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1. Introduction

Vibrios, members of the genus *Vibrionaceae* are gram negative, usually motile rod shaped, mesophilic and chemoorganotrophic bacteria. They are found in aquatic habitats and in association with eukaryotes (Thompson et al., 2004). Fish vibriosis is a disease of marine and brackish water finfish. The severity of vibriosis has increased with the worldwide expansion of fish farming. Vibriosis is the most common disease affecting marine finfish grow-out system in the Asian region, leading to catastrophic economic losses. The most commonly encountered fish pathogenic *Vibrio* species is *Vibrio anguillarum*. It constitutes the normal microflora of the aquatic environment and proportionally increases in the summer season. Consequently, the fish may be continually threatened by the pathogen which may be able to cause disease under certain conditions (Bullock, 1987; Austin and Austin, 1999).

Prior to the introducing of immunization, the most common method employed to control vibriosis was treatment of diseased fish with antibiotics and antimicrobial chemicals. The commonly used antibiotics were tetracycline, sulphonamides, nitrofuran, and quinolines. However, it is now considered ineffective as serious drug resistance have been reported to the most common antibiotics used in aquaculture farms (Aoki et al., 1981). Further, there is a risk of transfer of antibiotic resistance factors to other bacteria including human pathogens. It has been observed that R-plasmid carried by *V. anguillarum* is transferred even between different species. Beside the problem in treatment of vibriosis by antibiotics, the use of antimicrobial chemicals like copper compounds causes harm to the fish by accumulating in the tissues (Aoki et al., 1974).

Therefore, immunization is considered as the best strategy to protect fishes and aquatic animals from vibriosis. Several studies have shown protective effect of different vaccine formulae such as formalin killed or heat inactivated bacteria against *Vibrio* infection. However,
no effective and safe killed vaccine is available for these infections. Attenuated live vaccines stimulate an effective immune response but they may represent a risk by reverting to virulence (Ellis, 1988).

Recombinant techniques can be used to produce high amounts of recombinant antigen. This antigen can be administrated to the host by injection, or by immersion. The advantage of recombinant vaccine is that there is no chance of host being threatened by vaccine material. Moreover, each batch of vaccine has the same potency and is more stable during the long term storage. The technology can also be used to produce the antigenic epitopes which stimulate T-suppressor cells (Munn, 1994; Lorenzen, 1999).

The outer membrane proteins (OMPs) of Gram-negative pathogenic bacteria posses the vaccine characters, since they have important role in the interaction with hosts in adherence, uptake of nutrients from the host and eliminating host-defense mechanisms during bacterial pathogenicity. Also it has protective antigenicity, because the components of the outer membrane are easily recognized as foreign substances by immunological defense systems of the hosts (Maiti et al., 2009).

The OMPs can induce specific bactericidal antibodies, inhibit the bacterial colonization in hosts and induce cell-mediated immunity (Bricknell et al., 1999). OMPs are located in the outer membrane so that it affects surface structures or surface charges that may have important role in colonization of the bacteria. In contrast to most of bacterial invasion factors, the non enzymatic OMPs are considered as non toxic molecules. Therefore they are safe for animal administration (Seltman and Holst, 2002).

Against this background this study was aimed to develop a recombinant protein vaccine against *V. anguillarum* by targeting OMPs. The study was taken up with the following objectives:
Selection of suitable antigens that might be having immunogenic characteristics based on the genome sequences of *Vibrio* and bioinformatics tools.

Cloning and expression of the genes coding for the target antigens.

Immune characterization of the recombinant proteins.

Evaluation of protection level offered by the recombinant immunogenic antigens.

2. Literature review