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PROGRAMME BOOK

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DETECTION OF DNA METHYLATION OF DISC1 GENE USING METHYLITYL TAQMAN® ASSAY

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Introduction
Genetic functional studies have shown contributions of DISC1 gene in the pathogenesis of Schizophrenia [1]. However, the basis of the genetic defect is not yet established [2]. There has been a shift of emphasis from DISC1 gene variations to other genetic defects such as copy number and epigenetic [3]. Epigenetic of DISC1 has not been well studied [4].

Objective
To explore the DNA methylation status of DISC1 gene in patients with Schizophrenia.

Materials & Methods

Subjects
This is a case-control study design. Based on data from Carrard et al. (2011) [5], 122 blood samples (1:1 case-control ratio) were needed to give a 95% two-sided confidence level and an 80% power of detection (effect size 0.30).

239 Malay subjects (117 Schizophrenia patients; 122 healthy controls) were recruited from the Psychiatry Clinic, Hospital Tengku Ampuan Afzan, Kuantan, Pahang.

Inclusion criteria:
1. DSM-IV diagnosis of schizophrenia of at least six months duration.
2. Symptoms of psychosis not secondary to substance use or neurological disorders.

Exclusion criteria: Mentally retarded patients and age below 18 years old. Biodata and clinical data were gathered from interview and medical records. Clinical symptoms were assessed using the Positive and Negative Syndrome Scale (PANSS).

The study protocol was ethically approved by IIUM Research Ethics Committee (IREC PANSS).

The statistical tests used to the variants of the data were: Independent sample t-test, one way-analysis of variance (ANOVA), and bivariate correlation. p-value of <0.05 were considered as significant.

Results
Successful amplification curve recorded for DISC1 was limited to 1:729 of DNA serial dilution, and β-actin at 1:6501. Standard curve of DISC1 assay showed 101.2% efficiency with R²=0.999 (Figure 1) while for β-actin, the assay showed 101.8% efficiency with R²=0.762 (Figure 2). Both assays were sensitive and specific at the DNA methylated region.

There was no significant differences p-value >0.05 in the methylation level of DISC1 between Schizophrenia patients and healthy controls (Table 1).

There was no correlation between DNA methylation level of DISC1 and age (R²=0.011, p =0.454 ), age of onset (R²=0.017, p=0.152), duration of illness (R²=0.006, p=0.680), and PANSS sub domains (positive symptoms: R²=0.025, p=0.224; negative symptoms: R²=0.014, p=0.296; disorganization: R²=0.019, p=0.851; excitement: R²=0.005, p=0.242; emotional distress: R²=0.024, p=0.950) in Schizophrenia.

There was no significant difference in DISC1 gene methylation for smoking status and the types of antipsychotic drugs received using independent sample t-test.

One way-ANOVA was conducted to explore the impact of DISC1 methylation level in Schizophrenia on BMI status and there was no significant difference for each BMI group.

Conclusions
Study on epigenetic of Schizophrenia, DISC1 gene is still lacking. Although DISC1 gene has been one of the positive candidate genes for the pathogenesis of Schizophrenia since 1990 [7], i.e. via gene variations [8], our study suggested that the DNA methylation of DISC1 gene is most likely not the genetic basis of Schizophrenia.

References