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A review for health management strategies and future aquaculture sustainability
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Published in:
Heliyon

Published: 15/05/2024

Document Version:
Final Published version, also known as Publisher's PDF, Publisher's Final version or Version of Record

License:
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Publication record in CityU Scholars:
Go to record

Published version (DOI):
10.1016/j.heliyon.2024.e29793

Publication details:

Citing this paper
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Review article

Bacterial diseases of Asian sea bass (*Lates calcarifer*): A review for health management strategies and future aquaculture sustainability

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ARTICLE INFO

Keywords:
Barramundi
Infection
Pathogen
Nanoparticle
Vaccination

ABSTRACT

The advent of aquaculture has been one of the most significant shifts in world food supply during the last century. Aquaculture has rapidly expanded and become a global food industry, spurred by population expansion, increased seafood consumption, and decreased captured fisheries. Nonetheless, the exponential growth of aquaculture has emerged as a significant contributor to anthropogenic changes. Unexpectedly, the result has focused in the emergence and spread of new diseases. The Asian sea bass (*Lates calcarifer*) is an economically important species in aquaculture, contributing significantly to the global seafood market. However, bacterial diseases have emerged as a major concern, affecting both wild and cultured populations of this species. The most prevalent bacterial pathogens are streptococcus, vibriosis, nocardiosis, tenacibaculosis, and potbelly disease. Therefore, this review aims to comprehensively analyze both emerging and non-emerging bacterial diseases affecting *L. calcarifer* and explore potential management approaches for their control. Through an extensive literature survey and critical evaluation of research findings, this review highlights the current understanding of bacterial diseases in *L. calcarifer* and proposes strategies for better disease management. In addition, this review looks at the rise and characteristics of aquaculture, the major bacterial pathogens of *L. calcarifer* and their effects, and the specific attributes of disease emergence in an aquatic rather than terrestrial...
1. Introduction

The Asian seabass, *Lates calcarifer*, also called the barramundi/giant sea perch, is a popular aquaculture fish endemic to the Indo-Pacific region and found in tropical and sub-tropical locations between the 50°E and 160°W longitudes and 24°N and 25°S latitudes [1]. Asian seabass is bisexual, catadromous, and euryhaline; it is frequently found in estuaries and mangrove swamps near the sea, where the water salinity and depth range between 30 and 32 ppt and 10–15 m, respectively [2,3]. From 2023 to 2033, the Asian sea bass market is expected to grow at a compound annual growth rate (CAGR) of 5%. It is anticipated that the market will be worth US$ 960 million in 2023 and US$ 1492 million in 2033 (Future market insights, 2023). However, due to euryhaline, sea bass can attain maturity in freshwater too and migrate to the deep sea to breed. The Asian sea bass is a commercially important fish in northern and southeast Asia, the southern part of Australia, and west to East Africa [4]. This fish is widely and intensively cultured in these regions (Russell, 2013). At the crossroads of aquaculture intensification and climate change, Asian seabass is facing infections and diseases by various pathogens, e.g., bacteria, viruses, fungi, and parasites. Bacterial infections and diseases are reported as the most prevalent [5]. The rates of death from vibriosis and other causes were 16.23 and 12.68 %, correspondingly. *Vibrio* spp. mortality in hatcheries was a risk that accounted for 2.77 %. According to the stochastic model, the cost per kilogram of Asian seabass tail production was £2.69. According to the study conducted in the Peninsula of Malaysia, vibriosis costs the economy €0.19 per kilogram of tail, or 7.06 % of the total cost of producing 1 kg of Asian seabass (Mohd Yazid et al., 2021).

In fish, the relationship imbalance among the host, pathogens(s), and the environment result in a range of diseases (viz. bacterial, viral, and fungal infections), underscoring the importance of their interdependent interaction in disease formation and transmission [6,7]. Viruses and parasitic diseases incur more minor economic losses than bacterial diseases, and the latter severely affects the profit margins of the aquaculture business [8,9]. Globally, the disease is one of the most severe constraints to aquaculture [10]. Bacterial infections alone have been reported to destroy more than 10 % of all farmed aquatic species, resulting in substantial economic losses (>10 billion USD) (Evenson, 2016). Finfish is the major aquaculture species throughout the world. For finfish farming, the biggest barriers to sustainable aquaculture expansion are diseases and health management. Most bacterial diseases lack proven or licensed medications, immunizations, or management techniques, posing a significant challenge to the sustainability of aquaculture businesses.

An 'emerging disease', a different aspect of an existing disease (such as greater severity or the occurrence in a new species), or an existing disease that manifests in a different geographic location are all considered emerging diseases [11]. Emerging diseases have drawn more attention in the recent 20 years from researchers studying humans (Krause, 1998) and animals including fish [12]; Harvell et al., 1999). However, while non-emerging diseases are well-known and investigated, they can still represent considerable problems for fish populations [13]. These diseases are not recognized as a threat to fish populations. These diseases are not regarded as recently emerging or fast-spreading infections, yet they may represent a severe threat to fish populations [14]. The disease described has a well-established history of occurrences, and its features, transmissions, and effects on fish health are all reported [13]. Numerous non-emerging diseases have been examined in-depth, and there may already be treatment techniques and preventative measures to lessen their consequences.

Pathogens have a significant influence on aquaculture sustainability and profitability. Bacterial, parasitic, and fungal infections are just a few instances of how they may cause significant financial losses as well as ethical issues in aquaculture across the world. Several bacterial diseases are commonly encountered in finfish aquaculture, such as *Aeromonas hydrophila*, *Streptococcus iniae*, *Flavobacterium columnare*, and *Vibrio anguillarum* [15]. These bacterial diseases significantly affect finfish farming, impacting the economic viability and the overall health of the fish populations. In Asian sea bass, two emerging bacterial diseases, “Luminous bacterial disease” and “Streptococcal infections” have been a concern for the last two decades [15,16]. Among non-emerging bacterial infections, *Vibrio harveyi*, *Tenacibaculum* (previously known as *Flexibacter*, specifically *Tenacibaculum maritimum*, *Edwardiella tarda*, and *Pseudomonas* spp [17] are highly common. In Asian sea bass, non-emerging bacterial infections cause lower growth rates, increased mortality, and compromised immunity, resulting in economic losses and overall poor fish population health. Furthermore, controlling these conditions necessitate additional expenditures for disease diagnosis, treatment, and disease prevention. Previously, antibiotics were utilized; however, currently, inexpensively available monovalent and multivalent vaccines have been licensed for routinely used. Bacterial infections are a big hazard to sea bass aquaculture, and the market is offering an increasing number of treatments and approved immunizations. The fundamental goal of vaccination is to enhance specific protection against the illnesses that vaccinated fish are likely to encounter. By doing so, aquaculture practitioners can improve the overall health and resilience of their sea bass populations [18]. Asian Sea bass (*Lates calcarifer*) is susceptible to a wide range of infections, including bacteria, viruses, and fungi. Efficient vaccination strategies can significantly reduce the likelihood of disease outbreaks in aquaculture environments. Additionally, the integration of innovative technologies such as nanoparticles and biosensors can facilitate early detection of pathogen outbreaks, thereby enhancing fish health and combating various bacterial diseases in Asian Sea bass. This review aims to provide comprehensive knowledge about both emerging and non-emerging bacterial diseases in Asian Sea bass, encompassing clinical signs and symptoms, effective treatment measures for sustainable farming, successful vaccination, the latest technological tools for early disease detection at the farm level, the impact of symbiotics on disease treatment, and advancements in nanotechnology for controlling emerging diseases. By compiling information on these various aspects, this study aims to contribute to a deeper understanding of the threats posed by bacterial diseases to Asian Sea bass aquaculture. This piece of review will serve as a valuable resource for fish farmers, researchers, and
<table>
<thead>
<tr>
<th>Disease Name</th>
<th>Name of pathogen</th>
<th>Host range</th>
<th>Geographic distribution</th>
<th>Stage of infestation</th>
<th>Diagnostics</th>
<th>Clinical sign</th>
<th>Treatment</th>
<th>Zoonotic potential</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Streptococcosis</td>
<td>Streptococcus iniae</td>
<td>Various freshwater and marine fish species</td>
<td>Australia, Bahrain, Thailand,</td>
<td>Younger fish</td>
<td>Standard (TSA, NA,</td>
<td>Underneath of fins and on the ventral abdomen, there is bilateral exophthalmos,</td>
<td>Enrofloxacin,</td>
<td>Yes (Creeper &amp; Buller, 2006; Wanman et al., 2005)</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>like sea bass, tilapia</td>
<td></td>
<td></td>
<td>Blood agar</td>
<td>darker bodies, and redness of the surface can be seen</td>
<td>Oxytetracycline is very effective. Vaccine is also available but it only provides</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>six-month immunity</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Flavobacteriosis</td>
<td>Flavobacterium sp</td>
<td>Various cultured freshwater sea bass species</td>
<td>Thailand</td>
<td>Juvenile, fry</td>
<td>Special media needed (AOA-Anacker ordaI agar)</td>
<td>On the head, mouth, fins, flanks, and gills, ulcerative skin lesions can be seen</td>
<td>Antibiotic treatment (Quinolone) Standard Vaccine available</td>
<td>No (Chokmangmeepisarn et al., 2021)</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>worldwide</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tenacibaculosis</td>
<td>Tenacibaculum marinum</td>
<td>Various cultured freshwater sea bass species</td>
<td>Marine fish at global level</td>
<td>Juvenile, fry</td>
<td>Marine agar, AOA</td>
<td>External cutaneous ulcerations</td>
<td>Early treatment with potassium permanganate or copper bathing may be beneficial, Antibiotic treatment, Vaccine treatment can minimize the mortality</td>
<td>No (Avendaño-Herrera et al., 2006)</td>
<td></td>
</tr>
<tr>
<td>Pot-belly disease</td>
<td>Vibrio sp.</td>
<td>Cultured Asian sea bass</td>
<td>Singapore</td>
<td>Fry</td>
<td>DNaeasy Blood &amp; Tissue Kit (250) (QIAGEN)</td>
<td>Darkened and emaciated, with abdominal distensions, Skinny, darkened bodies, off feed, not schooling</td>
<td>Use of cyclophosphamides and cortisone to treat the condition usually results in the lesions stabilizing</td>
<td>Unrecognized</td>
<td>(Susan Gibson-Kueh et al., 2021)</td>
</tr>
<tr>
<td>Nocardiosis</td>
<td>Nocardia seriolae</td>
<td>Cultured sea bass</td>
<td>Malaysia, Singapore, Indonesia,</td>
<td>Fry</td>
<td>Tryptic soy broth medium</td>
<td>Moribund fish with pale gills, systemic granuloma</td>
<td>Only useful solution of preventing disease is to suppress pathogen activity using antibiotic treatment</td>
<td>No (S. Gibson-Kueh, 2012; Labrie et al., 2005)</td>
<td></td>
</tr>
<tr>
<td>Vibriosis</td>
<td>Vibrio anguillarum</td>
<td>In &gt; 50 fish species</td>
<td>Global</td>
<td>Juvenile, fry</td>
<td>Standard, with salt media TCBS agar a.o</td>
<td>Swollen red lesions on the ventral and lateral parts, as well as dark skin patches that ulcerate and release blood</td>
<td>Treatments for vibriosis include flumequine, oxytetracyclines, sulfonamides (+trimethoprim), and florfenicol, vaccine is available.</td>
<td>No (Frans et al., 2011; Sharma S R et al., 2013; Zhang et al., 2020)</td>
<td></td>
</tr>
<tr>
<td>Vibrio harvey</td>
<td></td>
<td>In culture of Asian seabass</td>
<td>warm regions, ubiquitous in seawater</td>
<td>Juvenile, fry</td>
<td>TCBS agar a.o</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Photobacterium</td>
<td>Photobacterium damselae</td>
<td>Marine fish</td>
<td>Japan, Taiwan, China, Israel, Portugal, Mediterranean</td>
<td>Juvenile, fry</td>
<td>Standard</td>
<td></td>
<td>Yes (Crum et al., 2009)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
policymakers, empowering them to implement proactive and effective measures to safeguard the health and productivity of Asian Sea bass populations.

