

GENOTYPING OF ENTEROTOXIGENIC *E. COLI* COLONIZATION FACTORS BY AFLP FINGERPRINTING (POSTER)

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Background: Enterotoxigenic *E.coli* (ETEC) is the major causative agent of diarrheal disease in young children and travelers in Thailand. ETEC colonize intestinal mucosa by means of colonization factors and the production of enterotoxins (LT and/or ST). Many types of colonization factor antigens (CFA) and putative colonization factors (PCF) have been identified throughout the world. However, only 36% of ETEC strains isolated in Thailand could be categorized by immunological method. This is probably due to either loss of antigenicity during storage or the presence of new variants. In this study, AFLP fingerprinting method was tested for its ability to demonstrate the relationship between different CFA types of ETEC and to categorize the group of untypeable strains.

Methods: Genomic DNA was extracted from ETEC strains with known and unknown CFA using phenol-chloroform method. The DNA was subsequently subjected to restriction (*MseI* and *EcoRI*)-ligation, preselective, selective amplification and electrophoresis on ABI310 Genetic Analyzer. Similarity values were computed using Dice's coefficient and Cluster analysis was carried out by UPGMA.

Results: Initially, two combinations of AFLP modules generating informative bands correlated with CFA types were obtained from 14 known strains. Fifty-nine known and 24 unknown strains were then investigated for DNA fingerprint profiles. Cluster analysis showed 5 major clusters with the percent similarity ranging from 59 to 75. Distinct CFA types were grouped into distinct clusters of dendrogram. However, CS 2 was present in the same cluster as CS2+3 and shared 82% similarity with CS1+3 suggesting very close related strains. Half of the unknown CFA strains could be grouped into particular known CFA types, while the rest belonged to distinct clusters suggesting new variants.

Conclusion: Besides immunological typing method that relies on the stability of antigens, the AFLP fingerprint technique provided a sufficient discriminatory power and high reproducibility within the group of closely related strains of *E. coli*.

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