITH RETROGRADE EJACULATION AFTER THE SURGICAL TREATMENT OF ACHALASIA: A CASE REPORT

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We report a rare cause of retrograde ejaculation on an infertility patient after suffering from the open surgery for treatment of achalasia 12 years before. He started recognizing dry ejaculation one month after that surgery. Although we succeeded

with the result of one ongoing pregnancy through an ICSI cycle by using the motile sperms extracted from diluted and alkalized post ejaculatory urine after failure of restoring his antegrade ejaculation by ejaculation on a full bladder and treatment with imipramine but the main result here is the patients with surgical treatment of achalasia could receive counseling about infertility complication before undertaking that surgery.

P205

THE ASSOCIATION BETWEEN SERUM ESTRADIOL LEVEL AND INTIMA-MEDIA THICKNESS IN POSTMENOPAUSAL WOMEN: A PRELIMINARY REPORT

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Objectives: To estimate whether intima-media thickness (IMT) of carotid artery is associated with serum estradiol (E2) level in postmenopausal women.

Methods: We performed a doppler ultrasonography of the carotid artery and measured intima-media thickness (IMT) in 124 postmenopausal women who did not take postmenopausal hormone therapy. Women were divided into two groups according to the IMT (< 1.0 mm and ≥ 1.0 mm). Serum estradiol (E2) level, lipid profile, bone mineral densities (BMD) of the lumbar vertebrae and femoral neck, current statin treatment status, and other coronary risk factors were analyzed in these two groups.

Results: Compared to women with higher IMT (≥ 1mm), women with lower IMT (< 1mm) had a significantly higher level of serum E2 (22.92 ± 7.07 vs. 8.47 ± 1.25 pg/ml, $P = 0.04$), lower tendency of BMI, mean age, time to since menopause (23.48 ± 0.33 vs. 24.14 ± 0.44 kg/cm², 56.19 ± 0.76 vs. 58.00 ± 0.84 years, 6.59 ± 0.77 vs. 8.34 ± 0.88 years) and were significantly less likely to have dyslipidemia (15.3 % vs. 32.3 %, $P = 0.03$). Women with IMT < 1mm were more likely to have a serum E2 level greater than 20 pg/ml (11.9 % vs. 4.6 %). However, multiple logistic regression analysis showed that any factors did not have a significant association with IMT except dyslipidemia.

Conclusion: Serum E2 level might be associated with IMT of carotid artery in postmenopausal women, but larger and prospective study would be needed.

P206

THE OCCURRENCE OF GLAUCOMA AND ASSOCIATION WITH SERUM ESTRADIOL LEVEL IN POSTMENOPAUSAL WOMEN

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Objectives: To investigate the occurrence of glaucoma and association with serum estradiol(E₂) level in postmenopausal women.

Methods: We evaluated serum E₂ level, female reproductive factors and glaucoma related risk factors including intraocular pressure and optical coherence tomography (OCT) findings in 30 postmenopausal women who visited Obstetrics and Gynecology outpatient clinic. Patients who showed abnormal findings in glaucoma screening test were classified to glaucoma suspect group, and underwent glaucoma confirmatory test. Serum E₂ level, female reproductive and other menopausal health-related factors such as lipid profiles and bone mineral densities were analyzed in the glaucoma suspect group and non glaucomatous group.

Results: Eight out of thirty participants (26.7%) were classified to glaucoma suspect group. One of them was diagnosed to have treatment requiring glaucoma, and the other two were found to have early glaucomatous change. Compared to the glaucoma suspect group, non glaucomatous group had a higher level of serum E₂ (19.40 ± 4.79 vs. 13.95 ± 4.55 pg/ml) The difference, however, was not statistical significant ($P=0.525$). Proportion of glaucoma suspect patients in the groups with a higher serum E₂ level (≥ 20 pg/mL) and a lower serum E₂ level (<20 pg/mL) was similar (25.0 and 27.3%, $P=0.645$). Multiple logistic regression analysis showed that no female reproductive factors were associated with the risk of glaucoma.

Conclusion: Comprehensive glaucoma screening using an OCT in postmenopausal women could detect more glaucoma patients than prevalence in the similar age group. Statistical significance was not found in the association between serum E₂ level and the risk of glaucoma.

P207

THE PRESERVATION OF FERTILITY IN PATIENTS WITH MALIGNANT DISEASE; A CASE REPORT

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Introduction: Recently the number of young cancer survivor is increasing due to improvement of cancer treatment. After conquering their life-threatening disease, many patients have to face their impaired fertility in future. We successfully preserved oocytes of a woman with malignant disease before her intensive cancer treatment.

Case report: A 34-year-old unmarried woman with a diagnosis of myelodysplastic syndrome visited our fertility clinic. She was scheduled to have chemotherapy and radiotherapy as a pretreatment of bone-marrow transplantation few weeks later. She was quickly able to make her mind to accept oocytes freezing as fertility preservation after careful consideration of provided information from cancer experts and gynecologists. The oocytes retrieval was performed without complications one day after platelet transfusion. Six mature oocytes were successfully preserved.

Discussion: This is the first cryopreservation of oocytes for women with malignant disease at our fertility clinic. Since then we have been trying to come up with more patients-friendly system in cooperation with cancer specialists. The cancer patients have to decide if they would have ART related treatment in a limited time while they are struggling against their serious illness. So we compiled a brochure on fertility preservation to help patients and medical personnel in other specialty understand easily. We believe close cooperation between cancer experts and ART experts is necessary to establish fertility preservation procedure. More than 20 women with malignancy successfully went through oocyte retrieval procedure and secured their oocytes at our clinic.

P208

STUDY ON DISABILITIES CAUSED BY INADEQUATE MATERNAL AND CHILD HEALTH AND THE TRANSITION OF MATERNAL AND CHILD HEALTH PATTERN IN CHINA

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Objective To compare the changes of prevalence rates, types and class distribution of disability caused by inadequate maternal and child health among 0-6 years old population from 1987 to 2006 of China and describe the transition of maternal and child health pattern in China based on the change of mortality and disability. **Methods** Quantitative analysis had been carried out based on the data from Two National Sample Surveys on Disability in 1987 and 2006 with the SPSS16.0 statistical software. **Results** The prevalence rate of disability caused by inadequate MCH in 2006 was 7.86‰, higher than that in 1987 (7.16‰). The prevalence rate of visual disability, hearing-speech disability and physical disability caused by inadequate MCH in 2006 were all higher than that in 1987, while lower than 1987 on intellectual disability. Compared to 1987, among the physical disability and intellectual disability caused by inadequate MCH in 2006, the percentages of class 1 and 2 rose, class 3 and 4 dropped, while the other two types disabilities were approximately contrary to them. **Conclusion** The disabilities caused by inadequate maternal and child health care became more and more serious, and the disability types and grades have both changed during the 20 years in China. The adverse outcome of inadequate maternal and child health was changing from death to living with disability, which indicated that China should take effective measures to improve maternal and child health care and pay more attention to associated disabilities.

P209

PREDICTORS OF ADHERENCE TO RELAXATION GUIDED IMAGERY DURING PREGNANCY IN WOMEN WITH PRETERM LABOR

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Objectives: The objective of this study was to examine the adherence to relaxation guided imagery in women experiencing preterm labor, as well as predictors that influence this adherence.

Design: This study employed a longitudinal follow-up approach. The 57 participating women each received a mini MP3 player containing a 13-minute relaxation guided imagery audio program, which they were instructed to follow daily until they gave birth. Follow-up interviews were conducted weekly. Generalized Estimation Equation was employed to predict the adherence.

Results: The total adherence rate was 58%. Higher adherence was predicted by women with at least college degree ($p=0.006$), greater perceived stress ($p=0.006$), higher risk for preterm delivery ($p<0.001$), and greater effects of

relaxation($p=0.028$), while higher maternal age was associated with lower adherence($p=0.001$). In addition, adherence was significantly decreased over time ($p<0.001$). Adherence was not related to marital status, employment, parous, the baseline of anxiety, and hospitalization.

Conclusions: Pregnant women with high risk for preterm birth and greater perceived stress had higher adherence to the relaxation guided imagery. A tailored relaxation for those women had lower for relaxation guided imagery and consideration of their personal preferences is necessary.

P210

AN ANALYSIS OF BACKGROUND AND PERCEPTION OF PATIENTS DISPOSING CRYOPRESERVED EMBRYOS

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Objective: Disposition of the frozen embryo is often marked by ambivalence, discomfort, and uncertainty and may be influenced by couples' conceptualization of their embryos. We analyzed background and perception of patients disposing cryopreserved embryos.

Design: Retrospective analysis at a single institution

Materials and Methods: 56 cases voluntarily terminating embryo cryopreservation were reviewed. The request was confirmed by telephone interview by a fertility nurse-specialist. Further counseling in-person was provided in selected cases.

