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DIETARY SUPPLEMENTATION OF *Sargassum johnsonii* ENHANCES DISEASE RESISTANCE AGAINST *Aeromonas hydrophila* IN *Labeo rohita* FRY

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ABSTRACT

Aquaculture intensification has led to more bacterial diseases, causing significant economic losses and lower productivity. This study examined how adding the brown seaweed *Sargassum johnsonii* to the diet affects the disease resistance of *Labeo rohita* fry against *Aeromonas hydrophila*. Five diets with 0% (T0), 3% (T1), 6% (T2), 9% (T3), and 12% (T4) *S. johnsonii* powder were given to *L. rohita* fry for 60 days. After the feeding trial, fish were exposed to *A. hydrophila* (1×10^6 cells mL^{-1}), and mortality was tracked for ten days. Survival percentage, cumulative mortality, and relative percent survival (RPS) were calculated. The results showed a significant increase in disease resistance in seaweed-fed groups compared to the control. The highest survival (90%), lowest mortality (10%), and greatest RPS (75%) were seen in fish fed the diet with 9% *S. johnsonii*. The control group had the lowest survival (60%) and highest mortality (40%). The findings suggest that adding *S. johnsonii* to the diet boosts the resistance of *L. rohita* fry against *A. hydrophila* infection, with 9% being the best level for improving disease resistance.

Keywords **:** *Sargassum johnsonii*, *Labeo rohita*, *Aeromonas hydrophila*, disease resistance, challenge test, relative percent survival, seaweed supplementation.

Introduction

Aquaculture is one of the fastest-growing food production sectors globally and plays a significant role in ensuring food security and nutritional sustainability. However, disease outbreaks remain a major constraint to aquaculture production, causing substantial economic losses worldwide (FAO, 2024; Bondad-Reantaso *et al*., 2005).

Among bacterial pathogens, *Aeromonas hydrophila* is an important opportunistic bacterium responsible for motile aeromonad septicemia in freshwater fishes. The disease is characterized by hemorrhages, ulceration, septicemia, and high mortality, leading to serious production losses in aquaculture systems (Sawyer, 1986; Das *et al*., 2015). Although antibiotics have been widely used for disease control, their indiscriminate use has raised concerns regarding antimicrobial resistance, environmental contamination, and food safety (Cabello *et al*., 2019). Therefore, the development of natural and sustainable alternatives for disease management has become increasingly important.

Marine macroalgae have gained considerable attention as functional feed ingredients due to their nutritional value and abundance of bioactive compounds. Seaweeds contain polysaccharides,

phenolic compounds, vitamins, minerals, and antioxidants that possess antimicrobial, antioxidant, and immunostimulatory properties (Holdt and Kraan, 2011; Wan *et al*., 2019). Several studies have reported that dietary supplementation of seaweeds improves immune responses, growth performance, and disease resistance in cultured fish species (Reverter *et al*., 2020; Siddik *et al*., 2023).

Brown seaweeds of the genus *Sargassum* are particularly rich in biologically active compounds such as fucoidans, alginates, and phlorotannins. Dietary inclusion of *Sargassum* species has been shown to enhance innate immunity and resistance against pathogenic infections in fish (Yeganeh and Adel, 2019; Negm *et al*., 2021; Zeynali *et al*., 2020). However, information regarding the application of *Sargassum johnsonii* as a dietary immunostimulant in Indian major carps is limited.

Labeo rohita is one of the most important Indian major carps cultured throughout South Asia owing to its fast growth, consumer preference, and economic value (FAO, 2009; Ahammad *et al*., 2022). Therefore, the present study was undertaken to evaluate the effect of dietary supplementation of *Sargassum johnsonii* powder on the disease resistance of *Labeo rohita* fry against *Aeromonas hydrophila* infection through an experimental challenge study.

Materials and Methods

Fresh *Sargassum johnsonii* was collected from the Veraval coast, Gujarat, India. It was washed well to remove impurities and then dried in the sun. The dried seaweed was ground into a fine powder, sieved for uniformity, and stored in airtight polyethylene bags for later use.

