

Beyond Blue Horizons

An Experiential Learning Manual for
B.Sc. (Agri.) Students of KAU, Thrissur

Edited by

Vipinkumar V.P.

Jayasankar J.

Jenni B.

ICAR-Central Marine Fisheries Research Institute

(Department of Agricultural Research and Education, Government of India)

P.B. No. 1603, Ernakulam North P.O., Kochi - 682 018



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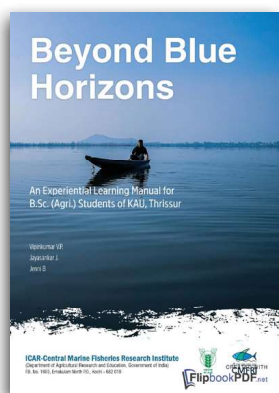


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Beyond Blue Horizons
Training Manual for BSc (Agri) students of Kerala Agricultural University

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FOREWORD

It is with great pleasure and deep satisfaction that I present this foreword to the Training Manual of the Science Camp titled “Beyond Blue Horizons: An Experiential Training Manual for B.Sc. Agriculture Students.” Conducted from July 14 to 18, 2025, at the STI Hub Digital Training Hall, ATIC, ICAR-CMFRI, Kochi, this programme exemplifies our continued commitment to innovative, experiential, and interdisciplinary learning in agriculture and allied sectors.

Organized by ICAR-Central Marine Fisheries Research Institute through its Agricultural Technology Information Centre (ATIC), the training served as a dynamic platform for B.Sc. Agriculture students from the College of Agriculture, Vellanikkara, Thrissur. The thematic focus on integrating field-based experiences with advanced laboratory analyses reflects a progressive approach to education—one that fosters a seamless continuum between knowledge generation, validation, and application. Such initiatives are vital in equipping students with the skills and perspectives required to address emerging challenges in agriculture and fisheries.

I place on record my sincere appreciation to Dr. Vipinkumar V.P., Principal Scientist and ATIC Manager, ICAR-CMFRI, for his exemplary leadership, meticulous planning, and unwavering dedication in organizing this programme. His efforts, along with those of the entire team, have ensured the successful conduct of this Science Camp, setting a high standard for future capacity-building initiatives.

The programme was thoughtfully designed to bridge the gap between theoretical understanding and practical application. It offered participants a rich blend of innovative lectures on emerging topics, hands-on training sessions, field exposure visits, and institutional interactions. The opportunity to access advanced laboratories, aquarium facilities, and the museum at CMFRI significantly enriched the learning experience. Equally important were the interactive sessions with farmers, which fostered meaningful exchanges between academia and practitioners, grounding scientific knowledge in real-world contexts.

Such experiential learning opportunities are invaluable in enabling students to appreciate the dynamic flow of information from field observations to laboratory insights, ultimately supporting informed decision-making and innovation in production systems. I am confident that the knowledge and exposure gained through this Science Camp will contribute significantly to the academic growth and professional development of the participants.

I extend my warm congratulations to all the students and faculty members who actively engaged in this programme. Your enthusiasm, curiosity, and commitment to learning are truly commendable. May this experience inspire you to strive for excellence and contribute meaningfully to the advancement of agriculture and fisheries.

I am confident that this training manual will serve as a lasting resource, capturing the essence of the programme and reflecting the collective efforts that made this initiative both impactful and memorable.



Dr. Grinson George
Director, ICAR-CMFRI
Kochi

PREFACE

It is with immense pleasure and a deep sense of fulfilment that I present this compendium, “Beyond Blue Horizons: A Training Manual for B.Sc. Agriculture Students of Kerala Agricultural University.” This volume encapsulates a unique and inspiring journey of experiential learning, meticulously designed and conducted at the STI Hub Digital Training Hall, ATIC, ICAR-CMFRI, Kochi, from July 14 to 18, 2025.

Envisioned as a transformative academic engagement, this Science Camp brought together bright and inquisitive B.Sc. Agriculture students from Kerala Agricultural University, Thrissur, and guided them through a rich continuum of learning—from field-level realities to the precision of laboratory analytics. At a time when agriculture is rapidly transitioning into a data-driven and innovation-led enterprise, the programme provided a vibrant platform for students to explore the convergence of traditional knowledge systems with modern scientific advancements, with a special emphasis on the fisheries sector.

The training was thoughtfully structured to deliver an immersive and practice-oriented learning experience. Through a dynamic blend of expert lectures, hands-on sessions, field exposure visits, institutional interactions, and meaningful dialogues with farmers, participants were encouraged not only to learn but to question, analyse, and innovate. The focus was on nurturing scientific curiosity, strengthening practical competencies, and inspiring a forward-looking approach to sustainable agriculture and fisheries development.

