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A PRELIMINARY STUDY ON THE GUT MICROBIAL COMPOSITION OF SELECTED CARP SPECIES

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ABSTRACT

This study explored bacterial diversity in the gastrointestinal tract of the Indian Major Carp (*Labeo catla*) using culture-dependent techniques. Results showed notable differences in bacterial load between the foregut and hindgut. Five bacterial strains were isolated, four from the hindgut and one from the foregut. Aeromonas (CC3) was the most prevalent strain, with colony counts from 2 to 53 across plates. Morphological and Gram stain analyses identified the isolates as belonging to five genera: *Serratia, Staphylococcus, Aeromonas, Bacillus*, and *Pseudomonas*; 75% were Gram-negative, and 25% were Gram-positive. Biochemical tests revealed genus-specific traits, with Aeromonas exhibiting the widest metabolic activity, including positive reactions for citrate, urease, catalase, oxidase, and sulphur reduction. These findings offer foundational insights into the focal animal's gut microbiota, emphasising the dominance of hindgut bacterial populations and the potential functions of certain strains.

INTRODUCTION

The gut microflora of riverine fish is a complex and dynamic community of microorganisms, mainly bacteria, along with contributions from archaea, fungi, protozoa, and viruses. These microbial populations differ notably among species, habitats, and developmental stages. The dominant bacterial groups typically include Proteobacteria, Firmicutes, Bacteroidetes, and Actinobacteria, with genera such as Aeromonas, Pseudomonas, Bacillus, and Clostridium frequently reported (1, 2). Many studies have examined the indigenous microflora of fish, especially the microbial ecology of the digestive tract (3, 4, 5). However, the microbial composition of the gastrointestinal tract varies greatly and is influenced by dietary changes (6, 7, 8). Studies show that facultative anaerobes, such as *Enterobacteriaceae, Aeromonas, Acinetobacter, Pseudomonas,* and *Flavobacterium*, are common, whereas *Bacteroides, Clostridium,* and *Fusobacterium* are obligate anaerobes (3, 4).

Microflora in the fish gut play essential roles in host physiology, including digestion, nutrient absorption, immune modulation, and pathogen resistance. Freshwater fish, especially those from river systems, often harbour more diverse microbiota than their marine counterparts due to fluctuating environmental conditions and dietary variability (9). In riverine fish, gut microbes are vital for adapting to seasonal changes in food availability and water quality. For instance, *Bacteroides* and *Clostridium* species ferment plant material in omnivorous fish. Conversely, herbivorous fish tend to have higher proportions of cellulolytic bacteria. In carnivorous fish, *Pseudomonas* and *Aeromonas* are involved in protein degradation (10). Moreover, the gut microbiota acts as a barrier against pathogens through competitive exclusion and the production of antimicrobial compounds.

In recent years, many studies have examined the indigenous intestinal microflora of various fish species (7, 11, 12, 13, and 14). These studies mainly concern the symbiotic relationship between fish and intestinal microflora, or the relationship between diet and intestinal microflora in various fish species. However, only a few studies have examined the gut microbiome of Indian Major Carps. Carps have been the most widely cultured species in India's freshwater aquaculture for centuries and account for about 70% of the sector. In 2022, Mondal carried out an in-depth investigation into the digestive enzyme-producing bacteria associated with the Indian major carp, Rohu. (15). Sundaray and colleagues (2025) successfully isolated members of the phyla *Proteobacteria*, *Fusobacteria*, *Bacteroidetes*, and *Firmicutes* from the intestinal microbiota of catla (16). The authors observed significant sex-specific differences, with higher Fusobacteria levels in males and higher γ-Proteobacteria levels in females. The gut microflora of fish varies significantly from place to place. Geographical location, habitat differences, and environmental factors, including water temperature, salinity, dissolved oxygen, and seasonal changes, drive distinct microbial compositions across populations (17). This study aims to explore the microbial communities in the gastrointestinal tracts of Indian Major Carp (*Labeo catla*) found in the southern part of Tamil Nadu. *L. catla*, commonly known as catla, is one of the three major Indian carps (along with rohu and mrigal) and a key species in freshwater aquaculture across South Asia, especially in India. This fish thrives in riverine and pond environments, growing rapidly to over 1 kg within the first year, and accounts for about 20-30% of polyculture systems due to its surface-feeding habits on zooplankton and insects. The key findings of this investigation open new avenues for scientific research, particularly regarding the microbiota's roles in digestion, nutrient absorption, immune modulation, and pathogen resistance. This promotes further growth in freshwater aquaculture.

MATERIALS AND METHODS

The present study was conducted from August 2024 to December 2024 at the Centre for Behavioural and Immuno-ecology, PG and Research Department of Zoology, St. John's College, Palayamkottai. To explore the gastrointestinal microbiota, healthy, active *Labeo catla* fingerlings were collected using mosquito nets in Sirukulam. The captured fish were immediately transferred to St. John's College. Subsequently, five fingerlings were selected and

starved for 12 hrs to clear their digestive tracts. The starved fingerlings were used for the experiment.