Results: The female demographics were: age at cryopreservation: 34.8 \pm 3.2 year-old, duration of infertility: 41.5 \pm 30.8 months, number of oocyte retrieval: 1.5 \pm 0.9 times. age at requesting disposition: 37.7 \pm 3.4 year-old, duration of cryopreservation: 30.8 \pm 14.7 months, number of disposing embryos: 2.7 \pm 2.3, number of birth: 1.4 \pm 0.8 times. The principal reasons of decision making were: one child achieved (39.3%), multiple children achieved (44.6%), termination of therapy (7.1%), age consideration (7.1%). 14 cases (25%) requested counseling before final decision (average 2.8 times by phone, and 1.3 times in-person). Fear of losing chance of pregnancy, lack of time due to busy child-care, difficulty to reach a consensus with a partner were the major reasons of postponing decision-making.

Conclusions: Approximately 85% of cases cited achieving child birth as the reason of terminating embryo cryopreservation. However, average 2.5 year was needed until disposition and 25% requested counseling before final decision. Understanding of diversity of the reasons, long-term nature, and complexity and uncertainty of decision-making is essential. Ability to assess and advice on issues of child care and barriers of in-couple communication may aid and facilitate the disposition process.

P211

ASSOCIATION STUDY OF ANTI-MÜLLERIAN HORMONE AND ANTI-MÜLLERIAN HORMONE TYPE II RECEPTOR POLYMORPHISMS WITH IDIOPATHIC PRIMARY OVARIAN INSUFFICIENCY

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BACKGROUND: AMH plays an important role in regulating both the primordial follicle recruitment and the cyclic selection of the antral follicles. Therefore, genetic variants in the AMH signal transduction pathway might affect the ovarian function of women. This study was performed to investigate whether the genetic polymorphisms of the anti-Müllerian hormone (AMH) and AMH type II receptor (AMHR2) genes are associated with idiopathic primary ovarian insufficiency (POI).

METHODS: The subjects consisted of 211 idiopathic POI patients and 233 post-menopausal controls. Genotyping for the AMH Ile⁴⁹Ser and the AMHR2 -482 A>G polymorphisms were performed by MGB primer/probe Taqman assay.

RESULTS: The median age (interquartile range) of onset of POI was 30.7 (23.5-37.8) years and the median values (interquartile range) of LH, FSH and estradiol in the POI group were 29.8 (18.7-40.0) mIU/ml, 67.9 (47.1-90.2) mIU/ml and 20.0 (11.2-31.0) pg/ml, respectively. The genotype distributions and allele frequencies for the AMH Ile⁴⁹Ser and the AMHR2 -482A>G polymorphisms were similar between the POI patients and the controls. Within POI population, the AMH Ile⁴⁹Ser and the AMHR2 -482A>G polymorphisms were not associated with age at the time of POI and LH, FSH, as well as estradiol levels. Haplotype analysis also showed no significant difference between groups.

CONCLUSIONS: Our findings suggest that genetic variants in the AMH signal transduction pathway may not influence the susceptibility of idiopathic POI. This is the first report on the association between the AMH and AMHR2 polymorphisms and idiopathic POI.

Key Words: AMH / AMHR2 / polymorphism / primary ovarian insufficiency

P212

PTEN, AKT AND FOXO3A EXPRESSION IN PRIMORDIAL FOLLICLES OF IN VITRO ACTIVATED NEONATAL MOUSE OVARIES

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The majority of primordial follicles are maintained in a quiescent state, only a few of them are activated for development. PTEN, Akt and Foxo3a are essential for maintaining the dormancy and activation of primordial follicles. Using PTEN inhibitor and PI3K activator in both mouse and human ovaries in vitro caused the suppression of Foxo3a and initiated the activation of primordial follicles. To study the effect of the use of the inhibitor and activator, we treated the neonatal mouse ovaries with PTEN inhibitor bpV(HOPic) and PI3K activator 740 Y-P for 0, 1, 6, 12 and 24h, and assessed the expression of PTEN, total Akt, phosphorylated Akt and phosphorylated Foxo3a in primordial follicles by immunohistochemistry and Western blot. The time course analysis showed that the staining intensity of PTEN became weak in oocytes of primordial follicles after 24 h of in vitro treatment, whereas the expression of Akt and phosphor-Akt in primordial follicles peaked at 1h and 6 h after incubation respectively. Moreover, treat the neonatal mouse ovaries for 1h, the nuclear export of phosphor-Foxo3a increased in oocytes of primordial follicles. The different staining patterns of PTEN, Akt, pAkt and pFoxo3a suggested that short-term treatment of neonatal mouse ovaries with PTEN inhibitor and PI3K activator repressed the expression of PTEN and promoted Akt activation and Foxo3 hyperphosphorylation, leading to activation of primordial follicles. In conclusion, primordial follicles could be activated in vitro via the PTEN-PI3K-Akt-Foxo3a signaling pathway. PTEN inhibitor and PI3K activator may have significant clinical potential for primordial follicles activation.

P213

EFFECT OF AUTOIMMUNE THYROIDITIS ON THE OUTCOME OF OVULATION INDUCTION IN PATIENTS WITH POLYCYSTIC OVARY SYNDROME

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Objective: To evaluate the prevalence of autoimmune thyroiditis (AIT) in patients with polycystic ovary syndrome (PCOS). Moreover, to investigate if AIT exerts any effect on the outcomes in polycystic ovary syndrome women undergoing ovulation induction. **Methods:** Over a period of 18 months, 116 patients with PCOS were recruited to this case-control study. 309 age-matched infertile women without PCOS were studied as a control group. A total of 104 PCOS Patients undergoing ovulation induction were divided into two groups: 31 patients (56 cycles) positive for antithyroid antibody (ATA+ group) and 73 women (106 cycles) negative for antithyroid antibody (ATA- group). **Results:** Thyroid function and thyroid-specific antibody tests revealed elevated thyroperoxidase (TPO) or thyroglobulin (TG) antibodies in 54 of 309 controls (17.5%), and in 39 of 116 patients with PCOS (26.9%; $P = 0.001$). The number of Gn days and early abortion rate was significantly higher. While the pregnancy rate was significantly lower in ATA+ group ($P < 0.05$). **Conclusion:** This prospective study demonstrates higher prevalence of AIT in patients with PCOS, correlated in part with an decreased pregnancy rate and an increased number of Gn days and early abortion rate undergoing ovulation induction.

P214

COMPARISON OF BIOCHEMICAL HYPERANDROGENISM IN LEAN AND OBESE WOMEN WITH POLYCYSTIC OVARY SYNDROME

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Obesity is not an essential feature in PCOS. Differences in biochemical hyperandrogenism among lean and obese patients with PCOS was investigated in this prospective study, on a cohort of diagnosed PCOS patients. They were subdivided into two groups based on BMI. BMI above 23 kg/m² = obese PCOS group, (n=54), BMI below 23 kg/m² = lean PCOS group. (n=54).

The mean ages of obese and lean PCOS groups were 24.6 ± 4.8 years, and 24.5 ± 4.7 respectively. The mean BMI of the two groups were 28.7 ± 5.7 kg/m² and 21.1 ± 1.5 kg/m². Mean Testosterone levels were 2.72 nmol/L and 2.18 nmol/L in obese and lean groups. Elevated Testosterone level was seen in 32 % of obese PCOS patients and 21.15% of lean PCOS patients. ($p = 0.000$). High FAI (FAI > 5) was found in 46.6% and 12.5% of obese and lean patients respectively. ($p = 0.043$). High BMI was found to be an independent predictor of high Testosterone level with an odds ratio of 2.984 (95% confidence Interval = 2.045 – 3.922.)

There is a statistically significant difference in Total Testosterone levels and FAI between lean and obese PCOS patients. Also, BMI is an independent predictor of elevated serum total Testosterone levels.

P215

INSULIN RESISTANCE AND PANCREATIC BETA CELL FUNCTION IN POLYCYSTIC OVARY SYNDROME: ARE THEY BMI DEPENDENT?

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Insulin resistance (IR) is a cardinal features of PCOS. A subgroup with IR develop pancreatic β -cell insufficiency that causes abnormal glucose tolerance. It is debated whether IR of PCOS is entirely BMI dependent, particularly in Asians

Aim was to compare IR and β -cell function in lean and overweight subjects with PCOS.

Retrospective cohort of 140 consecutive South Asians with PCOS were subdivided based on BMI as lean (≤ 23 kg/m²) and overweight (BMI ≥ 23 kg/m²). Fasting Blood Sugar (FBS) and serum fasting Insulin (FI) concentration were analyzed. Homeostatic Model Assessment was used to calculate insulin resistance (HOMA-IR) and pancreatic β -cell function. (HOMA-B)

N=140. Mean age 26.25 years \pm 5.61. BMI ≥ 23 was in 96 patients (68.6%) Mean BMI of overweight and lean groups were 28.56 \pm 4.65 and 21.15 \pm 1.31. Overweight subjects had significantly ($p=0.020$) higher mean FI (17.27 μ U/ml) compared to lean subjects (7.72 μ U/ml). HOMA-IR was 3.93 *versus* 1.77 in overweight and lean subjects respectively ($p = 0.057$). HOMA-B in overweight *versus* a lean was 1.40 and -1.28 ($p=0.321$).

Although overweight subjects with PCOS had higher fasting insulin; insulin resistance and β -cell function were not influenced by BMI. Insulin resistance and its consequence are independent of BMI in South Asians with PCOS.