2. Emerging and non-emerging bacterial diseases of Asian sea bass

Vibriosis is the most frequent disease affecting in Asian sea bass [19,20]. *Vibrio alginolyticus* results in a high mortality rate in the egg-rearing and larval stages. Since 2013, scale drop and muscle necrosis disease (SDMND) symptoms in farmed Asian seabass in Vietnam, have been regarded as an increasing concern. SDMND disease in fish shows noticeable exterior clinical indications such as scale loss, muscle deterioration, and eventually death. Naturally infected fish obtained from cage culture revealed symptoms of severe necrotic muscles with significant immune-related cell infiltration, acute hemorrhagic and plasma congestion in the brain, ruptured kidney tubules, and tissue macrophages sloughing into the lumen [21]. A hypothetical protein HP33(33 kDa) from *Vibrio harveyi* Y6 is supposed to cause SDMND in Asian sea bass [22]. Researchers found that only *V. harveyi*, *V. tubiashii*, *Tenacibaculum litopenaei*, *Tenacibaculum sp.*, and *Cytophaga* sp. killed the fish with identical clinical indications and histological abnormalities [21]. In Asian sea bass, *Streptococcus iniae* infections cause systemic infections of the liver, pancreas, heart, eye, and brain. The infiltration of vast numbers of macrophage centers in infected tissues [23]. Streptococcosis can manifest in juvenile fish with few clinical indications and a high death rate of up to 70 %. Affected fish show bilateral exophthalmos, darker bodies, and skin reddening on the fins and the abdomen in subacute instances. Cumulative mortality in subacute cases can reach 50 % over several weeks [24,25]. *S. iniae* was identified as a severe infection in two farms in South Australia, [26]. *S. iniae*-related disease occurrences in Thailand’s freshwater pond-cultured seabass were examined. Except for the exophthalmological lesions, the infected fish showed normal indications of streptococcal infection. Flavobacterium (formerly Cytophaga or Flexibacter) is a gram-negative filamentous bacterium with gliding locomotion that can be found in both fresh and ocean environments. Flavobacteriosis indeed a dangerous disease that can cause infections in cultivated finfish globally, both in freshwater and in the sea [27]. In 2018 and 2019 at Chachoengsao and Samut Prakan provinces in Thailand infections of *Flavobacterium columnare* were reported with clinical signs of gill necrosis, saddleback lesion, and fin erosion [28]. Tenacibaculosis is caused by species of the genus *Tenacibaculum* (family Flavobacteriaceae, phylum Bacteroidetes) is another frequent bacterial illness in tropical aquaculture in the Southeast Asian region, most significantly *Tenacibaculum maritimum*. The effect of tenacibaculosis on the microbiota of fish is still unknown. Symptoms of these diseases include ulcerative skin sores on the head, mouth, fins, flanks, and gills. Stressful events like improper handling, higher fish density, or lack of nutrients in the water have been linked to illness outbreaks [29]. Tenacibaculosis and vibriosis can develop simultaneously and cause identical external cutaneous ulcerations. In Southeast Asia, tenacibaculosis was most prevalent between 1 and 100 g sea bass, particularly in Singapore. Bacterial septicemia is produced by opportunistic pathogens, including *Aeromonas* spp. (pond water) and *Vibrio* spp. and *Photobacterium damsela* subsp. *damsela* (saltwater) has been observed to create significant mortality in farmed Asian sea bass [30,31]. A facultative intracellular Gram-negative bacterium (likely a novel *Vibrio* species) causes a big belly, commonly known as a skinny potbelly [32]. It can be found in Indonesia, Singapore, and Malaysia, among other Southeast Asian countries. Pot-belly disease has been documented in *L. calcarifer* fry as young as three weeks old, with up to 80 % mortality [33]. Big Belly is most common in 25-day-old nursery fry, causing significant internal organ clumping, abdominal distension, and muscle atrophy. Infected individuals often develop a darker coloration, become lethargic, detach from school, and lose balance by swimming on the surface or resting on the bottom of tanks. It is a delayed systemic infection with a gradual but constant start of mortality that has the potential to kill 90–95 % of the population if left untreated. Fish that had been affected will black with abdominal expansion. Multifocal granulomatous enteritis with clusters of large gram-negative cocccobacilli was discovered during the histopathological investigation. Combined with these big cocccobacilli, the disease commonly spreads throughout the body, causing granulomatous peritoneum, spleen, and kidney lesions. Despite the failure of growth, these cocccobacilli reacted favorably to a polyclonal antibody against *Edwardsiella ictaluri* in immunohistochemistry [33]. Researchers recently reported the first instance of big belly illness in a 3 to 4 week-old Asian seabass fry, in

<table>
<thead>
<tr>
<th>Disease name</th>
<th>Pathogens</th>
<th>Clinical symptoms</th>
<th>Preventive measures</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bacterial haemorrhagic septicaemia</td>
<td><em>Aeromonas</em> spp.; <em>Pseudomonas</em> sp.</td>
<td>Associated with inadequate environment &amp; skin injuries; intermittent reddish facial ulcerations; lethargic; anorexia; reddened abdomen fluid; pale gills</td>
<td>Antibiotic therapy and a better environment</td>
</tr>
<tr>
<td>Integumentary bacteriosis</td>
<td><em>Aeromonas</em> hydrophila; <em>Vibrio harvey</em></td>
<td>Intermittent reddish skin ulcerations; scale loss; linked to poor habitat and tissue damage.</td>
<td>Improved environment; increased water exchange</td>
</tr>
<tr>
<td>Bacterial gill disease</td>
<td><em>Flavobacterium</em> spp., <em>Cytophaga</em> spp.</td>
<td>White patches on the gills; swimming towards the surface of the water; fast opercular movement; copious mucus on the gills; especially prominent in the early stages of growth</td>
<td>Optimize water exchange; improve the water parameter with salinity alteration, KMno₄ or quaternary NH₄ baths; minimize stocking density</td>
</tr>
<tr>
<td>Bacterial peritonitis</td>
<td><em>Vibrio harvey</em>, <em>A. hydrophila</em></td>
<td>Adhesions and foul-smelling fluid in the abdomen; Darkened fish; lethargy;</td>
<td>Cull the diseased fish and treat them with antibiotics</td>
</tr>
<tr>
<td>Bacterial enteritis</td>
<td>Numerous bacteria (Gram-negative)</td>
<td>Anorexia, pin heads, darker color, and mortalities in intense larval rearing systems</td>
<td>Cull affected larval batch</td>
</tr>
<tr>
<td>Fin and tail rot</td>
<td><em>Pseudomonas</em> spp.; <em>Vibrio</em> spp.; <em>Flavobacterium</em> spp.</td>
<td>Soft tissue deterioration in the fins and tail, which may extend to the entire tail and caudal peduncle.</td>
<td>Optimize the ecosystem by lowering the stocking density</td>
</tr>
</tbody>
</table>
Singapore [34]. The injured fry was emaciated and had a large abdomen, according to the authors, and the ailment was dubbed "skinny potbelly" or "big belly" disorder. Further "Big belly" coccobacilli were detected in intralesional clusters in formalin-fixed paraffin-embedded tissues using dioxygenin-labeled positive PCR results. "Big belly" bacteria was likely an isolated Vibrio species, according to a study of 16S-rRNA gene-PCR findings from five fish that tested positive for the bacteria [34]. A detailed description of the major bacterial pathogens in L. calcarifer have been listed in Table 1 and Table 2.