The experimental animals were euthanised, placed on a sterile dissection board, pithed, and the body surface was cleaned with 1% iodine solution (Trust & Sparrow, 1974). Then, the animals were dissected aseptically, and the gastrointestinal tract was removed and thoroughly rinsed in sterile saline solution (0.9%). The digestive tract was divided into two regions (foregut and hindgut) and was homogenised separately with 5 ml of pre-chilled 1% NaCl solution.

The homogenate was filtered and collected in a sterile vial, labelled and stored at -20 °C. For bacterial isolation, the collected filtrate of each fish was used for serial dilution. After serial dilutions, 100 µL of the dilutions $10^{\wedge 4}$, $10^{\wedge 5}$, and $10^{\wedge 6}$ were spread using a sterile glass spreader onto pre-solidified Nutrient Agar (Srichem – SRL) petri plates in triplicate. The plates were incubated at 37°C for 48 hours.

After 48 hrs of incubation, plates with well-separated colonies were counted. The total viable bacterial colonies and individual bacterial colonies were counted using a Microprocessor-based Colony Counter (Deep Vision, Model No. 1363), and plates showing 30-300 colonies were considered countable. The bacterial population was quantified as colony-forming units per µl (cfu).

The isolated bacterial strains were subjected to morphological and biochemical characterisation. Morphological identification was conducted using the following criteria: 1. Shape of the colony, 2. Edge aspects, and 3. Colour. The Gram stain test was performed to determine whether the bacteria were Gram-positive or Gram-negative. Digital images of various isolated bacterial species were captured using an Olympus C220i Trinocular Microscope equipped with a MagVision 5 MP camera. Additionally, the isolated bacterial strains were identified and confirmed in accordance with Bergey's Manual of Systematic Bacteriology, Volumes I and II. After that, the following biochemical tests were performed.

Catalase Test

Catalase is the enzyme that cleaves hydrogen peroxide (H2O2) into H2O and O2. Catalase production was tested by adding 1 ml of an aqueous H_2O_2 solution (10%, v/v) to a 24-hour-old nutrient agar plate. The brisk evolution of gas (oxygen) bubbles indicated the presence of catalase in the culture.

Sulphur Production Test

H_2S production was tested on the slants of peptone-yeast extract-iron agar medium. The slants were inoculated with bacteria and incubated for 48 hours at 37 ± 1 °C. H_2S production was detected by inserting lead acetate paper strips into the neck of the culture tube. Blackening of the strip was a positive indication of H_2S production.

Oxidase Test

Cytochrome oxidase was detected in an 18-hour-old nutrient broth culture. 5 ml of the broth culture was taken in a clean tube. 0.3 ml of p-amino-dimethylaniline oxalate (1%) and 0.2 ml of α-napthol (1%) were added, and the mixture was shaken vigorously. The appearance of a blue colour indicated a positive test.

Citrate Utilisation Test

Bacteria were grown in citrate medium (Koser's synthetic medium), and the growth, as evidenced by turbidity, indicated the utilisation of citrate as the sole carbon source.

Urease Test

Inoculate a microorganism onto a urea-containing medium and incubate it, often at 37°C. A positive result is indicated by a colour change from yellow to pink or magenta, indicating that the urease enzyme has catalysed the hydrolysis of urea to ammonia, thereby raising the pH. A negative result remains yellow.

Motility Test

Stab a single colony from a bacterial culture into a semi-solid agar medium using a sterile straight needle. After incubating the tube for 24-48 hours, check for growth spreading away from the original stab line, which indicates motility. A non-motile organism will show growth only along the inoculation line.

RESULTS

The study was conducted to identify potential bacterial strains in the gastrointestinal tract of Indian Major Carp (*Labeo catla*).

Colony Enumeration

In the present investigation, the total viable bacterial load was determined in various dilutions prepared from the foregut and hindgut regions of the gastrointestinal tract. All dilutions showed considerable variation in bacterial load. The mean viable bacterial count observed in $10^{\wedge 4}$ dilutions of the foregut and hindgut regions was 12.72 ± 5.91 and 20.32 ± 9.15 CFUμ/L, respectively. However, the mean viable bacterial counts in 10^6 dilutions of the foregut and hindgut regions were 8.31 ± 4.33 CFU/μL and 4.21 ± 1.62 CFU/μL, respectively. Generally, the highest bacterial populations were observed in the hindgut regions compared to the foregut. Statistical analysis showed a significant difference in the occurrence of bacterial colonies between the foregut and hindgut regions (ANOVA, $F(2, 24) = 1.957$, $p = 0.015$).