P216

HOMOCYSTEINE LEVEL IN WOMEN WITH POLYCYSTIC OVARIAN SYNDROME

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Background: Polycystic ovarian syndrome (PCOS) is an endocrinopathy which generally involves obesity, ovulation disorders, and infertility. Cardiovascular complications are of long term risks of the condition. Plasma level of homocysteine (Hcy) is associated to cardiovascular risk and is an independent risk factor for the incidence of atherosclerosis.

Objective: To determine the mean homocysteine levels in women with PCOS.

Method: A cross-sectional study by measuring homocysteine levels in 20 PCOS women compared to 20 controls. The groups are matched based on body mass index (BMI) and age groups.

Results: 80% PCOS women are obese or overweighted, while the other 20% are normoweight. The mean Hcy levels of both groups are within the normal range (5-15 μ mol/L), in which PCOS women have lower Hcy levels compared to controls in the three BMI groups, which respectively are: obese (6.9 μ mol/L \pm 1.45 vs 8.41 μ mol/L \pm 1.6, $p=0.041$), overweight (6.04 μ mol/L \pm 1.26 vs 9.22 μ mol/L \pm 1.32, $p=0.002$), and normoweight (6.77 μ mol/L \pm 1.88 vs 7.05 μ mol/L \pm 0.47, $p=0.78$). Differences are statistically significant, except in the normoweight BMI group ($p>0.05$).

Conclusion: The majority of women with PCOS is a woman with above normal BMI. Mean Hcy levels in women with PCOS are within normal limits but lower than in women without PCOS. The mean Hcy level in this study represents very low risk of cardiovascular event in PCOS women, further studies are required.

Key Words: Polycystic ovarian syndrome, homocysteine, cardiovascular, body mass index.

P217

FOLLICULAR BCAA LEVELS ARE ASSOCIATED WITH OBESITY AND INSULIN RESISTANCE AND MAY COMPROMISE PREGNANCY OUTCOME

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Background: Disturbances in plasma branched-chain amino acids (BCAA, consisting of isoleucine, leucine and valine) metabolism have been implicated in obesity and polycystic ovary syndrome (PCOS), which may induce insulin resistance (IR) and further interfere with energy metabolism. However, evidence on BCAA metabolic profiles in the follicular fluid and its influence on pregnancy is sparse.

Methods: A prospective study including 63 PCOS patients and 48 controls was conducted in Peking University Third Hospital. Follicular BCAA levels were measured by the liquid chromatography-tandem mass spectrometric method.

Results: Isoleucine, leucine and valine levels were all positively correlated with BMI ($P < 0.05$). Overweight PCOS patients exhibited elevated BCAA levels compared with their lean counterparts ($P < 0.05$). Values of leucine and valine were increased in IR-PCOS patients in comparison with non IR group ($P < 0.05$). When patients were considered as a whole, the levels of BCAA were higher in the non-pregnant group than that in the pregnant group ($P < 0.05$). Subjects with BCAA levels greater than 239.10 μM showed a decreased pregnancy rate ($P = 0.036$) and higher abortion rate ($P = 0.004$) than those with BCAA levels below this cut-off value.

Discussion: BCAA metabolic disturbances are present in the local ovarian environment, especially in obese and PCOS patients with IR, which may negatively influence pregnancy outcome.

Key Words: PCOS, BCAA, obesity, follicular fluid

P218

CDKN2A-CDKN2B AND IGF2BP2 GENE POLYMORPHISMS IN WOMEN WITH POLYCYSTIC OVARY SYNDROME

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Objective: Insulin resistance is a core feature of PCOS. Genome-wide association studies (GWAS) have reported a number of SNPs with reproducible associations and susceptibilities to type 2 diabetes. We examined the potential association between the diabetogenic genes - CDKN2A-CDKN2B and IGF2BP2 - uncovered in the GWAS and PCOS in Korean women.

Design: Case-control study.

Measurements: DNA samples from 552 PCOS patients and 559 age-matched controls were genotyped. Four SNPs (rs564398, rs1333040, rs10757278 and rs10811661) in CDKN2A-CDKN2B and one SNP (rs4402960) in IGF2BP2 were evaluated. To investigate the association between the presence of PCOS and each individual SNP, logistic regression analyses were performed using the homozygote of wild-type allele as the reference category.

Results: Compared with the 559 controls, we found that PCOS was significantly associated with heterozygote of the rs10757278 in CDKN2A-CDKN2B, and their ORs ranged from 1.10 to 1.89. None of the remaining four SNPs (rs564398, rs1333040, rs10757278, rs10811661 and rs4402960) were associated with PCOS. For further analysis, the PCOS patients were divided into two or three subgroups according to genotype, and the associations between the genotypes and insulin resistance or insulin secretory capacity were assessed. No SNPs were significantly associated with HOMA-IR, HOMA _{β cell} (%), or 2-hour 75-g OGTT insulin levels in the PCOS patients; there were no significant associations with other serum hormonal and metabolic markers, such as androgen or glucose levels.

Conclusions: Our results suggest that except rs10757278 in CDKN2A-CDKN2B, most of the CDKN2A-CDKN2B and IGF2BP2 polymorphisms are not associated with PCOS.

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POLYCYSTIC OVARY SYNDROME (PCOS), THE SPECTRUM OF DISORDER IN EMIRATI WOMEN:

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Polycystic ovary syndrome (PCOS) is a common endocrine disorder affecting 10-21% of women. We have cases ranging from adolescents suffering from irregular periods, to married women suffering from infertility. Genetically, there is an association between the syndrome and insulin resistance. Weight gain and obesity are also common, along with a disturbance in the lipid profile. Psychological symptoms of PCOS include anxiety, depression, eating disorders, and psychosexual dysfunctions. Mood swings, sleep irregularities and an abnormality of social interaction are also common for this condition. Inheritance of PCOS appears to have a complex genetic basis which interacts with lifestyle and other environmental factors.

The goal of this study is to evaluate PCO cases in Qassimi hospital sharjah. A total 96 PCO patients were studied during one year duration. All patients underwent detailed history and thorough clinical evaluation. Ultrasonography and hormonal profiles were performed. The most common presentation was hirsutism (42.4%) more than 50% were overweight or obese according to the Body mass index (BMI). Fasting and postprandial Glucose and Insulin levels were abnormal in 30 %. About 40% of the women had a serum total Cholesterol level above 200 mg/dL, while Low Density Lipoprotein (LDL) Cholesterol was above and High Density Lipoprotein (HDL) cholesterol was lower than the desirable value.

The best first-line treatment for PCOS is a lifestyle modification that involves exercise and a balanced diet. Managing the patient's BMI (body mass index) is key to resolving PCOS and reducing the BMI to <30 kg/m² has been beneficial for most patients.

P220

POLYCYSTIC OVARY SYNDROME IN KERMANSHAH: CLINICAL AND ENDOCRINE

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Introduction: Polycystic Ovarian Syndrome (PCOS) is probably the most prevalent endocrinological disorder affecting women and it is the most common cause of menstrual disturbance during the reproductive age. The aim of this study is to investigate and analyze the clinical presentation, LH/FSH profile, and BMI of healthy women with polycystic ovary syndrome (PCOS).

Methods & materials: In this case - control study at Imam Reza in Kermanshah 71 patients with PCOS and 84 healthy women were studied. Body Mass Index (BMI) assessed to be matched in two groups. Clinical presentation, the LH / FSH, menstrual disorders in the two groups were compared. For data analysis, the chi-square test, Mann-Whitney U and independent sample T-test by SPSS software were used.

Results: The average age was 27.06±4.1 years in case and 30.27±5.6 years in control subjects. 16 cases (22.5%) and 12 control subjects had acne which were no significant differences (P = 0.212). 58 patients (81.7%) and 15 healthy women (17.9%) had oligomenorrhea, this difference was significant (P = 0.001). Mean of LH level was in the control group 5.4 ± 3.3 mIU / ml and in case group 9.8 ± 5.8 mIU / ml. The ratio of LH / FSH: 2.03 and 0.987 in cases and controls, respectively, which was statistically significant (P = 0.001).

Conclusion: No significant statistical correlation is found between LH/FSH ratio, menstrual pattern and obesity. This disproves the traditional concept about PCOS which regards that the heavier the patient is the higher the LH or the worst manifestations.

Keywords:

Polycystic ovary syndrome, Kermanshah, LH/FSH

P221

MANAGEMENT OF POLYCYSTIC OVARIAN SYNDROME: ROLE OF METFORMIN AND PIOGLITAZONE

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Introduction: PCOS is one of the most common endocrine diseases in women. This syndrome is characterized by hyper-androgenism, chronic anovulation, infertility and obesity. One of the modality of treatment in this disorder is insulin sensitizing agents. This study compared effects of metformin and pioglitazone in PCOS patients with menstrual disturbance. **Material and Methods:** We conducted a randomized clinical trial of 56 women aged 20-49 years, with AUB. Participants were randomly assigned to either cyclical 500 mg metformin TDS for three cycles or 30 mg pioglitazone once daily for 3 months. Clinical symptoms and Para clinical parameters such as (BP, BMI, FBS, TG, Chol, LDL, HDL, Testosterone, DHEAS) were recorded before and 3 months after initiation of the treatment. **Results:** Clinical symptoms such as hair loss improved after metformin treatment ($p = 0.08$). diastolic blood pressure reduced after metformin treatment, while after pioglitazone treatment was not significantly changed ($P = 0.02$). BMI didn't changed in before and after treatment in both groups ($p = 0.98$). Waist circumference significantly decreased only in metformin group. The mean of cholesterol before and after treatment in two groups were not significantly changed, while only Triglycerides in pioglitazone were statistically decreased. LDL, HDL and testosterone levels in two groups were not significant changed. However DHEAS and Insulin levels in both groups showed a significant decreased.

