

Occurrence and distribution of *Clostridium perfringens* in relation to conventional faecal indicator bacterial in Shrimp farm

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Biomonitoring indicates the microbiological quality of water. Though *E. coli* is a microbiological indicator, it cannot be considered as a perfect one. In this regard, attempts for finding a new indicator of faecal pollution in place of *E. coli* was carried out by performing a comparative study between *E. coli* and *Clostridium perfringens* as faecal indicator in shrimp farms. The presence of these organisms was determined by evaluating the brackish water and shrimp samples from various areas of Cochin, using MPN technique. The effect of disinfectant chlorine was examined over these organisms which revealed the sensitive nature of *E. coli*. *Clostridium perfringens* was found to be highly resistant in comparison with *E. coli* and thus suggesting it as a better indicator of faecal contamination in brackish water to evaluate the microbial quality of shrimp farms than *E. coli*.

Key words : Biomonitor, *E. coli*, *Clostridium perfringens*, Brackish water, Shrimp, MPN technique, Chlorine.

INTRODUCTION

Aquaculture has recently emerged as an alternative to the decline in marine resources. Shrimp, being the most widely cultured variety in India, plays an important role in the growth of seafood exports. The single most significant factor causing major decline in the population of many fish species is pollution.

Pathogens are always present in the aquatic environment. The main concern therefore, has been in introducing new disease organisms or causing environment deterioration that can result in increased population of indigenous pathogens (Reid *et al.*, 1976).

Biomonitoring is used to indicate the microbiological quality of water. Traditionally *E. coli* has been the chosen indicator of faecal pollution due to its occurrence in large numbers than other organisms. But *Clostridium perfringens* was found to persist for relatively longer time in the environment compared to coliforms (Herbett, 1992). As *E. coli* cannot be considered a perfect indicator, an investigatory attempt was done for searching a new indicator of faecal contamination to substitute *E. coli* by comparing *E. coli* and *Clostridium perfringens* as faecal indicators in shrimp farms.

MATERIALS AND METHODS

Sample preparation and processing:

Water and shrimp samples were collected from two

brackish water farms, namely Chellanum and Vaippin in and around the areas of Cochin. Shrimp sample collected include *Penaeus monodon* and *Penaeus indicus*. The sample preparation involved bringing shrimps in a sterile polythene bag and further the whole part being into small pieces under aseptic condition using scalpel. 10gm of sample was then homogenated in 100 ml of sterile physiological saline (0.85% NaCl). For processing the sample, 5 tube and 3 tube (MPN) method was followed to enumerate the amount of *E. coli* and *Clostridium perfringens* in the given sample.

Coliform isolation:

Isolation of *E. coli* was carried out using the presumptive, confirmatory and completed test 3 tube and 5 tube method. Dilutions were done for fish and water sample by inoculating MacConkey broth, with 10ml, 1ml, 0.1ml aliquots of homogenates and 10ml, 1ml, 0.1ml, 0.01ml of water sample, respectively and incubating the tubes at 37°C for 24 hours. Positive tubes were confirmed by inoculating into Brilliant green lactose bile broth and incubating at 37°C for 24 hours. The completed test involved transferring a loop full of inoculum from the positive tubes to E.C and tryptone broth and incubating at 46°C for 24 hours.

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