This manual, comprising ten thoughtfully curated chapters, reflects the thematic depth and diversity of the programme. It covers a wide spectrum of subjects including integrative analytics, digital interventions, field diagnostics, and emerging marine agri-technologies. What makes this volume particularly engaging is its strong practical orientation—each chapter offers insights, methodologies, and experiences that readers can readily connect with and apply. The concluding chapter, featuring the comprehensive report prepared by the students, stands as a testament to their active engagement and the effectiveness of the experiential learning model adopted during the camp.

I place on record my sincere gratitude to Dr. J. Jayasankar, Head of the FRAEE Division, and Dr. B. Jenni, ACTO, ATIC, for their scholarly contributions, editorial excellence, and steadfast support as co-editors of this compendium. Their efforts have been instrumental in shaping this manual into a valuable and enduring academic resource.

As the Course Director and Chief Editor, I consider this compendium not merely as a documentation of an event, but as a celebration of collaborative learning and an invitation to explore the vast and promising interface between agriculture and fisheries sciences. While this endeavour represents only a beginning—a glimpse into a much larger horizon—it is my earnest hope that this volume will inspire readers to delve deeper, think innovatively, and contribute meaningfully to this evolving domain.

I warmly invite students, researchers, academicians, and practitioners to engage with the chapters that follow—rich in practical insights, field-based observations, and scientific perspectives—and to draw inspiration for future learning and innovation.



A handwritten signature in black ink, appearing to read 'Vipinkumar V. P.', written over a light blue background.

Dr. Vipinkumar V. P.
Principal Scientist & ATIC Manager
ICAR-CMFRI, Kochi

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Disease diagnosis in marine fish and shellfish

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Sumithra T.G. and Krupesha Sharma S.R.

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Introduction

The global human population is anticipated to surpass 9 billion by 2050 and 11 billion by 2100 (UN, 2017), placing immense pressure on meeting growing food demands. In response to the rising need for marine protein, marine aquaculture presents a promising avenue for increasing seafood production. Mariculture, the farming of marine species, has emerged as a key sector for enhancing and diversifying global food systems, with ~249 species currently under cultivation (Hedge et al., 2023). Recognised as a major sub-sector of the aquaculture industry, mariculture contributes significantly to sustainable food production and supports the economic development of coastal communities. Presently, commercial mariculture operations are active in 102 countries across all continents except Antarctica. Over the past 30 years, mariculture production has grown nearly five-fold (Gentry et al., 2023). However, production remains concentrated in a few nations, with China alone accounting for over one-third of global output. Between 2000 and 2018, mariculture expanded at a compound annual growth rate (CAGR) of ~5.2%, compared to the CAGR of ~5.6% for the overall aquaculture sector (World Aquaculture, 2022). In 2020, global aquatic animal production reached an estimated 178 million tonnes, of which 63% (112 million tonnes) was sourced from marine environments, with 70% from capture fisheries and 30% from aquaculture (The State of World Fisheries and Aquaculture, 2022).

Diseases in mariculture

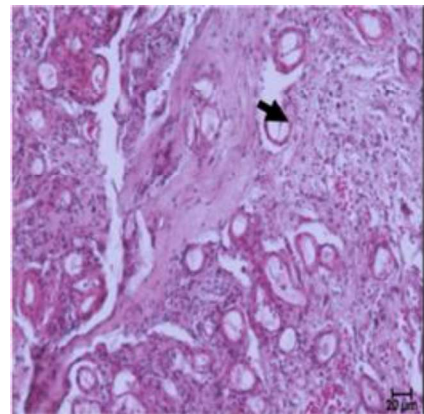
Marine fish health plays a pivotal role in the sustainability of global aquaculture operations. Infectious and non-infectious diseases can significantly affect fish populations, resulting in economic losses, reduced productivity, and disruption of ecological balance. The increasingly intense fish farming practices, however, have led to the emergence and re-emergence of various infectious diseases among the cultured animals. Noninfectious diseases usually arise from environmental stressors (poor water quality, ammonia/nitrite toxicity, low oxygen, changes in water physio-chemical characteristics), nutritional stressors like nutritional deficiencies, presence of toxins and pollutants, etc. Effective and accurate diagnosis and control measures are crucial for reducing the economic