Conclusion: We find a significant amelioration of endocrine and metabolic indices with pioglitazone in PCOs patients; it appears this drug offers a useful alternative treatment for women who couldn't tolerant metformin. **Keywords:** Poly cystic ovary syndrome, metformin, pioglitazone, endocrine and metabolic criteria

P222

CORRELATION BETWEEN HYPERINSULINEMIA AND SEVERITY OF HIRSUTISM IN PCOS PATIENTS: A PRELIMINARY STUDY

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Objective

It is still not clear whether only androgen responsible to the development of hirsutism in PCOS patients or there is other hormones such as insulin plays a role. One study has documented that insulin gave direct effect on the severity of hirsutism in PCOS and have a synergetic interaction with testosterone. The aim of current study is to determine whether hyperinsulinemia determines the severity of hirsutism in PCOS patients.

Method

This is a cross sectional study conducted in Yasmin Reproductive Clinic, Dr. Cipto Mangunkusumo General Hospital, Jakarta, Indonesia in 2013. Up until now, 23 PCOS patients participated in the study. All participants were examined for their hormonal profile, fasting insulin, fasting glucose, OGTT, SHBG level, free testosterone, total testosterone, and ferriman gallwey score (FGS). The groups were divided according to the presence and absence of insulin resistance based on the HOMA-IR level according to previous study (HOMA-IR level >3.308 is the cut of point for the diagnosis of insulin resistance in PCOS patients in Indonesia).

Result(s)

The groups consisted of 7 PCOS patients with insulin resistance (mean age: 29.2 ± 5.6 ; mean BMI 29.92 ± 5.2) and 16 subjects without insulin resistance (mean age 27.25 ± 2.712 ; mean BMI 27.58 ± 5.86). This study demonstrates that the mean of FGS in insulin resistance group (4.40 ± 2.96) is higher than in non-insulin resistance group (1.63 ± 3).

Conclusion

The present data suggest that development and the progression of hirsutism in patients with PCOS is associated with the circulating levels of insulin.

Keywords:

Hirsutism, hyperinsulinemia, PCOS

P223

PCOS patient in Indonesian showed a different characteristic of Ferriman Gallwey score

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Background

Polycystic ovary syndrome (PCOS) is a heterogeneous endocrine disorders marked by anovulation, androgen excess and polycystic ovaries, which is based on Rotterdam criteria. It is happens on 5%-10% women at reproductive age. Clinical hyperandrogenism are commonly used in clinical practice by assessing the hirsutism with Ferriman Gallwey score (FGS) ≥ 8 , due to impracticality of biochemical hyperandrogenism to confirm the diagnosis of hyperandrogenism. However, the universal score of FGS ≥ 8 are not found in the data with hyperandrogenemia from Yasmin clinic. Hence, It is important to find the cut off of FGS in Indonesia to reflex the free testosterone level.

Objectives

Find the cut off of Ferriman Gallwey score with free androgen index in PCOS patient in Indonesia

Methods

Cross sectional study was conducted at Yasmin Reproductive Clinic, dr.Cipto Mangunkusumo Hospital, Jakarta, Indonesia. Data was collected from the medical records of 20 women with PCOS. The data than will be analyzed using SPSS 11 and using ROC to find the cut off of FGS.

Result

The data obtained from 20 pcas patients found that between age 25-33 years old showed no data found of FGS ≥ 8 , 75% patient with higher level of free androgen index showed FGS score ≥ 3 with 5.61 (SD 4.97).

Conclusion

High free testosterone levels are not followed with FGS ≥ 8 in Indonesia, the universal score of FGS ≥ 8 cannot be used in Indonesia, score for Indonesian FGS is ≥ 3

Keywords: Ferriman Gallwey Score, Free Androgen Index, PCOS

P224

DOES TRIGLYCERIDE INFLUENCE HOMA-IR IN PCOS PATIENTS?

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Introduction: PCOS is a common condition in reproductive-aged women and frequently associated with impaired glucose tolerance (IGT), type 2 diabetes mellitus (DM2) and metabolic syndrome.

Objective: To determine the association between age, waist circumference (WC), BMI, and lipid profile with Homeostatic Model Assessment Insulin Resistance (HOMA-IR)

Material(s) and Methods: Data of 18 women with PCOS was collected. BMI was stratified as: normal, overweight, obese. Lipid profile, and WC was stratified as normal and high. PCOS was diagnosed by the Rotterdam criteria 2003 consensus workshop. Insulin resistance was measured using the HOMA-IR. Statistical analysis was performed using the SPSS 15.0.

Results: Among the PCOS subjects, the mean \pm SD age, BMI, WC, LDL, total cholesterol, and triglyceride were 27.56 ± 2.77 years, 27.8 ± 5.5 kg/m², 91 ± 12 cm, 125.44 ± 27.18 mg/dl, 184 ± 27.9 mg/dl, 123.7 ± 71.5 mg/dl. The median of HOMA-IR was 2.815. In bivariate analyses, the correlation between HOMA-IR and age ($p=0.569$), BMI ($p=0.515$), WC ($p=1.000$), LDL ($p=1.000$), total cholesterol ($p=1.000$), and TG ($p=0.083$). But in multiple linear regression analyses, TG was found to be associated with HOMA-IR ($p=0.021$)

Conclusion: There is no significant association between aged, WC, body fat percentage, BMI, total cholesterol and LDL with levels HOMA-IR in PCOS women. But increasing HOMA-IR level in PCOS women can be affected by increasing level of triglycerides.

Keywords: PCOS, HOMA-IR, BMI, LDL, total cholesterol, age, waist, triglycerides, body fat percentage

P225

CHARACTERISTICS OF PCOS AS THE CHANGES OF DIET PATTERN FOR A DECADE IN KOREA.

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Though there are significant racial variations in the incidence and clinical manifestations of PCOS, it is not well known about the delicate origin of those variations. We hypothesized that diet pattern is an important factor for the racial variations, and investigated time-based changes of diet pattern and clinical presentations of PCOS in Korean women. The medical records of PCOS women, who were diagnosed in their twenties, based on 2003 Rotterdam criteria, were reviewed: women who were diagnosed from Jan to Dec. 2001 were enrolled as group A and those from Jan to Dec. 2011 were selected as group B. Data from the 2nd (2001) and 5th (2011) Korean National Health and Nutrition Examination Survey were used for the evaluation of the changes of diet pattern. In group B, serum free testosterone level was higher than that of group A and hirsutism and acne were more frequent. According to 2011's KNHANES data, daily calorie intake of twenties women was reduced compared with 2001's. However, simple carbohydrate intake and fried foods, beverages, and animal foods consumption were increased. Mean BMI and the prevalence of obesity were also increased. As the change of diet pattern, increased incidence of hyperinsulinemia and related hyperandrogenism were expected and more PCOS women actually have expressed laboratory and clinical hyperandrogenism than 10 years ago. To conclude this matter, large scaled repeated studies with same design every ten years and population based cohort studies are needed.

P226

CAN ANTI MULLERIAN HORMONE BE A PREDICTOR OF METABOLIC SYNDROME IN POLYCYSTIC OVARIAN SYNDROME?

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Background: The purpose of this study was to evaluate whether Anti Mullerian Hormone (AMH) can be used as a predictor tool of metabolic syndrome in Polycystic Ovarian Syndrome (PCOS).

Method: This cross sectional study is conducted in Yasmin Clinic, Cipto Mangunkusumo General Hospital Jakarta between June to December 2012. Forty-one patients diagnosed with PCOS based on Rotterdam Criteria were enrolled. Secondary data was taken from medical record. The data consists of level of AMH, and risks of metabolic syndrome such as age, body mass index, waist circumference, fasting blood glucose, HDL, triglyceride, and insulin resistance have been evaluated using SPSS 11.0.

Result: There were twenty two patients found to have metabolic syndrome in this study. Mean AMH level in PCOS patient with metabolic syndrome is found higher than patient without metabolic syndrome (10.72 ± 6.23 ng/ml vs 7.97 ± 4.50 ng/ml, $p=0.12$). While AMH is being observed in each variable of metabolic syndrome, it was shown AMH has association with HDL, triglyceride and insulin resistance with r-value of -0.29, 0.23, and 0.21 respectively, with p-value of < 0.05 .

Conclusion: AMH can be considered as a predictor of metabolic syndrome in PCOS.