Myxobacterium marinum causes tuberculosis in Asian seabass and other finfish of Southeast Asia. Although there is not an incredible amount of data on this ailment. Furthermore, Nocardiosis is a non-emerging bacterial disease that can have significant effects on Asian seabass (L. calcarifer) in aquaculture (Gibson-Kueh et al., 2005). Nocardiosis is a systemic bacterial infection caused by a Gram-positive, somewhat acid-fast, aerobic filamentous bacterium [35]. Nocardiosis in Asian sea bass may result in lower fish development, higher death rates, and financial losses due to symptoms including skin ulcers, fin rot, and hemorrhages. Routine clinical examinations in farmed fish in Southeast Asia over the last few years have shown that Nocardiosis is endemic in countries like Taiwan, Malaysia, China, and Indonesia. There were varying degrees of death rates associated with this condition. Nodules in the gills, spleen, kidney, and liver and many skin ulcers and nodules were common clinical symptoms. Edwardsielliosis has been linked to increased mortality in farmed barramundi (L. calcarifer) in the United States [36]. According to phenotypic and genetic research, the etiological agents are Edwardsiella tarda and E. piscicida. External symptoms included erythema and/or bleeding on the ventrum, gill, pallor, and fin/oral cavity congestion, as well as internal hepatic pallor/mottling, and erythema of the gastrointestinal tract, but internal symptoms included splenomegaly, renomegaly, and small whitish miliary nodules in the liver, spleen, and kidney [36]. Sun et al., 2020 revealed the association of Pseudomonas plecoglossicida in barramundi (L. calcarifer) [37]. P. plecoglossicida is a highly deadly pathogen linked to significant financial losses in aquaculture farming. Lethargy, inappetence, disorientation, abdominal swelling with severe ascites, Bowman capsule space becoming wide, ascites, and numerous white patches coated on the surface of spleen tissue are common symptoms in L. calcarifer infected with P. plecoglossicida [37]. P. plecoglossicida could produce systemic infections in barramundi with typical clinical indications, making it a dangerous pathogen for the industry. Pseudomonas sp. KUMS3 had previously been linked to hemorrhages at the base of fins, mouth, and skin muscles in India in 2010 [38]. Black body syndrome (BBS) is another emerging disease for barramundi aquaculture reported in Indonesia in 2020 with clinical signs of a blackened body and bleeding under the belly [39].

For aquaculture, efficient disease identification and control are critical for maximizing productivity and guaranteeing optimal final product quality [40,41]. Nano biosensors provide a novel approach to issue-solving. These sensors, which can be made of a variety of nanomaterials, including carbon nanotubes, can detect low levels of pathogens such as bacteria, viruses, and parasites, as well as toxins [42]. Guo et al. (2016) created a microfluidic chip using indium tin oxide to construct an immunomagnetic NP-based microfluidic solution to track S. aureus. The detection approach does have the same specificity and sensitivity as the colony counting method, with a much quicker trace level without colony culture. Nanotechnology can help improve the identification and control of aquatic diseases in addition to detecting them. The adoption of more efficient formulations can address issues connected with traditional disinfec

Molle Aeromonas septicemia is caused by the ubiquitous aquatic disease A. hydrophila, which kills a variety of fish species around the world, including Asian sea bass. The identification of A. hydrophila pathogenicity via PCR is difficult, time-consuming, and costly. The recent invention of a quick, specific, and sensitive assay for detecting A. hydrophila DNA without prior amplification using a particular probe conjugated with gold nanoparticles (Aeromonas-AuNPs-probe assay) should allow for the visual detection of A. hydrophila DNA in 30 min without any prior amplification [44].

The use of gold nanoparticles for fish pathogen detection was originally reported with the use of an A. salmonicida antibody-gold nanoparticle combination for the specific immunological diagnosis of furunculosis in fish tissues [45]. Empa and ETH Zurich researchers are now working on nanoparticles that can be used to detect and kill multi-resistant bacteria that lurk inside human body cells (https://www.news-medical.net). Antibiotics have a hard time infiltrating animal cell, but because of their small size and structure, these nanoparticles can permeate the membrane of afflicted cells and combat bacteria. This revolutionary approach has the potential to expand the scope of aquatic disease study.

3. History and update of vaccines against bacterial diseases in sea bass

Immunization is currently one of the pivotal methods for the prevention of bacterial diseases in aquaculture species. As a preventative intervention, fish immunization can be the most effective alternative to the use of antibacterial drugs in aquaculture. In recent years, the occurrence of microbial infections in a range of aquaculture species and the use of vaccination strategies as prophylaxis measures have been expanded. For example, researchers examined the defensive efficacy of bacterial vaccinations against V. anguillarum in Asian sea bass in experimental trials [46,47]. Moreover, infections with V. harveyi [48], Streptococcus iniae [49], and infestation with Flavobacterium columnaris [50] in barramundi are also treated with experimental vaccines.

As a polycationic gene carrier, chitosan has been extensively investigated for more than 20 years. Researchers investigated at the effectiveness of a DNA vaccine against V. anguillarum in L. calcarifer, which were orally vaccinated with a chitosan-DNA complex and then exposed to a V. anguillarum infestation using chitosan nanoparticles encapsulation [51]. Vaccination with outer membrane protein (OMP) against V. anguillarum in Asian sea bass was undertaken in India. Experts used chitosan microparticle encapsulated DNA vaccines and saline mixed antigen formulations to deliver DNA vaccines via oral and intramuscular routes, respectively [47].

Another infection that causes enormous mortality in barramundi in Asian capture fisheries, mainly in Indonesia, is black body syndrome. For Barramundi, researchers demonstrated the impact of a bivalent vaccine developed from a local isolate for the prevention of black body syndrome (BBS). Pseudomonas stutzeri and V. harveyi were utilized in a 50:50 ratio in the bivalent vaccination
Antibody levels and lysozyme activity were higher in the vaccinated fish than in the control group. Immunization against a variety of different pathogenic bacteria has been investigated and determined to be technically possible. Vaccination has a net positive effect on farmed fish, lowering mortality. Nevertheless, it is equally critical for the future of the fish farming sector because immunization promotes sustainable biological production with little antibiotic use. To make progress in fish vaccination, more collaboration between basic and applied science is required. For booster immunization, biodegradable microparticle-based vaccines that improve oral immunity will be employed. The improvement of non-mineral oil adjuvants with minimal adverse effects is a pressing necessity in today’s world [52].

Streptococcus agalactiae and S. iniae pose major disease issues to Asian seabass farmers in Southeast Asia. Asian seabass was immunized with oil-based formalin-killed vaccines (FKVs) made from S. agalactiae and S. iniae (monovalent Sa, monovalent Si, and bivalent Sa-Si) and then given a water-based FKV as a supplementary booster. Scientists examined the mucosal and systemic antibody response kinetics (IgM) [53]. According to the findings of this study, the antibody reaction in Asian sea bass vaccinated with monovalent vaccines is better compared to that of fish vaccinated with bivalent vaccines in respect of particular antibody titer, and the mixture of S. agalactiae and S. iniae in a single injectable vaccine is plausible [53]. Photobacterium damselae subsp. piscicida (Phdp) causes photobacteriosis in marine fish and is blamed for declining revenues in marine aquaculture around the globe. Previously, Four heat-shock proteins (HSP90, HSP33, HSP70, and DnaJ) were found to be effective as recombinant vaccines against photobacteriosis in Asian seabass [54]. Additionally, the moonlighting protein fructose 1,6-bisphosphate aldolase (Fba) can protect sea bass from Photobacterium damselae subsp. piscicida [55]. They test the immunoprotective effects of recombinant proteins of catalase, superoxide dismutase, isocitrate dehydrogenase, fructose 1,6-bisphosphate aldolase (Fba), and a combination of all four proteins against photobacteriosis. They concluded that Fba is a promising candidate for developing a subunit vaccine against photobacteriosis in fish after immunizing experimental fish. Silvaraj et al. (2021) studied the effect of intraperitoneal delivery of recombinant outer membrane protein (OMP) of inactivated cells vaccine; r-OmpK on hematological parameters for fish health monitoring and histological inspection of fish. Following a vaccination trial with r-OmpK recombinant cell vaccine and a challenge with V. harveyi pathogenic strain VH1, researchers analyzed the transcriptome of L. calcarifer head kidneys. The investigation found that in response to V. harveyi infection, a more significant number of genes and pathways were regulated. Further protein-protein interaction analysis indicates the existence of unique immune-related signaling pathways in sea bass, including complement and coagulation cascades, chemokine signaling pathways, and toll-like receptor signaling pathways, all of which are important in immunity and anti-inflammation [56]. In Malaysia, 300 juvenile Asian sea basses tested positive for Vibriosis, Streptococcus, and motile aeromonad septicemia after receiving a newly designed feed-based whole-cell polyvalent vaccine [57]. The feed-based polyvalent vaccine may elicit strong innate and adaptive immune responses, providing potential for comprehensive immunization against vibriosis, streptococcus, and motile aeromonad septicemia in Asian seabass.

