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Title: Antimicrobial susceptibility and genotypic characterization of clinical Salmonella enteritidis strains isolated from a tertiary hospital in Malaysia by using multilocus variable number of tandem repeat analysis and pulsed-field gel electrophoresis

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Abstract: Background: Salmonella enterica serovar enteritidis (S. enteritidis) causes non-typhoidal salmonellosis (NTS) in humans. Twenty-eight percent of NTS serovars identified and reported to the laboratory based surveillance database of Malaysian Ministry of Health in 2005 was S. enteritidis. The increasing occurrence of multidrug resistant (MDR) S. enteritidis complicates available therapeutic options. Phage typing and pulsed-field gel electrophoresis (PFGE) are commonly used subtyping methods, but there are limitations. The advent of multi-locus variable number of tandem repeats analysis (MLVA) provided a better discrimination of S. enteritidis. This study aimed to determine the antibiograms and genotypes of clinical S. enteritidis strains isolated from a tertiary hospital in Penang, Malaysia.

Methods: A retrospective study involving 16 clinical S. enteritidis strains isolated from 2005 to 2006 was conducted. The resistance of the strains against 14 antimicrobial drugs was examined, and the donality of the strains was determined by both MLVA and PFGE of XbaI digested bacterial chromosomal DNA.

Results: Both invasive (n=9) and non-invasive (n=7) S. enteritidis were examined and a high percentage of multidrug resistance was observed (66% of invasive and 43% of non-invasive strains). MLVA (D=0.77) yielded five distinct types (M1 to M5), which correlated to antimicrobial resistance patterns of the strains. Strains of MLVA type M1 were resistant to tetracycline, type M2 resistant to nalidixic acid, types M3 and M4 resistant to both ampicillin and nalidixic acid, and type M5 resistant to sulfonamides, trimethoprim, trimethoprim-sulfamethoxazole and tetracycline. The VNTR loci were genetically homogeneous (Nei's diversity index ≤ 0.53) among the strains, PFGE (D=0.94) subtyped all strains into 11 pulsortypes, showing high genetic homogeneity (0.84<F<1.00) among the strains. There was a low concordance of the clustering of the strains based on PFGE and MLVA (Adjusted Rand value=0.023). MLVA provided a characteristic strain clustering according to resistance patterns but PFGE failed to do so. The invasive and non-invasive strains were not distinguished by both methods.

Conclusion: The occurrence of high percentage of MDR S. enteritidis strains is of public health concern. However, the ability of MLVA to rapidly subtype S. enteritidis strains into groups with characteristic antimicrobial resistance patterns may be of great advantage in clinical diagnostic.