Keywords: Polycystic Ovarian Syndrome, Anti Mullerian Hormone, Metabolic Syndrome

P227

EFFECTIVENESS OF RAT BONE MARROW STEM CELL THERAPY IN RATS POLYCYSTIC OVARIAN SYNDROME MODEL ON FOLLICULOGENESIS AND THE EXPRESSION OF TRANSFORMING GROWTH FACTOR-?

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The objective of this study was to treat PCOS using stem cell to enhance folliculogenesis to improve fertility. This laboratory experimental study used rats injected with testosterone propionate of 100 mg/kg BW for 14 days. On day 15 vaginal swab was done to determine the estrous cycle. The rats were given with intravenous injection through tail with single dose of 1×10^6 Rat Bone Marrow Stem Cell as therapy. The next day, the rats were sacrificed and the ovaries were taken to examine the estrous cycle and the folliculogenesis using Haematoxylin Eosin staining. The examination of Transforming Growth Factor (TGF- β) expression was done using immunohistochemistry. The estrous cycle of PCOS rats after being injected with RBMSC therapy could return to fertile condition since many of the rats gained estrous and pro-estrous phases. In control and treatment groups, primary follicles were 1.93 ± 1.03 vs 2.80 ± 1.01 ; secondary follicles 1.80 ± 1.45 vs 2.87 ± 1.59 ; and tertiary follicles were 0.93 ± 0.59 vs 3.40 ± 1.84 . The number of de Graaf's follicles increased 0.07 ± 0.02 in control group vs 1.07 ± 0.07 in treatment group. All folliculogenesis phases were different significantly. The expression of TGF- β also increased significantly compared to control group (1.4 in control group vs 2.4 in treatment group, $p = 0.0026$), indicating significant difference. In conclusion, the provision of RBMSC to PCOS rats can improve fertility, folliculogenesis and the increase of TGF- β expression.

Keywords: PCOS rats, stem cell, RBMSC therapy, folliculogenesis, estrous cycle

P228

PLASMA HEAVY METALS ARE ASSOCIATED WITH HORMONE DISTURBANCES AND NEGATIVE PREGNANCY OUTCOME IN PATIENTS WITH IVF TREATMENTS

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Background: Abnormal levels of heavy metals exposure is suggested to be related to reproductive dysfunction. However, its impacts on reproduction in patients seeking for in vitro fertilization and embryo transfer (IVF-ET) treatments especially patients with polycystic ovary syndrome (PCOS) are limited.

Methods: We conducted a prospective study in 40 PCOS patients and size-matched controls in the Peking University Third Hospital. Plasma levels of eight heavy metals were measured by inductively coupled plasma mass spectrometry.

Results: Compared with the control patients, PCOS patients exhibited higher plasma levels of Cr, Hg and Cu. No differences were found with regards to the plasma Mn, Pb, Se, Zn, and Mg levels. Cu level was found to be positively correlated with body mass index, while Hg level was correlated with luteinizing hormone and androstenedione levels. All the eight heavy metal levels were comparable in insulin resistant and non-insulin resistant PCOS patients as well as in patients with and without hyperandrogenism. Cu levels were elevated in non-pregnant patients in both PCOS and control group. Moreover, the risk of pregnancy failure with Cu elevation was 16.91 times increased and remained significant after further adjustment for age, BMI and homeostasis model assessment of insulin resistance.

Conclusions: PCOS patients exhibited altered heavy metal levels. Hg may serve as a negative factor for endocrine function, while Cu may contribute to pregnancy failure. This study provides valuable information to the impacts of heavy metal on reproduction.

Key Words: heavy metal, PCOS, pregnancy, IVF

P229

PCOS AND IVF: COMPARING FOUR PHENOTYPES UNDER THE ROTTERDAM CRITERIA

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Introduction: In 2006 Azziz defined four different phenotypes of PCOS under the Rotterdam criteria (Fertil. Steril. 2006).

Feature	Phenotype A	Phenotype B	Phenotype C	Phenotype D
Hyperandrogenism (clinical+/- biochemical)	X	X	X	
Oligoanovulation	X	X		X
Polycystic Ovaries on Ultrasound	X		X	X

Objective: To compare the outcome for the 4 phenotypes in the setting of IVF treatment.

Design: Retrospective Cohort study in a single center and single physician setting.

Materials and Methods: 107 patients meeting the diagnosis of PCOS under the Rotterdam Criteria were included. All patients were undergoing controlled ovarian stimulation (COS) for IVF between 2009 and 2012.

Findings: The PCOS patients were divided into four groups based on the above phenotypes. The relative prevalence was 49.5%, 3.7% , 28% and 18.7% for Phenotypes A, B, C and D respectively. Phenotype D had the lowest BMI.

Otherwise there were no significant differences. The live birth rate per transfer was 53.3%, 50%, 59.3% and 40% for phenotypes A, B, C and D respectively.

Conclusions: In this study of PCOS patients undergoing IVF treatment, the studied differentiation of PCOS phenotypes did not reveal any significant differences in IVF performance.

P230

AREG EXPRESSED IN CUMULUS CELLS AS A BIOMARKER FOR OOCYTE MATURITY IN PCOS PATIENTS

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The objective of this study was to explore whether the expression level of key genes (LHR, AREG, EREG and EGFR) involved in peri-ovulatory event induced by LH surge was altered in granulosa and cumulus cells in polycystic ovary syndrome (PCOS) patients and whether they were related to the clinical outcomes. 43 infertile PCOS women and 59 infertile women with tubal blockage or male factors were recruited. No matter in GCs or CCs, none of these genes was significant differentially expressed between PCOS and control patients. The AREG level in GCs was negatively correlated with oocyte retrieval number in PCOS patients ($r=-0.384$, $P=0.03$). We further cultured GCs and confirmed that the expression level of AREG and EREG mRNA peaked at 4 hours and gradually declined to basal level by 24 hours after HCG treatment. In contrast with GCs, the AREG level in CCs of PCOS patients was positively correlated with the oocyte retrieval ($r=0.71$, $P=0.01$), MII oocytes, available embryos ($r=0.782$, $P=0.003$) and good quality embryos ($r=0.678$, $P=0.015$). However, no correlation of the gene expression and laboratory outcomes was found in control patients. Our research confirmed that there is a time-dependent change of EGF-like factors in human beings. For PCOS patients, a delayed decrease in AREG in GCs may have a negative effect on laboratory outcomes, while elevated AREG and EREG expression in CCs may result in increased number of oocyte retrieval and available embryos. Thus the AREG might be a good marker to predict the outcome of PCOS patients.

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DYNAMICS EPIGENETIC MARKS IN THE BRAIN OF MOUSE CONCEIVED FROM PGD/PGS BLASTOMERE-BIOPSIED EMBRYOS

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Blastomere biopsy has been widely applied in the pre-implantation genetic diagnosis (PGD) or pre-implantation genetic screening (PGS). However, the long-term risks of PGD/PGS on offspring are still unknown. In the present study the effects of blastomere biopsy on the early development, post-implantation characteristics, and birth rate were determined. Moreover, the behavior of the offspring conceived from the biopsied embryos was evaluated with Morris water maze and pole climbing, and the dynamics epigenetic marks in the brain tissue were analyzed. Finally the correlation between aberrant behavior and dynamics methylation modification was evaluated. The results showed that the duration from 4-cell to blastocyst of biopsied embryos was prolonged significantly, and the blastocyst quality was also decreased. In the behavior testing, PGD/PGS mice spent more time on the non-trained quadrant and climbing down the pole. Furthermore, H19/Igf2 differential methylation regions showed decreased methylation patterns, but Snrpn and Igf2r was normal compared with the control group. Quantitative RT-PCR indicated significantly decreased Igf2 mRNA expression, but normal H19, Snrpn, Igf2r and Ube3a expression profiling. For the correlation analysis, the mice with abnormal H19 methylation modification tended to show impaired probing behaviors by Morris water maze, but there was no observed correlation between pole climbing ability and H19 methylation level. In conclusion, there are potential risks accompanying blastomere biopsies in PGD procedures on embryo development and the behavior of resulting offspring, which possibly arises from aberrant epigenetic modification and methylation patterns in brain tissues. Further studies are needed to better understand the risks of PGD.

P232

A NEW METHOD FOR PREIMPLANTATION GENETIC DIAGNOSIS OF MONOGENIC DISEASES BASED ON TARGETED NEXT-GENERATION SEQUENCING AND PEDIGREE HAPLOTYPE ANALYSIS

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Next generation sequencing (NGS) is a high-resolution, high-throughput and cost-effective technology. It has been used in many aspects of medical research and even some clinical applications. Preimplantation genetic diagnosis (PGD) of monogenic disease was established more than two decades ago which offered couples at risk of a genetic disease the chance to have an unaffected child. Here we report the application of target-enrichment technology and NGS for PGD of monogenic disease. We designed a novel DNA chip for SNPs enrichment, and then hundreds of SNPs closely linked to the target gene could be detected by NGS. The SNPs were used as genetic markers for pedigree haplotype analysis, and the genetic status of the embryo could be inferred by this method. Furthermore, we had completed the genetic analysis of 6 embryos and the results were consistent with those detected from MF-PCR provided by the reference laboratory. Compared with conventional method, the advantages of targeted-NGS are obvious. Firstly, targeted-NGS can provide more accurate diagnosis. This new application could dramatically reduce risk of misdiagnosis from ADO phenomenon and recombination because high-density and gene closely linked SNP markers were used for embryonic haplotype analysis. Secondly, embryo test can be completed within one month or even less, no additional individualized pre-test required. Thirdly, any known monogenic disease can be detected using this method, and the detection process is no difference among different genes and different genetic background people. Fourthly, the method is cost-effective because the cost of sequencing is falling drastically and less labor-time required.

