#### **ECONOMIC EVALUATION OF MMC**

Parameters Cost	t / L (Rs/-)
Variable Cost	
Enrichment Media and Substrate	15
Other Miscellaneous Expenditure (Electricity, maintenance etc.)	25
Total Variable cost	40
Initial Investment	
Instruments and equipment for the development of consortium (Autoclave, Industrial centrifuge, Laminar airflow, Bacteriological Incubator, Refrigerator)	4,50,000
Life span	10 years
Consumables	6000
Life span	5 years
Fixed Cost	
Depreciation	30
Interest on Fixed Capital @7% per annum	25
Total Fixed cost	55
Total Cost of Production	95

#### **MMC Product Efficiency**

- Invivo and invitro studies indicate that the Laboratory-developed MMC exhibits high antagonistic activity against fish pathogens and 82.3% enhancement of gut probiotic microbiota.
- The study also indicates higher growth rate (31.13%), yield (119.16%) and net profit (188.9%) than control with B:C ratio of 3.42.
- ➤ The consortium is highly bactericidal, bacteriostatic, and hydrophobic in nature. It improves fish gut health, disease resistance, survival, growth rate, and production of Indian pompano. The optimized concentration of the MMC product was 1.8 x 10<sup>15</sup> cfu/ml.
- ➤ CMFRI-MMC, with its high vibriocidal activity and economic viability, is the best feed supplement to enhance the growth rate, survival, and production of the Indian pompano.



## Product specifications Ingredients:

Bacillus sp., Shewanella sp., Pseudoalteromonas sp.,

Halomonas sp.

Concentration:

1.8 x 1015 cfu/ml

Recommended storage: Refrigeration at 4°C Cell viability: 90% viable cells after 3 months

#### Published by

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# ICAR - CMFRI - MMC PRODUCT: AN INDIGENOUS MARINE BENEFICIAL MICROBIAL CONSORTIUM AS A POTENTIAL SUPPLEMENTARY DIET IN MARICULTURE SYSTEMS



### VISAKHAPATNAM REGIONAL CENTRE ICAR-CENTRAL MARINE FISHERIES RESEARCH INSTITUTE

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#### **About Mariculture Systems in India**

Marine Aquaculture is gaining more attention in India with the transfer of several indigenous technologies such as marine fish breeding, cage culture, and Recirculating Aquaculture Systems (RAS) developed by ICAR-CMFRI. Expansion of marine fish farming and aquaculture production can meet the demand for large-scale production and supply of fish protein globally. Present and future challenges generated by disease outbreaks and antimicrobial resistance in aquaculture systems need to be addressed for the production of high-value fish protein.

#### **Need of Marine Probiotics in Mariculture**

The application of multi-strain probiotics as a supplementary diet for fish is a novel solution to reduce the usage of antibiotics and to prevent disease outbreaks in aquaculture. Although several commercial probiotics are available in aquaculture industry, the suitability of those probiotics for marine fish needs to be evaluated. The selection of suitable probiotics is the most crucial point in the development of a probiotic consortium, as the gut microbiota in each fish is different. Hence, isolation and production of beneficial bacteria from the fish gut as a probiotic supplementation to the same fish will give the best result for growth enhancement, better survival, and health of fish which further helps in attaining a sustainable production of mariculture.

#### About ICAR-CMFRI MMC

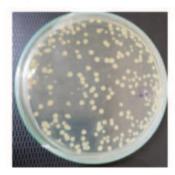
ICAR-CMFRI developed a potential Marine Beneficial Microbial Consortium (ICAR-CMFRI MMC) which improves the growth, survival, immunity, and gut health of Indian Pompano, *Trachinotus mookalee* 

#### Isolation of Marine Probiotics from Fish Gut

- Gut of Indian pompano, Trachinotus mookalee are collected from marine cages of ICAR-CMFRI, VRC
- The gut is processed for bacterial isolation, identification, and their antagonistic activity by spread plating of serially diluted samples on Zobell Marine Agar and incubated for 48 hrs
- Isolated colonies with typical characteristics are transferred to Marine broth for further analysis

#### **Identification of Probiotic Strains**

- 28 isolates exhibiting probiotic properties and antagonistic activity against pathogens are identified based on classical and molecular taxonomy (16S rRNA sequencing)
- Bacterial genera including 14.2% Gram-positive of three genera and 85.8% Gram-negative strains of 4 genera are identified





#### **Antagonistic Activity**

- The inhibitory effect of the gut isolates on selected fish pathogens are estimated using well diffusion and disc diffusion techniques and Minimum Inhibitory Concentrations (MIC) are estimated using broth dilution technique
- > Screened 250 isolates from fish gut out of which 11.2% of isolates exhibit antagonistic activity against fish pathogens *Vibrio harveyi, V. alginolyticus, V. neo-caledonicus*) isolated from infected Indian pompano.

#### Other Probiotic Properties

- Autoaggregation, coaggregation and cell hydrophobicity tests are performed for all the 28 isolates
- Sodium chloride, pH, temperature, acid and bile salt tolerance tests were conducted for all the 28 isolates, and found 10 isolates to exhibit a wide range of tolerance for the tests

#### Preparation of Marine Beneficial Microbial Consortium

- Co-culture of host-derived probiotic bacteria, belonging to four different genera, is a new approach to develop potential MMC.
- Marine Beneficial Microbial Consortia are prepared with different combinations of gut probiotic isolates that exhibit the maximum antagonistic and other probiotic characteristics
- MMC was prepared in simplified and cost-effective enrichment media and stored in at 4°C.



#### Advantages of MMC

High bactericidal, bacteriostatic and hydrophobic nature

- High inhibitory activity against fish pathogens
- Tolerance towards a wide range of pH, Sodium Chloride, Bile salt concentrations
- Improve survival percentage, weight gain, and biomass of cultured fish
- Improve gut health of cultured fish
- Eliminate Vibrios in the culture systems
- Enhances disease resistance in the fish