

APPLICATION OF RIBOSOMAL RNA GENE RESTRICTION PATTERNS ANALYSIS AND PULSED-FIELD GEL ELECTROPHORESIS IN DISTINGUISHING *SALMONELLA WELTEVREDEN* ISOLATES IN MALAYSIA

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Abstract. A representative sample of 20 isolates of *Salmonella weltevreden* strains from stool cultures of patients admitted at the University Hospital, Kuala Lumpur, Malaysia were analyzed. All the strains were susceptible to ampicillin, ceftriaxone, ciprofloxacin, chloramphenicol, tetracycline, trimethoprim, gentamicin and co-trimoxazole. Ribosomal RNA gene restriction pattern analysis of *Pst*I-digested DNA gave three ribotypes while pulsed-field gel electrophoresis (PFGE) analysis of *Xba*I-digested DNA gave ten distinct profiles. PFGE was more discriminative than ribotyping in distinguishing the strains. The majority of the strains analyzed were very closely related with similarity coefficient values ranging from 0.8 to 1.0. Both PFGE and ribotyping could distinguish one of the strains which was obtained from a patient following a bone marrow transplant for β -thalassemia major, indicating that this particular strain was unrelated to the rest of the strains from patients with acute gastroenteritis.

INTRODUCTION

Acute gastroenteritis caused by *Salmonella* spp continues to be a public health problem in many parts of the world especially in the developing countries, including Malaysia. The most common agent associated with non-typhoidal salmonellosis is *Salmonella enteritidis*. However, outbreaks of gastroenteritis caused by *Salmonella weltevreden* are rarely reported. In terms of its distribution, *S. weltevreden* was the fourth commonest serotype isolated (17/173 or 9.8%) from the University Hospital, Kuala Lumpur (Lee *et al.*, 1998). In another survey, Koe *et al.* (1991) in their study of 97 children with acute gastroenteritis, showed that the most common pathogen isolated was food poisoning *Salmonella* spp (25.8 %) and among these strains, 15% (3/20) were *S. weltevreden*. *S. weltevreden* was the second most common serotype isolated from clinical specimens in Malaysia (*ie* 326/1154) (IMR Annual Report, 1993). In addition, *S. weltevreden* also has one of the widest zoological distributions, which include cattle, beef, mutton, duck, prawn, dog, monkey and rats. In the past decade, molecular-based methods have been widely applied to study

the genetic relatedness of pathogens to track the source of infections, and to investigate the extent of genetic variations among isolates belonging to the same clone. As there are no published reports for this species, the objective of the work was to apply the technique of ribotyping and pulsed-field gel electrophoresis to study the extent of genetic variation of clinical strains of *S. weltevreden* from sporadic cases of acute gastro-enteritis in Malaysia.

MATERIALS AND METHODS

Bacterial strains

A total of 20 clinical isolates of *S. weltevreden* from feces or rectal swabs obtained from different individuals with sporadic cases of gastroenteritis admitted to the University Hospital, Kuala Lumpur were used in this study. The organisms were isolated, maintained and identified using standard methods at the Medical Microbiology Laboratory, University Malaya Medical Center. All isolates were tested for antibiotic sensitivity by standard disc diffusion procedures.

Preparation of DNA

Chromosomal DNA for pulsed-field gel electrophoresis (PFGE) analysis and ribotyping was prepared in agarose plugs as previously described (Thong and Pang, 1996). Very briefly, pelleted cells

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