P233

EUPLOIDY AND ANEUPLOIDY RATES COMPARISON OF CLEAVAGE STAGE AND BLASTOCYST STAGE USING MICRO-ARRAY COMPARATIVE GENOMIC HYBRIDISATION (MACGH)

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Introduction

In this study, we compared the euploidy and aneuploidy rates of PGD cases in Alpha International Fertility Centre, Malaysia following biopsy at the cleavage versus the blastocyst stage using Micro-array Comparative Genomic Hybridisation (MaCGH) for patients less than 37 years old.

Materials and Methods

130 patients had cleavage stage biopsy, while 18 patients had blastocyst stage biopsy MaCGH from July 2011 to August 2013. The mean maternal age of patients from the cleavage stage and blastocyst stage biopsy MaCGH cases are 30.5 and 29.8 respectively ($p=0.1$). 1151 cleavage stage embryos had a blastomere cell biopsied to perform MaCGH. Whereas for the blastocyst stage biopsy MaCGH, 108 blastocysts had 3 - 7 trophectoderm cells biopsied. The biopsied sample(s) and reference DNAs were amplified, labelled and hybridised according to manufacturer's (BlueGnome) specifications. Microarray slides were analysed using BlueFuse Software Version 3.1 (BlueGnome), revealing normal or whole and partial chromosome losses and gains. Cases excluded from the study were those with known altered karyotype of translocation and inversion; and indeterminate biopsies.

Results

The euploidy rates of cleavage stage embryos and blastocysts were 31.0% and 41.9% ($p=0.01$) respectively. The aneuploidy rates of cleavage stage embryos and blastocysts were 63.5% and 50.4% ($p=0.01$) respectively.

Conclusions

Blastocysts have significantly higher euploidy rate compared to cleavage stage embryos. We believe this is the primary reason for the higher implantation rate for blastocysts compared to cleavage stage embryos, and supports the practice of blastocyst culture and transfer.

P234

CHROMOSOMAL CHARACTERIZATION AT DIFFERENT EMBRYONIC DEVELOPMENTAL STAGES

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Objective: To explore the chromosomes status of the cleavage embryos and the blastocysts.

Materials and methods: 18 blastocysts were included in this study. Both one blastomere and the several trophectoderm cells were biopsied in all the 18 embryos. Biopsied blastomeres and the trophectoderms were examined by CGH or SNP chips.

Results: Chromosomes status of 11 embryos (61.11%, 11/18) at cleavage stage are completed consistent with those at blastocyst stage, including 7 chromosome balanced embryos and 4 abnormal embryos. Chromosomes status of 6 embryos (33.33%, 6/18) at cleavage stage are consistent with those at blastocyst stage. These 6 embryos are all abnormal embryos. In 5 embryos, chromosome abnormal at blastocysts stage is less than that at cleavage stage. Moreover, there is 1 embryo which chromosome is abnormal at cleavage stage but balanced at blastocyst stage.

Conclusions: 1. Most blastocysts keep the chromosome stabilization during the embryo development. 2. Self genetic correction could be happen in some embryos during development to the blastocyst stage. 3. Both CGH and SNP arrays are good technique to examine the micro-DNA. Compared with the blastomere DNA, trophectoderm DNA could provide a better result.

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IMPROVED EFFICIENCY OF MICROSURGICAL ENUCLEATED TRIPRONUCLEAR ZYGOTES DEVELOPMENT AND EMBRYONIC STEM CELL DERIVATION BY SUPPLEMENTING EPIDERMAL GROWTH FACTOR, BRAIN-DERIVED NEUROTROPHIC FACTOR AND INSULIN-LIKE GROWTH FACTOR-1

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Human embryonic stem cells (hESCs) hold great promise for future clinical cell therapies because of their unique potential to differentiate into all human cell types. However, the destruction of normal fertilized embryos and derivation of hESCs for research has resulted in polarized ethical debates, with most of the controversy centered on embryo destruction. Therefore, because of less ethical controversy surrounding them, abnormal fertilized zygotes that are usually discarded are a potential feasible resource for the derivation of hESCs. Microsurgery on human polyspermic zygotes can contribute to the derivation of human ESCs, but the efficiency is much lower. Here we reported a culture system to enhance the development competence of such microsurgical human polyspermic zygotes by EGF-BDNF-IGF-1 combination, which eventually resulted in the increased derivation efficiency of human embryonic stem cells from them. We found that the developmental efficiency of microsurgical enucleated trippronuclear embryos cultured with the EGF-BDNF-IGF-1 combination was significantly increased compared with control group. More importantly, when the microsurgical enucleated trippronuclear embryos were cultured in medium supplemented with EGF-BDNF-IGF-1, the frequency ratio of chromosome abnormality was reduced.

Our present study will facilitate the development of hESC lines derivation in subsequent studies and also provide an additional choice for infertile couples.

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DERIVATION OF HUMAN CLONED EMBRYOS WITH DIFFERENT CHROMOSOME PLOIDY BY NUCLEAR TRANSFER OF HUMAN SOMATIC NUCLEI INTO HUMAN NON-ENUCLEATED OOCYTES

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Therapeutic cloning has tremendous potential in regeneration medicine, however only one successful study was reported in 2013. Here we investigated the chromosome ploidy variation of human cloned embryos by transfer of human somatic nuclei into human non-enucleated oocytes using FISH method. The pronuclear position after activation and Y chromosome identification at 8-cell stage were used to analyze the chromosome ploidy. There are two types of chromosome ploidy in human SCNT embryos, including diploid and triploid. 14.3% embryos were not found pronuclear formation. 28.6% embryos were diploid and 57.1% embryos were triploid. In about 60% diploid embryos, the somatic nuclei were not activated successfully and only the pronuclear near the first polar body was found. In these embryos, Y chromosome could not be tested in blastomere biopsied from 8-cell embryos. In other diploid embryos, the somatic nuclei were activated successfully, but the nuclei of oocytes was failed to be activated. In these embryos, the pronuclear was far away from the first polar body, and Y chromosome could be tested in blastomere biopsied from 8-cell embryos. The cloned embryos with different chromosome ploidy were derived, however we found none of the cloned embryos with diploid chromosome ploidy developed via 8-cell stage, whereas about 35% the cloned embryos with triploid chromosome ploidy could develop to blastocyst stage. The present results suggested that human non-enucleated oocytes could reprogram the somatic nuclei successfully and form the cloned embryo with diploid or triploid chromosome ploidy, but the diploid embryos seemed not develop to blastocysts.

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FOLLICLE-STIMULATING HORMONE (FSH) DOWN-REGULATES PROFILIN-1 THROUGH ACTIVE OF THE IP3/DAG PATHWAY IN HUMAN GRANULOSA STEM CELLS (HGSCS) DERIVED FROM IN VITRO FERTILIZATION (IVF)

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Introduction:

FSH is a hormone to regulate the ovarian follicle and associated granulosa cell development, growth, maturation. These human granulosa cells play an important role in the process of oocyte maturation; however, the pathways involved in the differentiation of hGCs remains controversial. In this study, human granulosa stem cells (hGSCs) were treated with FSH to investigate the proteomic signature in hGSCs.

Materials and methods:

These hGSCs were collected from 10 patients undergoing IVF procedures after controlled ovarian stimulation. Cells were treated with rFSH for 21 days, qPCR and Proteomic analysis were used to detect the function of hGSCs.

Results:

These hGSCs expressed *Oct-4* (stem cell marker), *CD90*, *CD105*, *vimentin* and showed differentiation potential. In proteomics profile, these rFSH-treated hGSCs were compared with hGSCs using 2-D gel electrophoresis. 319 and 292

protein spots were detected in the hGSCs treated with/without rFSH, respectively. Total 291 protein spots were found to match each other with high homogeneity. LC-MS/MS sequence analysis, we found that FSH down-regulates profilin-1 protein expression in the hGSCs. The profilin-1 was regulated through the IP3/DAG pathway.

Conclusion:

Profilin-1 can bind to actin and affects the structure of the cytoskeleton and, inhibit the formation of IP3 and DG by binding to PIP2. These findings represent that FSH can down-regulates profilin-1 in gene and protein level. In the future, we will investigate the function of profilin-1 in the granulosa stem cells. This model may be beneficial in studying hormonal regulation associated with follicular maturation and the pathogenesis of polycystic ovary syndrome (PCOS).

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GET READY FOR FLOOD OF FETAL GENE TESTING

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INTRODUCTION-

A **DNA microarray** (also commonly known as DNA chip or biochip) is a collection of microscopic DNA spots attached to a solid surface. Scientists use DNA microarrays to measure the expression levels of large numbers of genes simultaneously or to genotype multiple regions of a genome.