The feeding trial took place at the Wet Laboratory, Department of Aquaculture, College of Fisheries Science, Kamdhenu University, Veraval, Gujarat, India. Healthy *Labeo rohita* fry, with an average initial body weight of 1.83 ± 0.001 g, were obtained from a local hatchery and acclimatized before the experiment. A total of 300 fry was randomly placed into 20 plastic tanks (40 L capacity) with 15 fish per tank. Continuous aeration was provided throughout the experiment. The water quality parameters were kept within the optimal range for rohu culture. Five experimental diets with the same nitrogen content were prepared by adding *Sargassum johnsonii* powder at different levels: 0% (T0, control), 3% (T1), 6% (T2), 9% (T3), and 12% (T4). The fish were fed twice daily at 10% of their body weight for 60 days. The feed amount was adjusted periodically based on the biomass.

The pathogenic bacterium *Aeromonas hydrophila* was grown in nutrient broth and incubated at 37°C for 24 hours. The bacterial culture was centrifuged at 3000 rpm for 10 minutes, and the supernatant was discarded. The resulting pellet was resuspended in sterile phosphate-buffered saline (PBS; pH 7.4). The bacterial concentration was adjusted using a spectrophotometer to an optical density (OD) of 0.2 at 456 nm, which is about 1×10^6 cells mL^{-1}. Before the challenge study, the median lethal dose (LD_{50}) of *A. hydrophila* was determined by preparing serial dilutions of the bacterial suspension (1×10^6, 1×10^5, and 1×10^4 cells mL^{-1}) in PBS. Experimental fish were exposed to these doses and observed for 24–48 hours after treatment. A mortality rate of about 50% was recorded at a concentration of 1×10^6 cells mL^{-1}, which was chosen as the challenge dose for the disease resistance study. At the end of the 60-day feeding trial, ten fish from each treatment were randomly selected and injected intramuscularly with 0.1 mL of a bacterial suspension containing 1×10^6 cells mL^{-1} of *A. hydrophila*. The challenged fish were observed for 10 days, and mortality was recorded daily. Survival (%), cumulative mortality (%), and relative percent survival (RPS) were calculated to evaluate the disease resistance of *Labeo rohita* fry fed different levels of *Sargassum johnsonii* supplementation. The experimental data were analyzed using SPSS software. A one-way analysis of variance (ANOVA) was conducted to determine significant differences among the treatment means.

Results and Discussion

The disease resistance of *Labeo rohita* fry fed different levels of *Sargassum johnsonii* was evaluated through an experimental challenge with *Aeromonas hydrophila*. The results revealed improved survival and reduced mortality in all seaweed-supplemented groups compared to the control treatment (Table 1).

The highest survival percentage (90%) was recorded in fish fed the diet containing 9% *S. johnsonii* (T3), followed by T2 (6%) and T4 (12%) with 80% survival. The control group (T0) exhibited the lowest survival (60%). Similarly, cumulative mortality was lowest in T3 (10%) and highest in the control group (40%).

Relative percent survival (RPS) increased with increasing dietary inclusion of *S. johnsonii* up to 9%, reaching a maximum value of 75% in T3. However, a slight decline was observed at the highest inclusion level (12%), where RPS was reduced to 50%.

Table 1 : Survival (%), cumulative mortality (%) and relative percent survival (RPS) of *Labeo rohita* fry following challenge with *Aeromonas hydrophila*.

Treatment	Survival (%)	Cumulative Mortality (%)	RPS (%)
T0 (Control)	60	40	-
T1 (3%)	70	30	25
T2 (6%)	80	20	50
T3 (9%)	90	10	75
T4 (12%)	80	20	50

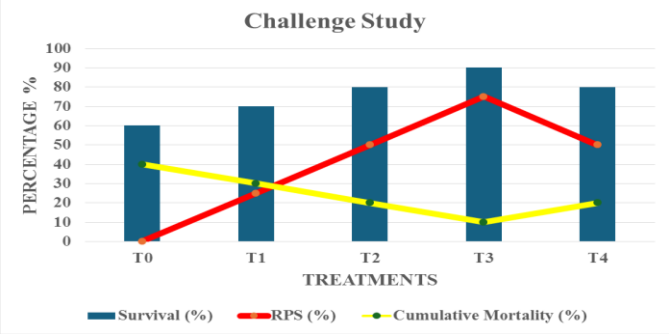

Fig. 1 : Effect of dietary *Sargassum johnsonii* supplementation on *Labeo rohita* fry following *Aeromonas hydrophila* challenge.

