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Genotypic and phenotypic characterization of *Escherichia coli* isolated from indigenous individuals in Malaysia

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ABSTRACT

Objective(s): The occurrence of asymptomatic verocytotoxin (VT)-producing *Escherichia coli* (VTEC) infections among humans in recent years is posing a high risk to public health. Thus, the role of asymptomatic human carriers as a source of dissemination should not be underestimated. This study aimed to elucidate the phenotypic and genotypic characteristics of *E. coli* in the stool samples collected from indigenous individuals in Malaysia.

Materials and Methods: *E. coli* strains (n=108) were isolated from stool samples obtained from 41 indigenous individuals. All strains were subjected to Repetitive Extragenic Palindromic-Polymerase Chain Reaction (REP-PCR) typing and confirmation of VTEC variants. Non-duplicate strains were selected based on REP-PCR profiles and further subjected to antimicrobial susceptibility test (AST). The genotypic and phenotypic characteristics of the strains were then correlated with the demographic data of the subjects.

Results: A total of 66 REP-PCR profiles grouped in 53 clusters (F=85%) were obtained. Four genetically distinct strains were confirmed as VTEC (*eaeA*-positive). The predominant resistance was against ampicillin (34.2%), followed by trimethoprim-sulfamethoxazole (32.9%), ampicillin-sulbactam (5.5%), and ciprofloxacin (1.4%). All isolates were sensitive to amoxicillin-clavulanate, cefuroxime, ceftriaxone, imipenem, and meropenem.

Conclusion: Genetically diverse *E. coli* and VTEC strains were found to colonize the intestines of the indigenous populations. This study is important for the prospective surveillance of *E. coli* among the indigenous individuals in Malaysia, especially in asymptomatic VTEC infection and antimicrobial resistance phenomenon.

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Introduction

Escherichia coli (*E. coli*) is one of the commensal bacterial species that are found in human and animal intestines and feces. These commensal *E. coli* strains rarely cause disease, except in immunocompromised hosts, or when gastrointestinal barriers are violated. However, only certain serotypes of *E. coli* may cause disease in both humans and animals (1).

Enterohaemorrhagic *E. coli* (EHEC), also known as Shiga-toxin producing *E. coli* (STEC), was first described as associated with pathogenicity in 1982 (1). It is an important foodborne pathogen that causes serious public health concerns as it is highly associated with severe gastrointestinal disease (1). Characteristics of EHEC infection include abdominal cramps, bloody diarrhea, as well as hemorrhagic colitis and hemolytic uremic syndrome (2). The major virulence factor of EHEC is Shiga toxins (Stx) (1, 2). The Stx family is composed of 2 major subgroups, namely Stx1 and Stx2. The organisms may express at least one subgroup of Stx. Shiga toxin was reported to be the same as verocytotoxin (VT) (3). Thus, the term can be fully interchangeable with VT nomenclatures (*i.e.*, Stx1 = VT1,

Stx2 = VT2).

EHEC can also be defined by the presence of intimin, encoded by the eaeA gene (2, 4). This protein allows EHEC to intimately attach to epithelial cells and efface microvilli in the large intestine. Strains that specifically encode the eaeA gene appear to colonize any intestinal site, followed by the presence of 'attaching and effacing' (A/E) lesions (2). The lesion in EHEC infection is characterized by destruction of microvilli, intimate attachment of the organism to the cell, and accumulation of polymerized actin beneath the site of bacterial attachment. Previous studies have shown that even if that strain of E. coli may produce Stx, it can be eaeAnegative and not associated with any disease (5-7). This has led to the use of VT-producing E. coli (VTEC) or STEC as a general term to name those strains that produce Stx, and the term EHEC is used only for the one that produces Stx together with the eaeA-positive genotype.

In Malaysia, the native indigenous populations generally live in poor conditions, low hygiene practices, and lack of functioning toilet facilities in their houses. These factors were found to be significantly related to microbiota diversity among them compared with the major ethnic groups

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