Four types of testing program are available-

- Newborn screening
- Carrier testing
- Prenatal testing
- Presymptomatic (predictive) testing

American College of Obstetricians and Gynecologists (ACOG) and European Best Practice Guidelines do not yet include offering CMA to all patients undergoing invasive prenatal testing

ACOG has endorsed CMA an appropriate test in prenatal cases with abnormal ultrasound findings and a normal karyotype

Advances in invasive prenatal diagnosis: Chromosomal microarray (array-CGH, molecular karyotyping) will replace conventional karyotyping.

OBSERVATION & RESULTS

We in our study of 10 cases of ultrasound detectable anomalies and abnormal serum marker subjected these to amniocentesis for Karyotype and CMA studies. All 10 cases with increased NT and Nasal Bone anomalies and raised MOM in serum marker tests showed a Normal Karyotype, while CMA picked up gene defects.

DISCUSSION:

CMA can also be applied to free fetal DNA in Maternal blood at 8 weeks gestation to get a complete genome study of the fetus. This in future will make accurate prenatal genetic diagnosis possible with just a few ml of maternal blood.

Gene testing is here and in the next few years there will be a flood of gene tests available.

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LEUCINE-RICH REPEATS AND WD REPEAT DOMAIN CONTAINING 1 (LRWD1) IS ASSOCIATED WITH REACTIVE OXYGEN SPECIES (ROS) RESPONSE IN TESTICULAR CELLS

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Abstract:

The human leucine-rich repeats and WD repeat domain containing protein 1 (LRWD1) is richly expressed in the testes. In human spermatozoa, LRWD1 colocalizes with centrin in the centrosome. We suggest that the testis-enriched LRWD1 is an important factor in human spermiogenesis and tail formation, yet its roles in male germ cell development and spermatogenesis remain unclear. The objective of this study is to investigate the expression and function of LRWD1 in human sperms. We used the PROMOTER SCAN software (<http://www.bimas.cit.nih.gov/molbio/proscan/>) to analyze the 1-kb segment immediately upstream to the LRWD1 translational start site and revealed the NF-κB (nuclear factor kappa-light-chain-enhancer of activated B cells) and NRF2 (nuclear factor (erythroid-derived 2)-like 2) binding sites at -133 to -125 and -9 to -5, respectively. NF-κB and NRF2 are activated by reactive oxygen species (ROS) exposure of NTERA-2 cl.D1. We used a series of deletion constructs with a dual luciferase reporter system to identify the core promoter of LRWD1. Site-directed mutagenesis and electrophoretic mobility shift assays (EMSA) confirmed an NF-κB binding element within the LRWD1 core promoter. Overexpression of NF-κB in cells enhanced the LRWD1 promoter activity *in vitro*. Our results suggest that NF-κB regulates the expression of LRWD1. In future work, we will investigate how NRF2 affects LRWD1 expression and focus on the regulatory role of NF-κB and NRF2 in LRWD1 expression under ROS and/or electrophiles exposure. Understanding the LRWD1 expression regulated by NF-κB and NRF2 will help develop diagnosis and/or treatment of male infertility and other diseases.

P240

SPERM AUTOSOME ANEUPLOIDY HAS A GREATER EFFECT ON EMBRYO GROWTH RATES THAN SEX CHROMOSOME ANEUPLOIDY

K. Harrison¹, N. Martin², J. Irving¹

¹Embryology, Queensland Fertility Group, Brisbane, Australia

²Cytogenetics, Queensland Fertility Group, Brisbane, Australia

Background

We previously demonstrated slower embryo development and decreased embryo morphology with increasing overall levels of male partner sperm chromosome aneuploidy.

Aim

Whether raised sperm sex chromosome and autosome aneuploidy levels contribute equally to slower embryo growth.

Method

55 couples having ICSI where the male had sperm aneuploidy assessed by 5-probe chromosomal FISH (X,Y,13,18,21). Normal chromosomal aneuploidy threshold was set at 2% for each chromosome. Embryos were assessed for early cleavage at 24hrs post-injection and subsequent developmental milestones through to D5 blastocyst formation.

Results

Control group (20 couples, 141 embryos) with all sperm aneuploidy levels <2% showed 30% cleavage at 24 hours, 62% with >3 cells on D2, 73% with >5 cells on D3, and 44% blastocysts on D5. The equivalent levels for raised levels of sex chromosomes >2% (5 couples, 30 embryos) were 23%, 47%, 37% and 35%. The embryo parameters for raised levels of a single autosome >2% (14 couples, 101 embryos) were 11%, 50%, 51% and 17%. Embryo parameters for raised levels of two autosomes >2% (4 couples, 20 embryos) were 11%, 50%, 51%, and 17%. For raised levels of two autosomes plus the sex chromosomes all >2% (6 couples, 48 embryos) the embryo parameters were 4%, 62%, 41% and 13%.

Conclusion

Raised levels of sperm autosome aneuploidy and raised levels of mixed sex chromosome and autosome aneuploidy have a greater negative effect on 24hrs zygote cleavage and D5 growth to blastocyst than do raised sex chromosome aneuploidy levels alone. This aids counselling cases with raised sperm aneuploidy.

P241

HYPERMETHYLATION OF THE LEUCINE-RICH REPEATS AND WD REPEAT DOMAIN CONTAINING 1 (LRWD1) PROMOTER IN SPERMS

Y. Teng¹, N. Yin-Mei Su¹, N. Shi-Kae Wee¹, N. Che-Chia Tsao¹, N. Yung-Chieh Tsa², N. Hsien-An Pan³, N. Pao-Lin Kuo⁴, N. Ping-Chi Hsu⁵

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⁵Department of Safety Health and Environmental Engineering, National Kaohsiung First University of Science and Technology Kaohsiung Taiwan., Kaohsiung, Taiwan

LRWD1 (Leucine-Rich repeats and WD repeat domain containing 1) is highly expressed in the testes but down-regulated in the testicular tissues of patients with severe spermatogenic defects. We used the EMBOSS CpG Plot to analyze the 500-b.p. fragment immediately upstream to the LRWD1 translational start site and revealed the CpG islands are located approximately between positions -253 to +5 from the LRWD1 transcription start site (TSS). A total of 34 CpG sites were located within the predicted CpG islands (total length 258 b.p.). The CpG islands contain the LRWD1 core promoter at -198 to +1 from the TSS. Within the core promoter region, we identified the NF-κB (nuclear factor kappa-light-chain-enhancer of activated B cells) and NRF2 (nuclear factor (erythroid-derived 2)-like 2) binding sites at -133 to -125 and -9 to -5, respectively. In addition, we cloned the methylation-specific PCR products of the LRWD1 promoter in bisulphate-modified DNA from 30 normal semen samples. We suggest that LRWD1 is down regulated via CpG island methylation in its promoter. In future work, we will investigate how DNA methylation of the CpG island affects the LRWD1 promoter activity in human sperms. The present study represents a significant step forward to understand the LRWD1 expression regulated by DNA methylation and to develop diagnosis and/or treatment of male infertility and other diseases.

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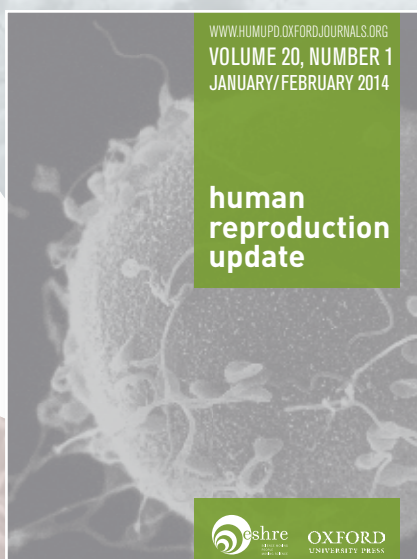


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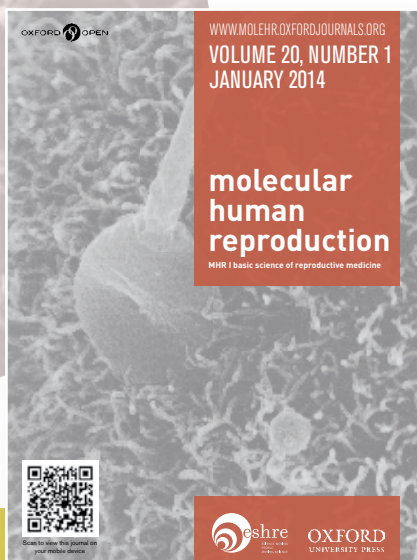
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MSD has a long-term commitment to patient centered IVF, with the introduction of Elonva (corifollitropin alfa), Puregon (recombinant FSH follitropin beta), Orgalutran (ganirelix), Pregnyl (Chorionic Gonadotropin). MSD also supports the development of assisted reproductive technology through its ongoing support of FSA.