Nocardiosis in fish is a chronic granulomatous bacterial disease caused mainly through three pathogenic bacteria: Nocardia seriolae, N. asteroides, and N. salmonicida, as well as a developing disease in Asian sea bass (L. calcarifer) (Gibson-Kueh, 2012). DnaK, a molecular chaperone, and GroEL, a shared antigen of three pathogenic Nocardia species, were discovered by researchers [58]. Hybrid snakeheads were vaccinated to test the immune protective effect of two DNA vaccines encoding DnaK or GroEL against fish nocardiosis. The results indicated that it took at least seven days for the DnaK or GroEL gene to travel from the injected muscle to the head kidney, spleen, and liver, stimulating the host’s immune system for later protection after DNA vaccine immunization. The protective efficacy of pcDNA-DnaK (53.01 %) and pcDNA-GroEL (80.71 %) in terms of relative percentage survival (RPS) was good, and researchers suggested that DnaK and GroEL were promising vaccine candidates for the development of DNA vaccines against fish nocardiosis in finfish aquaculture [58]. This approach can also effectively control mass mortalities in sea bass farms. Researchers in Thailand created a mucoadhesive immersion F. columnare nanoencapsulation vaccine to provide better protection for Asian seabass against different columnarins isolates, and they found that the prepared vaccine candidate has more potential as compared to whole-cell immunization [59]. This can also be tested for Asian seabass to see if this works. S. iniae is another significant aquatic pathogen that causes invasive diseases in farmed marine and freshwater fish globally, including sea bass, and an orally attenuated vaccine is currently the best option for protecting against these invasive infections [59]. Researchers looked at the safety, stability, and immunogenicity of the S. iniae attenuated strain YM011 and discovered that it was a good option for tilapia disease resistance [59]. S. iniae infection is also widespread in sea bass aquaculture, and future genetic studies will reveal more about the YM011 strain, allowing us to design an attenuated vaccine to control streptococcus at the farm level. Although an inactivated vaccination against S. iniae is available, it only protects sea bass for six months.

Another unique technique for controlling motile aeromonad infections in farmed species is to use transcutaneous immunization (TCI) with dissolving microneedle (MNs) patches to immunize fish against Aeromonas hydrophila infections [60]. TCI employing dissolving MNs has gotten a lot of attention; it has already been explored for a range of human diseases such as tetanus, diphtheria, malaria, and influenza, and it can also be effective against motile aeromonads in finfish [61,62]. Besides, A. salmonicida 70 kD serine proteases fusion protein has been cloned, produced, and practiced as a prototype vaccine. It was recently discovered that A. hydrophila native main adhesion, a 43 kD outer membrane protein, has the potential to be employed as a fish vaccine. In addition, some fish bacterial infections, such as A. salmonicida and A. hydrophila, have been treated with genetically modified live vaccines. Many procedures are utilized to create virulent mutant bacteria that can be used as live vaccines, including homologous recombination, chemical mutagenesis, and transposon mutagenesis (Khati et al., 2021). Moreover, the nanocellulose base fish vaccine can revolutionize the aquaculture industry in terms of disease management, particularly for Vibriosis (ScienceDaily, 2021). Nanocellulose is a biologically derived natural polymer found in the cell walls of wood and plants and may offer several benefits as a vaccine component. Nanocellulose is biocompatible, meaning it has no known harmful effects on tissue and is unlikely to cause cellular damage. Vaccines developed with nanocellulose may also be less expensive to manufacture than their oil-based counterparts, according to researchers
Nanomaterials have been used to improve fish health and immune status and prepare fish vaccines. Following bacterial infection, fish-fed diets supplemented with nano-selenium grew faster and had better immunity [64]. Chitosan NPs, which are prized for their intrinsic qualities of mucoadhesion, low toxicity, biodegradability, and biocompatibility, have also been proven to improve tilapia growth [65]. Recent research is focusing on using NPs in combination with gene manipulation to improve the dietary conversion of nutritious substances like carbohydrates. The researcher combined chitosan-tripolyphosphate NPs with a plasmid that promotes the expression of SREBP1a, a transcriptional factor that improves glucose metabolism [65]. Improved dietary carbohydrate conversion to fatty acids and cholesterol because of metabolic regulation could contribute to protein sparing. It is an example of NPs being used to influence endogenous host metabolic processes via a delivery method. Chitosan NPs are frequently used as carriers in vaccines against various aquatic diseases, including DNA vaccine against *Vibrio anguillarum* through an oral route using chitosan nanoparticles encapsulation already developed. This oral immunization of Asian sea bass with chitosan nanoparticles encapsulated DNA vaccine induced moderate protection against the OMP38 protein of *V. anguillarum* [51]. Scientists have determined that Chitosan is non-toxic to both experimental animals and humans and is derived from crab shells [66]. Chitosan is also employed as a provider for many types of DNA and vaccinations in fishes via various modes of delivery, owing to such characteristics (orally or an injection). As an example, dietary RNA in carp (Ferosekhan et al., 2014) is being encapsulated and found to be satisfactory in CS-based applications. Nano-capsules, which are resistant to degradation and protect compounds such as short strings of nucleic acids, are generated in one method. These nano-capsules are delivered to cells, which are subsequently chemically or physically triggered to release the DNA (i.e., pH, ultrasound). Once released, a host immune response is generated as a targeted technique of providing long-term protection. PLGA (poly lactic-co-glycolic acid) [67] and variants of chitosan are common macromolecules used in nano-capsules and have been used to target pathogens [68].

Recently it has recommended a nano-delivery mechanism as an alternate technique for vaccine delivery in fish, which is cheaper and far more effective. Until now, various encapsulation approaches have been proposed and tested in fish. Alginate particles were identified as experimental contenders for oral vaccination administration to aquatic species [69]. Alginate is indeed a copolymer of b-Dmannuronic acid (M) and a-L-guluronic acid (G) found in many brown algae and as a carbohydrate in bacterial pathogens. It is identified as experimental contenders for oral vaccination administration to aquatic species [69]. Alginate is indeed a copolymer of b-Dmannuronic acid (M) and a-L-guluronic acid (G) found in many brown algae and as a carbohydrate in bacterial pathogens. It is recognized for its chemical and microstructural resilience, including its mucoadhesive qualities, which enable it to adhere to epithelium cell membranes, making it a better candidate for oral delivery (Sosnik, 2014). Furthermore, emulsification (Leal et al., 2010), which is one of the orders to achieve the optimum for NP synthesis and is fully accessible (Cheng et al., 2008), and the orange-spotted grouper and brown-marbled grouper *Streptococcus* sp. (Yeh et al., 2008). In addition, a nanoparticle composed of zinc-layered hydroxy chloride and yeast-glucan (ZG) enhances fish species’ innate immune responses [70].

Another successful and less dangerous method for controlling fish bacterial infections is phytotherapy combined with nanotechnology. *Aeromonas hydrophila*, *E. tarda*, and *S. iniae* were all successfully treated with zein nanoparticles combined with eugenol and garlic essential oil [71]. In investigations using a biomarker, Zein nanoparticle formulations containing botanical components demonstrated decreased toxicity (*Artemia salina*).

Emerging nanobubble technology in freshwater aquaculture, to lessen harmful fish bacteria concentrations in freshwater while also determining if nanobubbles are safe for *A. hydrophila* treatment. The size, concentration, disinfection property, and impact on fish health are all factors in ozone nanobubble (NB-O3) treatment. A 10-min treatment with NB-O3 in 50 L water produces around 2–3 × 10^7 bubbles/mL, with the bulk of bubbles less than 130 nm in diameter and an oxidation-reduction potential (ORP) of 834 ± 22 mV. The bacterial load can be effectively reduced by a single treatment of water spike with either *S. agalactiae* or *A. veronii*. NB-O3 technology may also be effective in minimizing the danger of bacterial disease outbreaks in farmed fish [72].

Table 3
Available vaccine for bacterial fish disease of Sea bass.