The present study demonstrated that dietary supplementation of *Sargassum johnsonii* improved the resistance of *Labeo rohita* fry against *Aeromonas hydrophila* infection. Following bacterial challenge, fish fed seaweed-supplemented diets exhibited higher survival and lower cumulative mortality compared to the control group. Among the dietary treatments, the 9% inclusion level (T3) showed the highest survival (90%), lowest mortality (10%), and maximum relative percent survival (75%), indicating enhanced protection against bacterial infection.

The improved disease resistance observed in the present study may be attributed to the presence of biologically active compounds in *Sargassum johnsonii*, including sulfated polysaccharides, phenolic compounds, vitamins, minerals, and antioxidants. These compounds are known to stimulate innate immune responses and enhance the defensive capacity of fish against pathogenic microorganisms (Holdt and Kraan, 2011; Wan *et al*., 2019). Brown seaweeds of the genus *Sargassum* are particularly rich in fucoidans, alginates, and phlorotannins, which have been reported to possess immunostimulatory and antimicrobial properties.

The present findings are in agreement with the observations of Yeganeh and Adel (2019), who reported improved immune status and disease resistance in juvenile great sturgeon fed diets supplemented with *Sargassum ilicifolium*. Similarly, Zeynali *et al*. (2020) observed enhanced innate immunity and increased expression of immune-related genes in Asian seabass juveniles following dietary inclusion of *S. ilicifolium*. Negm *et al*. (2021) also reported improved immune response and stress tolerance in Nile tilapia fed diets supplemented with *Sargassum aquifolium*.

The higher survival recorded in seaweed-fed groups may be associated with improved physiological condition and enhanced activation of nonspecific immune mechanisms. Previous studies have shown that dietary seaweed supplementation stimulates phagocytic activity, respiratory burst activity, antioxidant defense systems, and leukocyte function, thereby improving resistance against pathogenic infections (Reverter *et al*., 2020; Siddik *et al*., 2023). Such immunostimulatory effects may have contributed to the reduced mortality observed in the present study following *A. hydrophila* challenge.

Interestingly, survival increased progressively with increasing dietary inclusion of *S. johnsonii* up to 9%, whereas no further improvement was observed at 12% inclusion. This suggests that moderate supplementation levels may provide optimal biological benefits, while excessive inclusion may not necessarily result in additional enhancement of disease resistance. Similar trends have been reported in studies evaluating dietary seaweed supplementation in aquaculture species (Shi *et al*., 2019; Hu *et al*., 2024).

Overall, the findings of the present investigation demonstrate that dietary incorporation of *Sargassum johnsonii* can effectively enhance resistance of *Labeo rohita* fry against *Aeromonas hydrophila* infection and may serve as a promising natural alternative to synthetic chemotherapeutics for sustainable aquaculture practices.

Conclusion

The present study demonstrated that dietary supplementation of *Sargassum johnsonii* improved the disease resistance of *Labeo rohita* fry against *Aeromonas hydrophila*. Fish fed the diet containing 9% *S. johnsonii* exhibited the highest survival (90%), lowest cumulative mortality (10%), and highest relative percent survival (75%) following bacterial challenge. The results indicate that *Sargassum johnsonii* can be utilized as a natural immunostimulatory feed additive to enhance resistance against bacterial diseases in freshwater aquaculture. Among the tested inclusion levels, 9% dietary supplementation was found to be the most effective.

References

Ahammad, A. S., Asaduzzaman, M., Ferdusy, T., Islam, M. M., Wong, L. L., Rahman, M. M., ... & Hassan, J. (2022). Morphogenetic diversity of the selected hatchery populations of an Indian major carp (*Labeo rohita*, Hamilton, 1822) in the Mymensingh region of Bangladesh. *Animal Gene*, **25**, 200131.

Bondad-Reantaso, M. G., Subasinghe, R. P., Arthur, J. R., Ogawa, K., Chinabut, S., Adlard, R., Tan Z. and Shariff, M. (2005). Disease and health management in Asian aquaculture. *Veterinary Parasitology*, *132*(3–4), 249–272.

Cabello, F. C., Godfrey, H. P., Tomova, A., Ivanova, L., Dölz, H., Millanao, A., & Buschmann, A. H. (2019). Aquaculture as yet another environmental gateway to the development and globalisation of antimicrobial resistance. *The Lancet Microbe*, **1**(2), e84–e85. https://doi.org/10.1016/S2666-5247(19)30046-8

Das, R., Raman, R. P., Saha, H., & Singh, R. (2015). Effect of *Ocimum sanctum* Linn. (Tulsi) extract on the immunity and survival of *Labeo rohita* (Hamilton) infected with *Aeromonas hydrophila*. *Aquaculture Research*, **46**(5), 1111-1121.

