Hyaluronate lyase activity in Malaysian isolates of *Streptococcus pneumoniae*

Yusof AH[1,2], Desa MNM[3], Masri SN[1], Malina O[1], *Jamal F[1]
[1]Department of Medical Microbiology and Parasitology, Faculty of Medicine and Health Sciences, Universiti Putra Malaysia, Selangor, Malaysia
[2]Department of Biomedical Science, Kulliyyah of Allied Health Sciences, International Islamic University Malaysia, Pahang, Malaysia
[3]Department of Biomedical Science, Faculty of Medicine and Health Sciences, Universiti Putra Malaysia, Selangor, Malaysia

Abstract

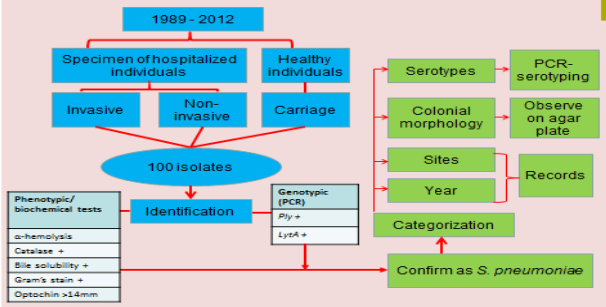
Streptococcus pneumoniae is an important pathogen cause various severe illness in human particularly pneumonia, septicaemia and meningitis. One of its virulence factors is hyaluronate lyase, an enzyme which degrades hyaluronic acid (HA), a major component in extracellular matrix of human connective tissues. Degradation of HA decrease host tissue viscosity and eventually increase tissue permeability to allow migration of pneumococcus or its product into human tissues. The study attempted to screen production of hyaluronate lyase in 100 isolates of *S. pneumoniae* from various sites and year of isolation, and other bacterial characteristics. Ninety-six isolates (96%) showed positive hyaluronate lyase through hyaluronic acid diffusion agar method. Few positive hyaluronate lyase isolates were selected and proceed to enzymatic activity determination through hyaluronic acid turbidity reduction assay. The study found that non-vaccine serotypes and carriage isolates showed high hyaluronate lyase activity than invasive isolates. This finding suggest that hyaluronate lyase in *S. pneumoniae* might play more important role in pneumococcal colonization than invasion

1 Introduction

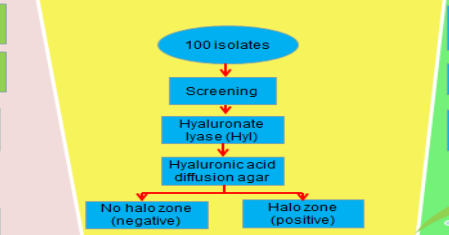
Hyaluronate lyase produced by wide range of microbes and recognized as spreading factor due to its capability to degrade hyaluronic acid in human connective tissue (Duran-Reynalds, 1933). Degraded hyaluronic acid decrease tissue viscosity and increase its permeability which allow bacteria to enter host body through broken tissue. Streptococcus pneumoniae is one of Gram positive bacteria producing this enzyme, but not all pneumococcal strains. Factors that might influence the production of this enzyme is currently unclear. Current study attempted to screen and determine activity of hyaluronate lyase in pneumococcal isolates with different characteristics including site and year of isolation, colonial morphology and serotypes.

2 Methodology

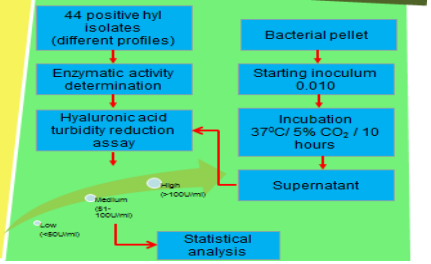
Bacterial identification & characterization



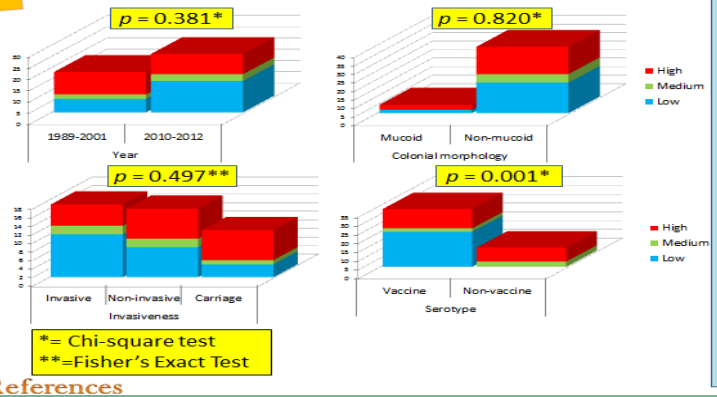
Hyaluronate lyase screening



Enzymatic activity determination



3 Results & Discussions



- High number of isolates (96%) showed hyaluronate lyase (Hyl) production. 100% carriage isolates produces the enzyme. This finding in agreement with Marion *et al.* (2012), but contrast to Kostyukova *et al.* (1995) which found high and low Hyl production in carriage isolates, respectively. Finding of high production of Hyl among carriage suggest role of this enzyme for pneumococcal colonization. In respiratory tract, there are low level of carbohydrate sources. Hyaluronic acid (HA) which abundantly present in this niche triggered activity of Hyl to degrade HA and serve carbon sources from end products of degraded HA.
- Enzymatic activity determination showed most of carriage and non-vaccine serotypes isolates produced high activity (>100 U/mL). High Hyl activity detected among isolates from respiratory-sites, while low activity detected among isolates from blood-origin sites. This suggest that activity of Hyl might be regulated by presence of its substrate, hyaluronic acid in the environment. Polissi *et al.* (1998) found association of Hyl in pneumonia, not in septicaemia. Hyl might facilitate bacterial residing in lung tissues rather than bacterial surviving in blood stream

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Acknowledgements

We would like to thank all staff and students, Microbiology Lab, Faculty of Medicine and Health Sciences , UPM for their help . This study was supported by Research University Grant Scheme (RUGS) (No. Grant: 04-02-11-1376RU) and Fundamental Research Grant Scheme (FRGS) (No. Grant: 01-03-11-987FR), and International Islamic University Malaysia for sponsoring study fees.