<table>
<thead>
<tr>
<th>Disease</th>
<th>Disease agent</th>
<th>Antigens</th>
<th>Type of vaccine</th>
<th>Method</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vibriosis</td>
<td><em>Vibrio harveyi</em></td>
<td>Inactivated <em>Vibrio</em></td>
<td>Inactivated</td>
<td>IP or IMM</td>
</tr>
<tr>
<td>Pasteurelosis</td>
<td><em>Photobacterium damselu subsp.</em></td>
<td>Inactivated <em>P. piscicida</em></td>
<td>Inactivated</td>
<td>IMM</td>
</tr>
<tr>
<td>Flavobacteriosis</td>
<td><em>Flavobacterium columnare</em></td>
<td>Attenuated <em>F. columnare</em></td>
<td>Attenuated</td>
<td>IMM</td>
</tr>
<tr>
<td>Flexibacteriosis</td>
<td><em>Tenacibaculum maritimum</em></td>
<td>Inactivated <em>T. maritimum</em></td>
<td>Inactivated</td>
<td>IP</td>
</tr>
<tr>
<td>Infection with motile Aeromonas</td>
<td><em>Motile Aeromonas sp.</em></td>
<td>Inactivated <em>S. agalactiae</em></td>
<td>Feed-based polyvalent vaccine</td>
<td>IP</td>
</tr>
<tr>
<td>Streptococcus</td>
<td><em>Streptococcus iniae</em></td>
<td>Inactivated <em>S. agalactiae</em></td>
<td>Inactivated</td>
<td>IP</td>
</tr>
</tbody>
</table>

IP: Intraperitoneal injection; IMM: Immersion.
4. Probiotics and Prebiotics prospects in Asian sea bass aquaculture

Increasing a fish’s immune defense naturally is one of the important research topics in aquaculture. The main focus has been on compounds that can be integrated into feed and given to fish orally, although other substances could be administered with vaccinations. Many of the early commercial benefit reports were not supported by investigations into the mechanism of action, and indications of immune system involvement could not be substantiated. More recent studies now include data on the effects of treatment on a variety of immune bioassays, and there appears to be some immunomodulation. Immunostimulants either directly or indirectly...
stimulate particular or nonspecific defensive mechanisms, depending on their mode of action [73]. Immunostimulants (probiotics and prebiotics) are one of the most promising alternatives to prophylactic antibiotics in aquaculture for disease prevention, allowing for an eco-friendlier approach. These natural chemicals boost fish’s immune systems, help with stress relief, and function as anti-pathogenic agents due to their bioactive compounds [74–76].

Symbiotics, which combine probiotics (beneficial bacteria) and prebiotics (substances that promote the growth of beneficial bacteria) enhance survival rates and intestinal microbiota regulation [77]. This synergistic strategy improves gut health and immune function by creating a favorable environment for the development and colonization of beneficial bacteria. Studies have shown symbiotics to improve survival rates and modulate gut microbiota in various animals, including fish [78]. Additivity, synergism, and potentiation are the three patterns that occur from the use of two or more feed additives in general [79]. Prebiotic activity may be enhanced as a result of this component’s contribution to probiotic bacteria growth, metabolism, and activation [80]. Research on symbiotics suggests that they increase the host’s enzymatic digestion, the generation of acetic, lactic, and butyric acids (products of probiotic bacteria fermenting prebiotics), and innate immune system activation (Fig. 2) [81].

In aquaculture, a variety of microorganisms have been tested as probiotics. Shewanella putrefaciens, Bacillus subtilis, Lactobacillus acidophilus, Lactobacillus sakei, and Bacillus subtilis are only a handful of them. They can be utilized to avoid sickness and enhance weight gain in farmed fish and other animals. Probiotics can be added to the meal or directly to the water. Encapsulation is the other administration strategy. Encapsulation aids in the transfer of microbes to the host while minimizing the waste of living organisms [82]. To physically and chemically preserve the microorganisms, the organism’s cells are enclosed in a colloidal matrix made of alginate, chitosan, carboxymethylcellulose, or pectin [83]. Bacteria are protected from low pH and digestive enzymes by encapsulation in alginate matrices. Probiotic immobilization is another technology applied to a LAB, which is widely employed in the dairy and pharmaceutical industries. Cell immobilization, in particular, has been found to provide numerous advantages for biomass and metabolite synthesis when compared to free cell systems, including high cell density [83].

In addition, prebiotics are non-toxic, stable chemicals that leave no residues and can be found in several places, including seaweed. Mannan oligosaccharides (MOS) are a component of Actigen (commercial prebiotics) generated from the yeast cell wall. It binds to pathogens, preventing their colonization in the intestine, and changes the intestinal shape, resulting in increased microvilli density, reduced tight junction exposure, and improved nutrient absorption. Chitosan is a probiotic that is formed when chitin is deacetylated and has been shown to boost fish immune activity [84]. Prebiotics and probiotics must be carefully chosen for symbiotic preparation since improper combinations might cause animal physiology and microbial diversity difficulties.

For seabass, only a few studies have reported on the effects of prebiotics and probiotics [85,86]. Sargassum (S. polycystum), a brown seaweed, was discovered to have prebiotic potential when employed in an experimental diet formulation on Asian sea bass fingerlings [87]. The addition of graded amounts of seaweed to conventional meals resulted in a considerable change in mineral content, particularly [88]. Prebiotic feed supplements are thought to help normalize the bacteria flora in the GI tract, preventing disease-causing opportunistic pathogens from invading. Lactic acid bacteria (LAB) from the intestine of juvenile seabass (L. calcarifer) was recently isolated, identified, and characterized as a new potential probiotic by researchers [89]. The intestines of ten healthy seabass juveniles yielded four LAB strains. In the in vitro screening process utilizing spot lawn assay, Enterococcus hirae was detected...
Nanomedicine has the potential to change disease treatment in animal models. Existing research has established the feasibility of nanotechnology-based drug delivery systems can now be used to treat chronic intracellular infections in both humans and animals. Development of pathogen-free fish seedlings has been reported as a nano-encapsulation carrier. For agriculture, these nanomaterials are proven to be highly beneficial in the absorption and utilization of trace elements for improved physiological functions. Limiting viruses, bacteria, and fungi would require early identification and removal of pathogens, which might be accomplished using nanomaterials, which work on the same scale as a virus or disease-infecting particle. To successfully treat pathogenic bacteria and viruses, vaccines wrapped in chitosan have been reported as a nano-encapsulation carrier. For agriculture, these nanomaterials are proven to be highly beneficial in the development of pathogen-free fish seedlings.

5. Application of nanotechnology in fish bacterial disease management

Nanotechnology is thought to be offered a solution for preventing and monitoring diseases and infections, as well as multiplying the benefits of fish farming. Some applications of nanotechnology for improving fish health are the development of antibacterial or antifungal surfaces using porous nanostructures, the use of nanosensors in freshwater aquaculture for bacterial identification in waterways, and the administration of veterinary items and fish medicines via fish meals. Nano-trace element usage involves utilizing trace elements in nanoscale forms, which can be up to 100 times more effective compared to standard inorganic trace element usage. This enhanced efficacy is attributed to the ability of nano-sized particles to directly penetrate animal organs, facilitating better absorption and utilization of trace elements for improved physiological functions. Limiting viruses, bacteria, and fungi would require early identification and removal of pathogens, which might be accomplished using nanomaterials, which work on the same scale as a virus or disease-infecting particle. To successfully treat pathogenic bacteria and viruses, vaccines wrapped in chitosan have been reported as a nano-encapsulation carrier. For agriculture, these nanomaterials are proven to be highly beneficial in the development of pathogen-free fish seedlings.

6. Antibacterial drug

In the field of drug delivery, the era of nanotechnology has enabled new research methodologies to develop antibacterial drug. Nanotechnology-based drug delivery systems can now be used to treat chronic intracellular infections in both humans and animals. Nanomedicine has the potential to change disease treatment in animal models. Existing research has established the feasibility of introducing nano shells and nanotubes into animal systems to kill specific cells. Drugs have been delivered into cells using nanoparticles with few adverse effects. Through co-enzymatic systems, the creation of nanoparticles from metals involves a variety of biological processes. Chelation interaction between these nanoparticles and physiologically active ligands in the mammalian system. Novel antimicrobial medicines are needed due to an increase in bacterial illness outbreaks in the aquaculture business and the development of bacterial resistance. Silver nanoparticles are one of the most effective metallic nanoparticles with antibacterial activity against a variety of bacteria and fish pathogens. Silver ions (Ag+) are released and linked to bacterial cell wall proteins, causing cellular membranes to rupture and apoptosis. Researchers tested the antibacterial properties of ZnO nanoparticles on the pathogenic V. harveyi bacteria and discovered that nanoparticles had better bactericidal effects than bulk ZnO. Another fascinating work conducted A. hydrophila to make ZnO nanoparticles and these nanoparticles showed antibacterial efficacy against the same bacterium and additional species: Candida albicans, Pseudomonas aeruginosa, E. coli, Enterococcus faecalis, Aspergillus flavus. Furthermore, biological zinc nanoparticles (BIO-ZnONPs) were found to have good antibacterial action against harmful fish pathogens in Nile tilapia (Oreochromis niloticus). After activation by light, TiO2-NPs doped with magnetic Fe3O4-NPs displayed a bactericidal effect against S. iniae, E. tarda, and Photobacterium damselae. Because fish infections link to nanoparticles, which can then be easily removed from water using a magnet, these particles can be utilized to treat water. Nevertheless, researchers found that TiO2-NPs influence the fish immune system by decreasing the antibacterial activity of fish neutrophils, making the fish more susceptible to infection and thus higher mortality, particularly during illness outbreaks.