losses caused by diseases in marine aquaculture and ensuring its sustainable development. Common pathogenic diseases in marine fish mainly include bacterial, viral, fungal, and parasitic diseases. Lafferty et al. (2015) estimated that bacteria, viruses, protists, and metazoans constitute about 25, 34, 19, and 18% of the total infectious agents causing diseases in marine aquaculture. The common bacterial diseases include vibriosis, streptococcosis, furunculosis, and photobacteriosis (Remuzgo-Martínez et al. 2014). In marine fish, the most critical etiologies for vibriosis are *V. parahaemolyticus*, *V. harveyi*, *V. alginolyticus*, *Vibrio anguillarum*, and *V. vulnificus*. Viral infections are also a significant concern in aquaculture and can cause high mortality rates in infected populations. Some common viral diseases affecting marine fish include *Red Sea Bream Iridoviral* disease, viral hemorrhagic septicemia, viral nervous necrosis, infectious pancreatic necrosis, infectious hematopoietic necrosis, and lymphocystis disease virus. Parasitic diseases cannot usually cause a direct loss due to fish mortality, but increase production costs through treatment or reduction in growth and the product quality. Many parasites affecting sea cage mariculture are ectoparasites. Amyloodiniosis caused by *Amyloodinium ocellatum*, a parasitic dinoflagellate, is a major threat to marine aquaculture, causing significant mortality in various fish species. *Ichthyophonus hoferi* is another fish pathogenic protist which causes a granulomatous systemic infection, called ichthyophoniasis, resulting in heavy mass mortalities of more than 80 freshwater and marine species, leading to huge economic losses. Fungal infections in aquaculture are less common than bacterial, viral, and parasitic diseases, but can still cause significant economic losses. Pathogens in the fungal diseases are divided into two groups. One of them is marine Oomycetes, which include members of the genera *Lagenidium*, *Haliphthoros*, *Halocrusticida*, *Halioticida*, *Atkinsiella*, and *Pythium*. The fungal diseases caused by mitosporic fungi in marine fish and shellfish include members of the genera *Fusarium*, *Ochroconis*, *Exophiala*, *Scytalidium*, *Plectosporium*, *Cladosporium* spp. and *Acremonium*.

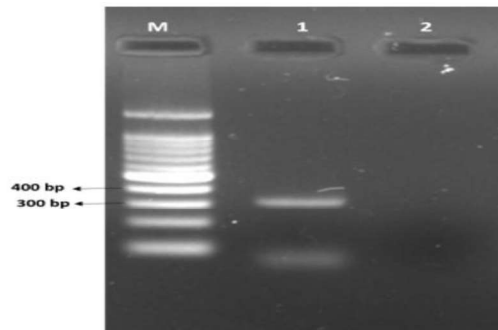
Disease diagnostic techniques in marine fish

Monitoring mortality patterns in a culture system can provide valuable insights into potential disease diagnosis. A relatively uniform pattern of mortality involving several different species often points to fluctuations in environmental parameters such as temperature, dissolved oxygen, salinity, or pH. In contrast, sporadic or random mortality typically suggests a potential infectious or localized issue, warranting immediate submission of affected animals to a diagnostic facility. Any mortality rate exceeding 0.3% per day should be investigated to identify underlying causes. A daily mortality rate greater than 1.5% should be considered an epizootic, requiring urgent response. For clinical disease cases with mortality above 0.5% per day, a diagnostic sample of 10 moribund fish or shellfish is generally adequate. In the absence of noticeable mortality or clinical signs, a larger sample size, typically around 60 healthy-

looking individuals is recommended for routine screening or surveillance. A typical disease diagnosis of fish includes mainly five steps, as collection of detailed history/anamnesis, inspection of the farming site, sampling, necropsy and detailed investigation in the lab. During sampling, along with animals, we have to sample water, sediment and feed if possible. Collecting the physiochemical characteristics of the water is very important in fish disease diagnosis. Careful observation of external symptoms and behaviour prior to necropsy plays a crucial role in the early diagnosis of fish diseases. Clinical signs such as abnormal swimming patterns, skin lesions, excessive mucus production, fin erosion, or changes in body coloration often serve as initial indicators of underlying health issues. Following this, post-mortem examination provides further insights by revealing gross pathological changes in internal organs such as enlarged spleen, necrotic liver tissue, or fluid accumulation in the body cavity. While not conclusive on their own, these clinical and gross pathological findings offer valuable preliminary clues that help narrow down the list of potential pathogens. During necropsy, after documenting gross lesions, standard protocols should be followed for parasitological examination (Palm & Bray, 2014; Hennersdorf et al., 2016). Initially, fish should be inspected for any visible ectoparasite infestations immediately upon sample collection. In the laboratory, a thorough visual examination of the skin, mouth, fins, and gills is carried out to detect any macroscopically visible parasites, cysts, or nodules. This is followed by direct microscopic examination of wet impression smears prepared from the skin, fins, gills, and various internal organs, as well as from the stomach and intestinal contents, to identify the presence of microscopic parasites or cysts. For bacteriological investigations, blood and internal organs (kidney, spleen, liver and brain) are collected aseptically. The pooled tissue samples are homogenized in sterile normal saline solution, serially diluted and spread onto different media, like Zobell marine agar (Himedia), 1/10th strength nutrient agar (Himedia) and thiosulphate citrate bile salt sucrose agar (Himedia). Morphologically unique colonies at the end of incubation are purified and identified based on conventional microbiological tests (Bergey et al. 2012) and 16S rRNA gene sequence analysis. For screening viruses, supernatant prepared from tissue homogenate of various internal organs is inoculated onto a confluent monolayer of fish cell line and examined for cytopathic effects. Histopathological examination of tissue sections stained with hematoxylin and eosin (H&E) is an important diagnostic technique and helps to detect tissue damages, granulomas, and intracellular organisms. An example of histopathological examination showing the typical penetrating hyphae of fungi is shown in Fig. 1.



