9.0. SUMMARY AND CONCLUSION

The results of the present study are summarized as follows:

- A total of 148 total heterotrophic bacteria (THB) strains and 21 actinomycetes (ACT) strains were isolated from 36 marine sponge samples. The maximum counts of THB was found in November (monsoon season) and actinomycetes in May (summer season).

- Morphologically different five bacterial fish pathogens such as FPAU01, FPAU02, FPAU03, FPAU04 and FPAU05 were isolated from infected fish of *Mugil cephalus* and the pathogenecity was confirmed by the re infection studies.

- Of the sponge associated microbial strains, the THB-131 and ACT-21 showed potential antibacterial activity against 5 bacterial fish pathogens and the results of the MIC values revealed that, THB-131 and ACT-21 showed varied MIC and MBC values ranged from 125-1500µg.ml⁻¹, which depends upon the bacterial fish pathogens.
Molecular characterization fish pathogens reveals that, the sequence of the FPAU01 (680 nucleotides) showed maximum similarity (99%) with *Bacillus* sp. (GenBank Acc. No. HM179550), followed by FPAU02 (720 nucleotides) showed maximum similarity (99%) with *Bacillus* sp. (GenBank Acc. No. DQ079008), the FPAU03 (640 nucleotides) showed maximum similarity (100%) with *Bacillus cereus* (GenBank Acc. No. FR878075), the FPAU04 (730 nucleotides) showed maximum similarity (99%) with *Bacillus* sp. (GenBank Acc. No. JF935274) and FPAU05 (560 nucleotides) showed maximum similarity (94%) with *Bacillus* sp. (GenBank Acc. No. EU571145). But, the sponge associates of THB-131 (770 base pairs) showed maximum similarity (96%) with *Bacillus* sp., (GenBank Acc. No. FJ392727) (Fig. 16) and ACT-21 (240 base pairs) showed maximum similarity (99%) with *Streptomyces* sp.’ST-1 Sivasamy BU’ (GenBank Acc. No. GQ423725).

As the bacterial species are belongs to *Bacillus* genus, the present study also made an attempt to find out the any dissimilarity in the RNA secondary structure. It reveals that, the stem and loops in the secondary structure shows variations among the *Bacillus* species. Even
though the species are seems to be similar but there are some inherent variations in the genetic system.

- Immunological parameters such as glucose, cholesterol, bilirubin, SGOT, SGPT and ALP were found increased with the pathogen injected animals but, the extract of *Bacillus* sp. RPAUTHB-131 and *Streptomyces* sp. RPAUACT-21 treated animals were showed decreased level of glucose, cholesterol, bilirubin, SGOT, SGPT and ALP parameters and increased level of WBC, RBC and total protein hematocrit parameters.

- The present study was also made an attempt to find out the maximum average percentage protection of antibacterial activity (A) and the results revealed that, the maximum protection of antibacterial activity was identified against fish pathogenic *Bacillus cereus* with sponge associated *Streptomyces* sp. RPAUACT-21 extracts (76.29%) and fish pathogenic bacteria *Bacillus* sp. RPAUOCAS2 with sponge associated *Streptomyces* sp. RPAUACT-21 (71.84%). Moreover, minimum percentage was observed in fish pathogenic bacteria *Bacillus* sp. RPAUOCAS3 with the sponge associated *Bacillus* sp. RPAUTHB-131 extract (44.25%).
It is concluded from the present study that, the sponge associates of *Bacillus* sp. RPAUTHB-131 and *Streptomyces* sp. RPAUACT-21 isolated from the Palk Strait region has potential antibacterial activity against bacterial fish pathogens and its opens a new way for the development of immunostimulants so as to enable to enhance the health of the fish and for the treatment of fish diseases. Further studies are highly warranted to develop a vaccine from the sponge associated microorganisms for the management of fish diseases in *M. cephalus*. 