7. Phytochemicals for bacterial disease treatment in Asian sea bass

Farmers utilized a range of antibiotics to treat fish diseases to enhance productivity and profit. Antibiotic-resistant microorganisms have risen because of the misuse of broad-spectrum chemotherapeutics. Drug-resistant genes have also been transferred to bacteria and viruses that attack animals and humans, resulting in residual antibiotic levels in food. Antibiotics included in feed are consumed by fish. However, the whole antibiotic is not metabolized and absorbed and thus excreted into the environment. To tackle this unsustainable tendency, researchers are already exploring antibiotic alternatives in aquaculture.
For example, citrus fruit, leaves, whole plants, and peels are physiologically active in a variety of ways that benefit animal health, including being antiangiogenic, anti-inflammatory, antibacterial, antioxidant, immune system regulating, anti-obesity, and hepatoprotective [106,107]. Aquaculture has also benefited from citrus by-products. The antioxidant properties of yuzu (Citrus junos) as an aquafeed additive, for example, protect yellowtail fish (Seriola quinquergiata) from black muscle discoloration during storage [108]. In addition, citrus byproducts fermented with Bacillus subtilis and fed to juvenile olive flounder (Paralichthys olivaceus) for six weeks increased their immune response and illness resistance to E. tarda [109]. Barramundi (L. calcarifer) fed on a diet containing 5% C. depression was found to be healthier. Against A. hydrophila, Hayata leaf meal has a considerably superior innate immune response and disease resistance [110]. The effects of supplementing the diet with Lactobacillus plantarum fermented lemon peel (FLP) on Asian seabass (L. calcarifer) revealed a significant improvement in the immune system. For Asian sea bass, a daily feed of fermented lemon peeling from 1% to 3% can improve intestinal health [111].

Sargassum polycystum as a supplement in the diets of Asian sea bass fingerlings for up to 55 days encourages the growth of possible Lactobacillus paracasei subspecies paracasei in the fish intestine and acts as a prebiotic [105]. Subsequent research revealed that carvacrol, eugenol, and thymol had acceptable antibacterial action when given alone or in combination with fluorfenicol or oxytetracycline to bacteria isolated from silver catfish [112]. Linalool with florfenicol or oxytetracycline demonstrated promising effects against A. hydrophila and citral with oxytetracycline against Citrobacter freundii. Against bacterial pathogens, all phytochemicals decreased biofilm formation and hemolysis activity, while phytochemicals had no antagonistic associations with antimicrobials [112].

The neem (Azadirachta indica) possesses significant antibacterial [113] and antiviral properties [114]. Furthermore, neem has been shown to have anti-inflammatory, antioxidant, hepatoprotective, and cancer chemo-preventive properties [115,116]. Researchers studied the effects of supplementing diets with A. indica (neem) leaf and garlic (Allium sativum) on the innate immune response in Asian seabass fingerlings, L. calcarifer, against Vibrio harveyi infection [117,118]. In the aquaculture system, neem leaf food supplementation can considerably improve phagocytic activity, superoxide anion generation, serum lysozyme, serum bactericidal activity, and serum anti-protease activity. A neem leaf diet can considerably influence the immunological parameters, hematological parameters, and blood biochemical indices of fish [117]. Another phytochemical, Jerusalem artichoke (JA) (Helianthus tuberosus), was utilized to successfully treat A. hydrophila in juvenile Asian seabass (Lates calcarifer) [119]. Researchers reported that supplementing L. calcarifer juveniles with 20 g kg⁻¹ JA improves their metabolic, immunological, and illness resistance [119]. Moreover, Adenostemma viscosum, Allium sativum, Alstonia scholaris, Amaranthus spinosus, Anocardium occidentale, Averrhoa bilimbi, Casuarina equisetifolia and Citrus grandis verum these plants also showed significant antibacterial and antiviral activity [120]. Terminalia catappa methanol leaf extracts and Eucocera agallocha are two essential phytochemicals delivering good results against S. agalactiae in tilapia farms in Malaysia [121,122].

Salvia deserta Schang is an herbaceous perennial plant with numerous diterpenoids that showed antimicrobial activity against S. iniae and other Streptococcus spp in fish [123]. Four diterpenes (Taxodione, Ferruginol, 7-O-Acetylhorminone, and Horminone) from the root of this plant have optimum IC₅₀ and are not harmful to any animal application. Further in vivo and in-silico approaches can explain better this diterpene’s effectiveness against S. iniae in Asian sea bass.

8. Nutrigenomics and dietary supplementation for disease treatment

Antimicrobials should be minimally used in aquaculture, according to the current consensus, as they may lead to the development of microbial resistance [124,125]. Since fish health has become a major concern in aquaculture industries, many feeds have become functional feeds, which are considered to enhance fish health, reduce disease outbreaks, and increase post-infection recovery [126]. The effects of diet on the immune system are becoming apparent because of emerging omics technologies such as transcriptomics (microarray and RNA-seq) and proteomics. Modules of genes can be used to show how both local (intestinal) and systemic immune function is affected utilizing molecular pathway enrichment analysis. Although significant progress has been made in defining alterations in host immune function, additional study is needed to fully comprehend the interaction between fish nutrition, the gut microbiome, and the immune system.

Due to its antioxidant efficacy and diverse biological roles, astaxanthin has considerable applications in the modern aquaculture business as a feed ingredient and nutritional supplement (e.g., immune system boosting, stress alleviation, and survival) [127]. Researchers studied the effects of astaxanthin supplementation on hematopoiesis, nonspecific immunity, and disease resistance in Asian seabass infected with the virulent V. alginolyticus, with a focus on dose-response relationships and variations over time after infection [128]. When given diets with increasing supplemental dosages of astaxanthin, supplemented fish showed significant improvements in hematological parameters (WBC and RBC counts, as well as hemoglobin and hematocrit levels). Furthermore, increasing dietary astaxanthin doses improved sea bass serum’s biochemical profile (aspartate aminotransferase, alanine aminotransferase, glucose, cortisol, cholesterol, and triglyceride contents), leading to improved welfare. The immune defense systems of fish (lysozyme activity, phagocytotic activity, respiratory burst activity, and total serum immunoglobulin) were also considerably stimulated following astaxanthin ingestion [128].

With considerable success, researchers have recently concentrated on the role of organic acids and their salts in preventing and controlling infections in aquaculture. Organic acids have been shown to have a strong antimicrobial impact when challenged with the pathogenic bacterium Streptococcus agalactiae, as well as a high potential to benefit tilapia development, nutrient utilization, and disease resistance. Lipophilic organic acids are those that have not been dissociated and can readily pass through the plasma membrane of bacteria. Organic acids break down into anions and protons once within the cells, where pH values are normally more neutral than outside. Additionally, autolyzed yeast immune-stimulant compounds (which contain the cell walls and accessible nutrients) improved the survival rate of Asian sea bass after a bacterial challenge with S. iniae. The entire blend of immuno-stimulants in autolyzed yeast is
more effective than a single-glucan or nucleotide treatment (Gephart et al., 2020).

The effects of fasting on fish immunity are poorly known and vary by species. Farmed fish are regularly exposed to short-term food limitations (fasting) as part of a seasonal feeding cycle or in response to overproduction or disease outbreaks [129]. Pathogen exposure in fish begins shortly after they emerge from their protective chorions and is amplified during the mouth and gut opening stages and when external feeding begins [130]. Because their nutritional state heavily influences on fish’s ability to resist diseases and cope with stress, the need for dietary treatments to promote fish health has become generally acknowledged as critical to the industrial long-term viability.

Supplementing with in-feed plant-derived additives (1 % immunostimulant and 3 % anti-attachment compound), glucosinolates (GLs, 7.3 mmol/g, and 26.4 mmol/g), additives (nucleotides, mannooligosaccharides, fructo oligosaccharides, vitamin C, and vitamin E), and fasting helps control bacterial, viral, and parasitic infections in aquaculture [131–133]. Although the relationship between nutrition and the immune system is well understood, basic and applied research on the relationships between diet and health in fish, especially aquaculture species, has lagged. The latest omics tools, particularly transcriptomics combined with complete genome sequences, hold immense promise for studying the intricate link between fish nutrition and immunology in health and disease. The effects of nutrition on the development of the digestive and immune systems in the early life stage are yet unknown. Such research is critical because it may lead to the development of nutritional programming and intestinal tolerance strategies at the time of the initial feeding. Understanding both transgenerational epigenetic regulation of immune gene expression as well as life-long epigenetic control of immune gene expression established during the initial feeding will be required to comprehend the relationship between nutrition and immune system ontogeny. Fig. 4 elucidates the concept of the tetrad, highlighting the emerging paradigm of nutrigenomics as a promising alternative to traditional antimicrobial drugs in addressing health challenges, promoting a holistic approach to sustainable health management in aquaculture.

9. miRNAs research prospects in antibacterial immunity of Asian sea bass

miRNAs are also important in the control of the antibacterial immune response in fish, according to research. Through complementary pairing with the mRNA, miRNAs can inhibit the target gene adversely. miRNAs are involved in the metabolism of glucose, lipids, and cholesterol in fish and govern the production of long-chain polyunsaturated fatty acids. They can boost fish species’ immunological response and help bacteria or viruses escape the immune system. Targeting pattern recognition receptors and downstream signaling pathways is one major component of miRNA regulation on fish immunity [134].