Molecular techniques like polymerase chain reaction (PCR), real-time PCR (qPCR), and loop-mediated isothermal amplification (LAMP) also provide rapid, sensitive, and specific detection of a wide range of pathogens. An example of a gel visualization after agarose gel electrophoresis after a diagnostic PCR for *V. vulnificus* is shown in Fig. 2. These methods can detect pathogens even in asymptomatic carriers or early infections. Additionally, emerging technologies such as next-generation sequencing (NGS) and metagenomics are now being explored for comprehensive disease profiling.



Role of ICAR-CMFRI in fish disease diagnosis

The ICAR-Central Marine Fisheries Research Institute (ICAR-CMFRI) plays a pivotal role in enhancing fish health management in India, particularly within the marine and brackishwater sectors. As a premier research institute under the Indian Council of Agricultural Research, ICAR-CMFRI is actively involved in the surveillance, diagnosis, and management of diseases affecting marine finfish and shellfish. The institute has established well-equipped fish health laboratories across its regional centers, providing diagnostic services that support mariculture, hatchery operations, and monitoring of wild stocks.

ICAR-CMFRI also offers decision-support services to farmers during disease outbreaks, guiding them in adopting timely and effective interventions. When a disease is reported, ICAR-CMFRI undertakes sample collection following standardized guidelines. A comprehensive case history (anamnesis) is documented, and water quality parameters are assessed to identify deviations from optimal ranges. This is followed by detailed diagnostic procedures, including bacteriological, virological, parasitological, and mycological analyses of blood and tissue samples. Based on the definitive diagnosis, tailored management strategies, encompassing preventive and remedial measures, are recommended to the farmers to mitigate losses and restore stock health. Further, ICAR-CMFRI plays a critical role in pathogen screening during the quarantine of imported aquatic and marine species to prevent the introduction and spread of transboundary aquatic animal diseases. The institute supports the Animal Quarantine and Certification Service (AQCS) Centre at Kochi, Kerala, by screening imported marine fish and shellfish for World Organization for Animal Health (WOAH)-listed pathogens. This ensures compliance with international biosecurity standards and safeguards the sustainability of India's aquaculture sector. Additionally, ICAR-CMFRI conducts regular screening of wild

and farmed bivalve populations along India's east and west coasts, including the Lakshadweep and Andaman Islands, under the National Surveillance Program for Aquatic Animal Diseases (NSPAAD).

Through its dedicated research efforts, ICAR-CMFRI has already identified and documented several novel diseases and pathogens affecting India's marine and brackishwater ecosystems. Recognizing the importance of timely and accurate disease identification, the institute has developed and employed advanced biotechnological tools for pathogen detection in marine fish and shellfish. Furthermore, ICAR-CMFRI has established and maintained diagnostic clones representing antigenic regions of major fish pathogens. These clones serve as positive controls in molecular diagnostics, contributing to the standardization and quality assurance of PCR-based protocols across various national fish disease surveillance programs.

Conclusion

Effective disease diagnosis is fundamental to sustaining marine aquaculture and safeguarding wild fish populations. A combination of clinical observations, parasitological, microbiological, histopathological, and molecular techniques enable accurate identification of pathogens affecting marine finfish and shellfish. Timely and precise diagnosis aids in implementing appropriate control measures and helps in minimizing economic losses and ensuring biosecurity in farming systems. In this context, the ICAR-CMFRI plays a pivotal role in strengthening aquatic animal health management in India.