

Detection of Virulence Genes and Class 1 Integron Associated Gene Cassettes in *Vibrio Cholerae* Isolated from Ice of Streetfoods in Jakarta, Indonesia

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Vibrio cholerae is a noninvasive Gram negative bacterium that lives naturally in the aquatic environment. Ice used in beverages could be one of the sources of *V. cholerae* transmission. Almost all of the beverages are served with ice cubes in Jakarta. But there is no regulation to control the quality of ice. Street food is considered as one of the potential major public risks in Jakarta.

The isolates were collected from ice used for street foods in Jakarta. In this study, we isolated *V. cholerae* using selective medium and continued with biochemical assays (KIA, lysine decarboxylase, and indole). Serological tests were done using polyvalent antiserum to determine O1 or non-O1 and monovalent antisera to determine Ogawa or Inaba. Hexaplex PCR is used for rapid detection of virulence genes such as *ctxA*, *ompU*, *tcpA*, *ace*, *zot*, and *toxR*. The hemolysis assay were done by using Brain Heart Infusion medium. Antibiotic resistance test were examined by using disc diffusion test for ampicillin (10µg), streptomycin (10µg), kanamycin (30µg), trimethoprim (5µg), tetracycline (30µg), ciprofloxacin (5µg), erythromycin (15µg) and sulfamethoxazole (25µg). All of the samples were screened for the presence of class 1 integron by PCR using specific primers 5'CS and 3'CS.

From six hundred and seventy one presumptive *V. cholerae* isolates, we found one hundred and eight (16.10%) were positive *V. cholerae*. We recovered eighty four isolates of *V. cholerae* classified as non-O1 and twenty four isolates were O1. From O1 serotype, twenty isolates were classified as Ogawa and four isolates were Inaba.

Based on the results from the hexaplex PCR, we found eighty samples (74.07%) gave positive results for *toxR*, twenty five samples (23.15%) for *ctx*, fifteen samples (13.88%) for *ompU*, one samples (0.93%) for *zot* and all samples gave negative results for the *tcp* and *ace* genes.

Ninety isolates (74.07%) were positive in hemolysis assays. From the antibiotic resistance assays, 70.37% isolates were resistance to ampicillin, 19.45% isolates to tetracycline, 61.11% isolates to streptomycin, 51.85% isolates to kanamycin, 2.77% isolates to ciprofloxacin, 37.97% isolates to erythromycin and 42.59% isolates were resistance to sulfamethoxazole-trimethoprim.

From the detection of class 1 integron by PCR using primer 5'CS and 3'CS. 42.59% were positive for class 1 integron.

A lot of ice in Jakarta was contaminated with *V. cholerae*. Some of the isolates were classified as toxigenic and others were pathogenic strains. The isolates which do not have the virulence genes detected in this study might have different virulence mechanism. A majority of the isolates were resistant to more than one antibiotics, most of them are correlated with class 1 integron. Finding of antibiotics-resistant *V. cholerae* in this study need to be aware of due to their correlation with integron as one of the resistance dissemination factors.

Keywords: *V. cholerae*, ice, class 1 integron, hexaplex PCR, virulence genes, Jakarta