The expression of miR-203 was shown to be upregulated in the host, suggesting that it may limit the activation of NF–B signaling by targeting the expression of IRAK4, preventing the development of excessive inflammatory reactions following V. anguillarum disease [135]. Fang et al. (2019) discovered that following A. hydrophila infection, the expression of CiGadd45g was up-regulated in several

Fig. 4. The tetrad is based upon the role of nutrigenomics as an alternative to antimicrobial drugs.
tissues of grass carp, and miR-429b could control CiGadd45g and its downstream genes, limiting the activation of immune-related cytokines (p38 and JNK) and proinflammatory cytokines (IL-8, IFN-1, and TNF-α) [136]. Using microRNAs, bacteria may control and evade the host’s immunological response in some instances. Researchers discovered that E. tarda infection may up-regulate the expression of polmiR-194a in Japanese flounders. IRF7 expression was selectively inhibited by pol-miR-194a, which suppressed type I interferon sensitivity. As a result, enhanced pol-miR-194a expression may assist bacteria in avoiding the host immune response [137]. Researchers discovered that the expression of pol-miR-171 dropped during 48 h of bacterial infection and rose considerably two days later in the same infection scenario. Because PoFAM49B is a pol-miR-target new 171 gene, it can inhibit gram-negative bacterial development as well as block the synthesis of key apoptotic genes, preventing the cell from dying. By targeting PoFAM49B, pol-miR-novel 171 may influence apoptosis and bacterial infection [138]. According to certain research, miRNAs have been linked to antibacterial immunity in various grass carp clones. Xu et al. (2015) found differences in miRNA expression between A. hydrophila-susceptible grass carp (SGC) and A. hydrophila-resistant grass carp (RGC), with five miRNAs being significantly expressed in SGC. It was discovered that the nifI3-6 and thr4 genes were direct targets of let-7i and cid-miRn-118, and it was hypothesized that several miRNAs play a role in grass carp antibacterial immunity through target prediction [139]. Although a study on miRNAs and bacterial illnesses in Asian bass is needed to understand the harmful effect of pathogens such as V. anguillarum, A. hydrophila, F. columnare, and E. tarda, understanding miRNAs can aid in the long-term sustainability of Asian sea bass (L. calcarifer) aquaculture.

10. Biosensor for detection of fish bacterial pathogens

The identification of harmful microorganisms that cause infections and diseases in fish is also critical in aquaculture. In earlier studies, immunomagnetic separation was created as a sensitive magnetic separation approach for concentrating bacteria in fish samples [140]. Scientists used high gradient immunomagnetic separation to boost the detection limit in the water body (HGIMS) [141]. Magnetizing a thin metal magnetic material, such as stainless steel wire, serves as a filter for HGIMS [141]. The microcantilever sensor, amperometric sensor, potentiometric sensor, and surface plasmon resonance (SPR) biosensor, as well as lateral flow tests, are all used to target viral RNA [142] or the bacteria cells [143], these are some other categories of biosensors. Identification of bacteria cells by biosensors eliminates the need for extraction and may be more suitable for onsite use if the sensitivity meets the requirements. The lateral flow test is the most portable type of biosensor since it incorporates sample separation, interaction, and detection into a single chromatographic strip. The SPR sensor study is notable for its attempt to screen an aptamer for Vibrio parahaemolyticus and its reasonable selectivity for V. parahaemolyticus compared to a few others, such as E. coli, V. Fischeri, and S. Soneti. Culture-based approaches represent the “gold standard” in bacteria identification among the methods. The sensitivity of molecular-based approaches is higher, but they require specialized and expensive equipment. Fast, dependable, simple to use, sensitive, and technologies are always in high demand for usage in the field to forecast and respond to epidemics. Biosensors, if they can increase their performance, offer a lot of potential [144], especially for bacterial disease identification in sea bass farms.

11. Quorum sensing and quorum quenching for bacterial diseases

Quorum sensing (QS) is an intercellular communication phenomenon first found in bacteria. A QS system consisting of QS signal molecules and regulatory protein components might govern the physiological features and virulence gene expression of bacterial pathogens. As a result, blocking QS might be a novel way to fight diseases. QS inhibitors (QSI) can be collected from various sources, including tiny chemical compounds and quorum-quenching enzymes, in both marine and terrestrial environments [145]. QS systems have been studied in-depth in a variety of bacterial species, most notably Vibrio. Bacteria release signal molecules in QS systems in a cell density-dependent manner, and signal transduction is induced by cascaded QS regulatory proteins [146]. Multiple phenotypic and key physiological activities of bacteria would be regulated by signal molecule-mediated QS, including cell metabolism, virulence factor release, stress response, and so on [147]. QS-mediated virulence expression, biofilm formation, and pathogenic bacteria colonization have recently sparked widespread concern in aquaculture. Because some bacteria are multidrug-resistant, QS disruption or suppression appeared to be a viable technique for combating pathogen infection. Blocking the creation of signaling molecules, degradation of signaling molecules, and hindering the binding of signaling molecules to receptor proteins in QS pathways are the three basic approaches to interrupting QS pathways.

According to Romero et al. (2010), T. maritimum, a fish pathogen that can cause tenacibaculosis in Asian sea bass, may create an acylase that targets C10-HSL [148]. After degradation, the acidity of the culture medium did not allow the recovery of C10-HSL, indicating acylase-type degradation activity. Even if the physiological processes under the control of AHL-mediated QS in T. maritimum need to be further described. Weiland-Bräuer et al. (2015) looked for ORFs in marine Eukarya-associated bacterium isolates that conferred quorum quenching action. Photobacterium sp., Pseudoalteromonas sp., and Vibrio parahaemolyticus ORFs encoded proteins with unknown functions or functions that differed from recognized lactonases or acylases. The functions of these new proteins are still unknown [149]. Suppressing the production of signal molecules is another method for inhibiting QS. In Aeromonas hydrophila, Kozlova et al. (2012) discovered an S-ribosyl homocysteinase (luxS)-based autoinducer 2 (AI-2) QS system [150]. In the septicemic mouse model, the luxS isogenic mutant of A. hydrophila showed altered dynamics and architecture of biofilm development, decreased motility, and increased pathogenicity. A. hydrophila contains an AHL-based QS system in which ahl and ahlR act as luxI and luxR homologs, in addition to the production of AI-2 through the luxS gene [151]. According to previous research, sodium houttuynfate reduces QS activity and its pathogenicity in A. hydrophila via downregulating ahl. Although quorum inhibition holds great promise in the treatment of fish diseases, it must be balanced against the risk of bacterial resistance.

Researchers recently discovered that aqueous extracts of medicinal rhizome plants inhibited A. hydrophila growth and quorum
Asian seabass nursery rearing is endangered by bacterial infections, which may result in significant death rates and financial losses. Adding probiotics to the culture water [154] and then combat potentially pathogenic microorganisms [166]. This has grown fairly popular in hatcheries, with some farms even using immunostimulants to improve innate immunity and, thus, resistance to diseases in young fry can be beneficial [165]. Supplementation with intracellular actions, such as florfenicol, have shown to be effective in treating PBD on some farms. Because diseased fry frequently has minimal clinical indications or abnormalities, determining the actual origin of sickness can be challenging. As a result, for effective disease prevention and to avoid the emergence of antibiotic resistance, treatment must be preceded by correct laboratory disease sequencing. They were successful in degrading both short and long-chain AHLs associated with many pathogenic Vibrio spp. in fish, including N-[(RS)-3-Hydroxybutyryl]-N-Hexanoyl-l-homoserine lactone (3-oh-C4-HSL), -l-homoserine lactone (3-oh-C4-HSL) N-[(Ketocaproyl)-l-homoserine lactone (C6-HSL), N-homoserine lactone (3-oxo-C6-HSL), -l-homoserine lactone (3-oxo-C6-HSL) (3-Oxodecanoyl)-l-homoserine lactone (3-oxo-C10-HSL), -l-homoserine lactone (3-oxo-C10-HSL) (3-Oxotetradecanoyl) -lactone of N-homoserine (3-oxo-C14-HSL). Using the well-diffusion method and thin-layer chromatography, five QQ isolates (QQIs) from the Bacillus and Shewanella genera showed a solid capacity to digest both synthetic and natural AHLs generated by V. harveyi and V. alginolyticus (TLC). Bacillus thuringiensis QQ1 and Bacillus cereus QQ2 were found to have suitable probiotic features, barramundi safety, and antibiotic sensitivity, and might be employed to reduce vibriosis in Lates calcarifer [153].

12. Disease management of sea bass at the farming level

Outbreaks of disease at the farming level are known to be directed by interactions between the host, the ecosystem, and microorganisms [154]. While major infections are frequently linked to specific aquatic viruses, little is recognized about the risk factors that lead to epibricks of fish disease. Increased output and fewer disease outbreaks have resulted from the formulation of appropriate management systems related to risk factor assessments in shrimp farms [155,156]. Stressful situations, such as net transfers, recent handling, or low water quality, frequently result in disease epidemics [157]. Diseases are frequently caused by infectious agents found in the culture conditions, including the Vibrio species or the parasite Trichodina spp. [158]. Feeding, culture conditions, and farming procedures all have an impact on the health and immunological system of the fish host [159]. While additional research is needed to enhance the detection and management of specific diseases, common health management measures should be implemented at different phases in the culture of sea bass to reduce epidemics [154].