Food and Agriculture Organization (FAO). (2009). *Labeo rohita*. In Cultured aquatic species information programme. FAO Fisheries and Aquaculture Department, Rome. Retrieved from https://www.fao.org/fishery/docs /CDrom/aquaculture/I1129m/file/en/en_roholabeo.htm

FAO. (2024). The State of World Fisheries and Aquaculture 2024; Blue Transformation in action, Food and Agriculture Organization of the United Nations, Rome, Italy. P. 264. https://doi.org/10.4060/cd0683en

Holdt, S. L., & Kraan, S. (2011). Bioactive compounds in seaweed: functional food applications and legislation. *Journal of applied phycology*, **23**, 543-597.

Hu, P., Shi, Q., Wen, Z., Wang, J., & Qin, C. (2024). Potential effects of *Sargassum kjellmanianum* as a dietary supplement on the growth performance, immunity response, antioxidant status, and ammonia resistance of yellow catfish (*Pelteobagrus fulvidraco*). *Aquaculture Reports*, **36**, 102181.

Negm, S. S., Ismael, N. E., Ahmed, A. I., Asely, A. M. E., & Naiel, M. A. (2021). The efficiency of dietary *Sargassum aquifolium* on the performance, innate immune responses, antioxidant activity, and intestinal microbiota of Nile Tilapia (*Oreochromis niloticus*) raised at high stocking density. *Journal of Applied Phycology*, **33**, 4067-4082.

OECD/FAO (2023), OECD-FAO Agricultural Outlook 2023-2032, OECD Publishing, Paris, https://doi.org/10.1787/08801ab7-en [Accessed on 22/06/2024]

Reverter, M., Tapissier-Bontemps, N., Sarter, S., Sasal, P., & Caruso, D. (2020). Moving towards more sustainable aquaculture practices: A meta-analysis on the potential of plant-enriched diets to improve fish growth, immunity and disease resistance. *Reviews in Aquaculture*, **12**(3), 1795–1813. https://doi.org/10.1111/raq.12485

Sawyer, T. K. (1986). Morphological and biochemical characteristics of *Aeromonas hydrophila*. *Microbiology Reviews*, **50**(3), 384–409.

Seyedalhosseini, S. H., Salati, A. P., Mozanzadeh, M. T., Parrish, C. C., & Shahriari, A. (2023). Effects of dietary seaweeds (*Gracilaria spp*. and *Sargassum spp*.) on growth, feed utilization, and resistance to acute hypoxia stress in juvenile Asian seabass (*Lates calcarifer*). *Aquaculture Reports*, **31**, 101663.

Shi, Q., Rong, H., Hao, M., Zhu, D., Aweya, J. J., Li, S., & Wen, X. (2019). Effects of dietary *Sargassum horneri* on growth performance, serum biochemical parameters, hepatic antioxidant status, and immune responses of juvenile black sea bream *Acanthopagrus schlegelii*. *Journal of Applied Phycology*, **31**, 2103-2113.

Siddik, M. A. B., Howieson, J., Fotedar, R., & Partridge, G. J. (2023). Use of seaweed and seaweed-derived products in aquafeeds: Effects on growth performance, immune response and disease resistance of fish. *Aquaculture*, **562**, 738746.https://doi.org/10.1016/j.aquaculture.2022.738746

Wan, A. H. L., Davies, S. J., Soler-Vila, A., Fitzgerald, R., & Johnson, M. P. (2019). Macroalgae as a sustainable aquafeed ingredient**.** *Reviews in Aquaculture*, **11**(3), 458–492. https://doi.org/10.1111/raq.12241

Yeganeh, S., & Adel, M. (2019). Effects of dietary algae (*Sargassum ilicifolium*) as immunomodulator and growth promoter of juvenile great sturgeon (*Huso huso Linnaeus*, 1758). *Journal of Applied Phycology*, **31**(3), 2093-2102.

Zeynali, M., Nafisi Bahabadi, M., Morshedi, V., Ghasemi, A., & Torfi Mozanzadeh, M. (2020). Replacement of dietary fishmeal with *Sargassum ilicifolium* meal on growth, innate immunity and immune gene mRNA transcript abundance in *Lates calcarifer* juveniles. *Aquaculture Nutrition*, **26**(5), 1657-1668.