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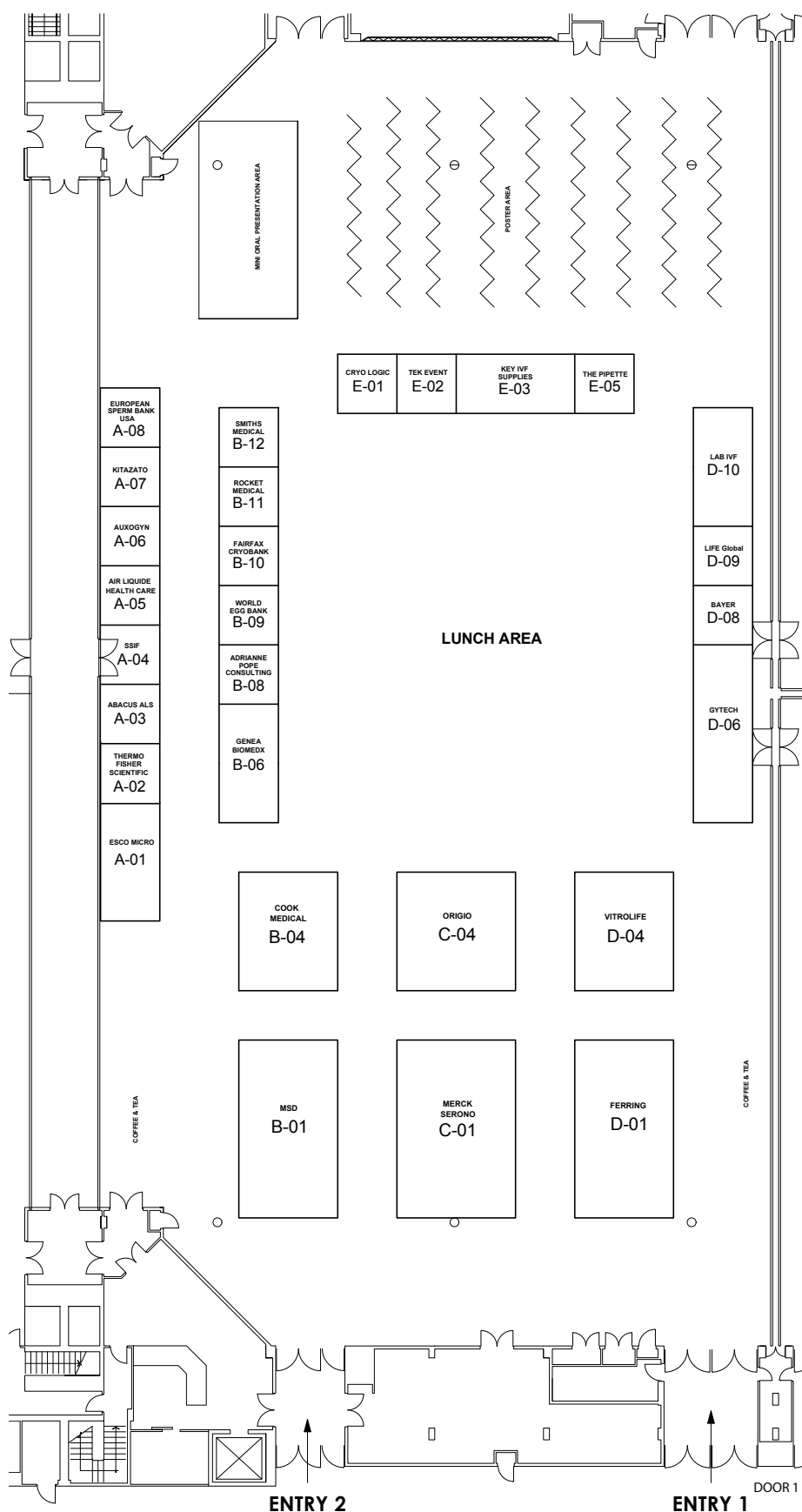
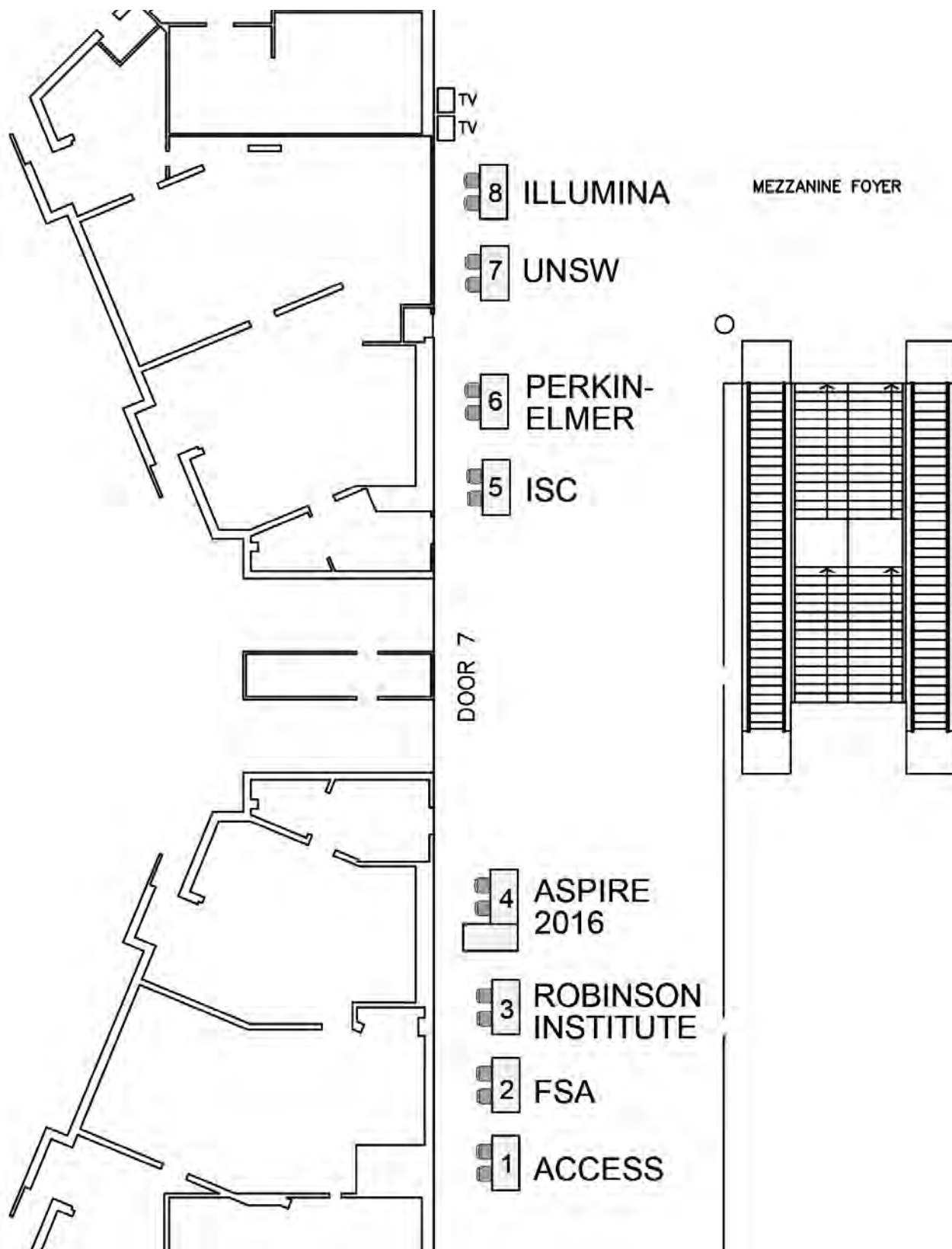


TABLE TOP EXHIBITION FLOORPLAN



EXHIBITORS' LISTING

EXHIBITORS	BOOTH
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AIR LIQUIDE HEALTHCARE	A-05
AUXOGYN	A-06
BAYER	D-08
COOK MEDICAL	B-04
CRYOLOGIC	E-01
ESCO MICRO PTE LTD	A-01
EUROPEAN SPERM BANK USA	A-08
FAIRFAX CRYOBANK	B-10
FERRING PHARMACEUTICALS	D-01
GENEA BIOMEDX	B-06
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LIFEGLOBAL GROUP/ IVFONLINE.COM, LLC	D-09
MERCK SERONO	C-01
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ORIGIO A/S	C-04
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SERONO SYMPOSIA INTERNATIONAL FOUNDATION	A-04
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VITROLIFE	D-04
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ILLUMINA	T-08
INTERNATIONAL STANDARDS CERTIFICATIONS (ISC)	T-05
PERKINELMER	T-06
THE UNIVERSITY OF ADELAIDE AND THE ROBINSON INSTITUTE	T-03
UNSW AUSTRALIA	T-07

EXHIBITORS' PROFILES

ABACUS ALS

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Email: p.hawkins@abacus-als.com

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Abacus ALS will be displaying the ASTEC incubators and Nikon microscopy products on our booth. A representative from ASTEC will be available to discuss the reasons behind the popularity of their incubators for IVF applications.

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Contact: Sandra K Dill

Email: access@access.org.au

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Email: sgowan@illumina.com

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Contact: Maki OGAWA

Email: trading@kitazato.co.jp for company / ogawa@kitazato.co.jp for private contact

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Fax: +65-6896 7096

Contact: See Huilin

Email: sales@labivf.com

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STAND: T-07

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