12.1. Hatchery

Aquaculture farms have seen fewer cases of potbelly disease (PBD) since they reduced the number of fish in their tanks and cleaned them more thoroughly. By lowering the number of fish per unit of water volume and keeping the environment clean and healthy, the chance of PBD breakouts and the bad effects on fish health and growth that come with them can be reduced [160,161]. With a decreased stocking density and strict sanitation, aquaculture farms reported fewer cases of potbelly disease (PBD) [154]. Even though the role of vertical or horizontal transmissions in PBD is undetermined, it has been noticed that lower stocking densities may be linked to lower disease outbreaks due to reduced horizontal transmission. Dead fish should be removed from rearing tanks daily to help lower the load of highly contaminated substances and thus disease transmission [162]. In hatcheries, all-in-all-out stocking management will allow for proper disinfection among batches of spawn [163]. During the hatchery phase, weaning fry from a live diet to an artificial pellet is traumatic. Certain stressful events can reduce fry immunity and make them more susceptible to infection [157]). Modifications in the feed supplied to fry must be made gradually and closely examined to assure that they are getting enough to eat. Micro diets have been shown to help transition from life to inert feed and reduce weaning-related death [164]. Several experiments have shown that using immunostimulants to improve innate immunity and, thus, resistance to diseases in young fry can be beneficial [165]. Supplements are currently incorporated throughout many commercial fish feeds, as they were shown to maintain a balanced gut microbiome and then combat potentially pathogenic microorganisms [166]. This has grown fairly popular in hatcheries, with some farms even adding probiotics to the culture water [154]. T. maritimum was shown to be more viable in sterile seawater than in natural seawater, implying that the natural microbial community in natural seawater may help keep this bacterium in account [167]. Antibiotics with intracellular actions, such as florfenicol, have shown to be effective in treating PBD on some farms. Because diseased fry frequently has minimal clinical indications or abnormalities, determining the actual origin of sickness can be challenging. As a result, for effective disease prevention and to avoid the emergence of antibiotic resistance, treatment must be preceded by correct laboratory disease identification. Vaccination is effective in controlling bacterial and other infectious diseases in experimental studies [168,169].

12.2. Nursery

Bacterial diseases may threaten the health and survival of juvenile fish, which provide serious problems for Asian seabass nurseries. Asian seabass nursery rearing is endangered by bacterial infections, which may result in significant death rates and financial losses. V. harveyi and Streptococcus spp. are the two most prevalent bacterial infections infecting Asian seabass nurseries. If these germs are not managed, they may cause fatal diseases that kill many people [170]. Another significant bacterial disease that affects Asian seabass nurseries is streptococcosis. The main culprits are Streptococcus species, especially S. agalactiae and S. iniae. Systemic infections brought on by streptococcal infections may result in significant fatality rates and financial losses [171]. The bacterial infections that affect Asian seabass might spread more quickly and be more severe in the nursery environment [171]. Fish may get stressed and have
their immune systems weakened by overpopulation and poor water quality, increasing their susceptibility to diseases. The introduction of contaminated or carrier fish, lack of biosecurity controls, and poor sanitation procedures may also influence disease outbreaks in nursery facilities.

Managing bacterial diseases in Asian seabass nurseries requires a comprehensive approach. Implementing strict biosecurity protocols, including proper quarantine measures and screening of incoming fish, is crucial to prevent the introduction and spread of pathogens. Good water quality management, optimal stocking densities, and adequate nutrition are also essential to minimize stress and enhance the fish’s immune response [172]. Most excellent farming methods described in the hatchery stage will also be useful in the nurseries. Furthermore, due to the rapid increase and the resulting significant differential in fish size, cannibalism is a significant issue in the production of sea bass during the nursery and initial grow-out periods [154]. It can cause serious mortality, either directly or indirectly, and skin irritation on the head and tail, leading to bacterial transmission. To eliminate size differences, early nursery stages (20–35 mm body length) should be graded every third to four days, whereas larger fish (50–100 mm body length) should be evaluated every seven days [164]. External damage from the grading procedure can result in secondary bacterial infections such as tenacibaculosis and vibriosis. Therefore, assessment must be done using well-thought-out procedures. The oxygen level in intensively filled tanks with rapidly growing fish can quickly go under four ppm, increasing the organic load. More excellent thermal rotations keep DO levels in check and prevent the buildup of organic material and bacteria that can cause disease occurrences [173]. In higher temperatures with less dissolved oxygen, streptococcosis and tenacibaculosis were linked to a greater mortality rate [174,175]. Avoiding stressful handling techniques, keeping appropriate aeration, and delaying feed until mortality has decreased are all acceptable ways to manage diseases with pronounced effects on blood oxygen. Despite antibiotic medication, high mortality is common in Streptococcus at nursery and grow-out stages. For streptococci in fish weighing less than 3 g, a monovalent inactivated vaccine or bath immunizations may be an alternative. Intraperitoneal immunizations are routinely given in fish weighing more than 20 g to extend protection during the grow-out period. Vaccines, on the other hand, may not cross-protect against distinct streptococcus biotypes, hence appropriate diagnosis is essential for creating a successful vaccination program [176]. Tenacibaculosis vaccines are currently unavailable on the market. It is critical to check nursery fish for any underlying diseases that could become clinical during shipment, transfer to saltwater, or other handling operations [154].

12.3. Grow-out

Before being stocked into marine cages, fish should be inoculated against major infections that can develop on every aquaculture site. Fish in sea cages might be challenging to treat. Bath treatments are both time-consuming and distressing for the fish. Because fish are typically not fed when the disease is detected, oral treatments are difficult to administer [154]. In disease outbreaks, reducing mortality is vital to reducing the vulnerability of other fish to potential pathogens. Dead fish attract giant predators and cause net damage if not removed. While transfers and grading might cause diseases like vibriosis and tenacibaculosis, these husbandry practices are unavoidable. Farmworkers must follow simple fish health so that these operations may be carried out with the least stress on the species [154]. With adequate planning and the right equipment, handling time may be cut in half, and conditions can be maintained at their best with optimum aeration, good water exchanges, and anesthetics. Cannibalism can be a concern in L. calcarifer during the early stages of growth, grading should be done until the species weigh about 300 g [154]. Cannibalism and direct mortality will occur due to the large size gap, as will predisposition to opportunistic bacterial or parasitic pathogens. Streptococcosis outbreaks in L. calcarifer were linked to adverse weather conditions and increased suspended solids in Lake Argyle, Western Australia, and the Northern Territory, Australia [25]. It has been proven that suspended solids can behave as physical carriers of infectious pathogens [177]. Streptococcus outbreaks could be reduced by locating prospective marine cage locations farther from significant river runoff. The efficiency of Streptococcus iniae immunization in fish has been demonstrated to be reduced by parasitism or immunosuppression [178]. Parasite control programs and fish husbandry should back up vaccination effectiveness measures that reduce stress. Even though vaccines are accessible, many research groups hold opposing perspectives on the efficacy of Streptococcus vaccines [179]. Therefore, farms in Southeast Asia that implemented S. iniae immunization saw a reduction in L. calcarifer mortality. According to anecdotal data from established farms in Southeast Asia and Australia, stocking thirty to forty gm barramundi in sea cages is linked to a greater survival rate. Marine fish species’ adaptive immune systems are known to mature significantly later than their freshwater competitors. Studies of disease outbreaks in farmed L. calcarifer in Asia have revealed age-related vulnerability to disease, further validating the stocking of older, more immune-capable fish into marine cages [180]. Because each farm site frequently has multiple age groups of L. calcarifer, older carrier fish can be a source of infectious pathogens. To facilitate disease control in the future, some thought should be given to maintaining sites as single-year classes.

13. Prospects of rapid genomic detection in sea bass Aquaculture disease management

Pathogen detection must be done quickly to control disease in aquaculture. The use of antibody probes and DNA primers/probes in pathogen detection and identification has significantly influenced the development of quick diagnostic procedures. However, a lack of information and instruments to identify fish infections, determine their origins and manage their spread limits development. Whole-genome sequencing reveals how viruses alter and travel through habitats, allowing evidence-based biosecurity to be implemented to reduce disease effects. Sequencing services performed off-site are costly and cause significant delays.

WorldFish (https://www.worldfishcenter.org) has developed a compact “lab-in-a-backpack” for pond-side identification and quantification of diseases affecting tilapia in partnership with the University of Queensland and Wilder lab. A portable DNA extraction equipment, a hand-held DNA sequencer (MiniION), a battery-operated minicomputer (MiniIT), and a user-friendly purpose-built
software suite are all included, with low electricity and internet access; users with no prior training in molecular biology or bioinformatics can identify fish pathogens from both water samples and affected tissues remotely and in real-time. These low-cost methods will enable tilapia breeding, quarantine, and biosecurity facilities to detect disease outbreak-causing organisms in a fraction of the time and money necessary for external laboratory analysis. Although this method has yet to be used in Asian sea bass farming, it has the potential to play a significant role in disease control in the sea bass aquaculture industry.

14. Conclusion and recommendation

Disease incidence is among the most significant determining factors in sea bass farming. Pathogens are easily transmitted between individual fish in intensive aquaculture systems where monoculture or polyculture of fish species are grown at high densities. Despite biosecurity techniques such as isolation, sterilization, and sanitation, and the use of synbiotics, disease-free broodstock, and high-quality feed, which have been demonstrated to minimize disease transmission, they do not always guarantee the complete removal of infectious organisms. Antibiotic use increases the likelihood of pathogens developing drug resistance, rendering therapy unproductive and transferring resistant strains to other bacteria and antibiotic residues into feed. Vaccination is the most eco-sustainable practical strategy for preventing disease outbreaks and excessive antibiotic use in aquatic ecosystems. On the other hand, commercial vaccine development for the sea bass sector is still infancy. Aside from that, researchers should focus on novel strategies based on emerging technology, including nanosensor, image-based machine learning technique, surface drone for early bacterial disease detection, and phytosynthesis drugs as an alternative to antibiotics and nanoparticle immunizations that helps to preserve antigens from low pH hydrolysis in the abdomen.

CRediT authorship contribution statement


Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgments

The author(s) would like to acknowledge Qatar National Library for providing Open Access fees for this work. In addition, the authors extend their appreciation to Taif university, Saudi Arabia, for supporting this work through project number (TU-DSPP-2024-02).

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