Table 1: Total bacterial count in the foregut and hindgut regions of *Catla catla*

S. No	Regions of the Digestive Tract	Dilutions	Bacterial Load (CFU/μL)	
			Mean	SD
1.	Foregut	$10^{\wedge 4}$	12.72	5.91
		$10^{\wedge 5}$	7.33	2.15

		$10^{\wedge 6}$	4.21	1.62
2.	Hindgut	$10^{\wedge 4}$	20.32	9.15
		$10^{\wedge 5}$	13.54	4.91
		$10^{\wedge 6}$	8.31	4.33

Morphological Characterisation of Bacterial Strains

In the present study, five bacterial strains were isolated from the gastrointestinal tract of carp. They were *Serratia* (CC1), *Staphylococcus* (CC2), *Aeromonas* (CC3), *Bacillus* (CC4) and *Pseudomonas* (CC5). *Serratia* species are rod-shaped, motile, peritrichously flagellated, and often produce smooth, moist colonies with a characteristic red pigment (prodigiosin). *Staphylococcus* species form grape-like clusters, are non-motile, and typically form round. *Aeromonas* species are also rod-shaped and motile with polar flagella; they form smooth, convex colonies. *Bacillus* species are large, often form chains, and can form endospores; their colonies are usually dry, irregular, and may have a ground-glass appearance. Finally, *Pseudomonas* species are rod-shaped and produce flat, irregularly edged colonies. The morphological characteristics of isolated colonies are given in Table 2. Among the isolated strains, four strains were isolated in the hind gut region, and only one strain was recovered in the foregut region. The CC3 isolate was dominant compared to the others. The minimum and maximum number of CC3 isolates recovered in all plates was 2 to 53. The minimum and maximum of respective bacterial strains recovered in all the culture plates are given in Figure 1. The results of Gram staining revealed that 75% of isolates were Gram-negative and the remaining 25% were Gram-positive (*Staphylococcus* and *Bacillus*) (Table 3).

Table 2: Morphological characteristics of isolated bacterial colonies

S. No.	Isolate Code	Morphology		
		Shape	Edge	Colour
1.	CC1	Rod	Smooth/Irregular	Red
2.	CC2	Round	Smooth	Golden yellow or white
3.	CC3	Rod	Circular	Greyish
4.	CC4	Round	Irregular, undulate, or lobate	Pale white
5.	CC5	Rod	Flat/Irregular	Greenish blue

Figure 1: Minimum and maximum number of isolated colonies

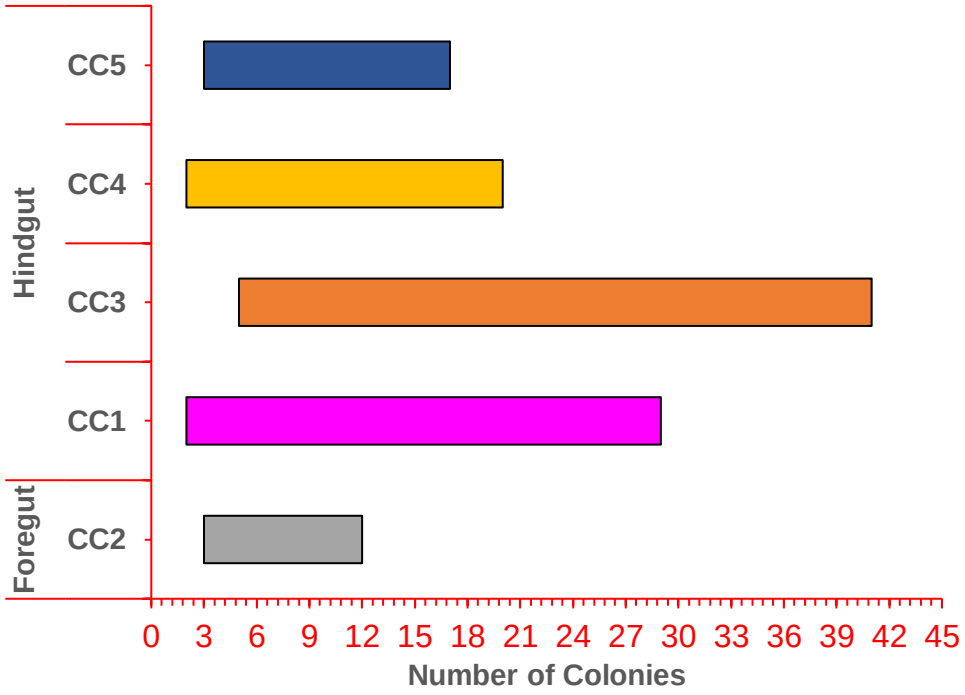

Table 3: Results of Gram staining of isolated bacterial colonies

S. No.	Isolate Code	Strain	Gram
1.	CC1	*Serratia*	Negative
2.	CC2	*Staphylococcus*	Positive
3.	CC3	*Aeromonas*	Negative
4.	CC4	*Bacillus*	Positive
5.	CC5	*Pseudomonas*	Negative

Biochemical Characterisation

In the present study, various biochemical tests were performed to characterise the isolated bacterial strains. *Serratia* species are usually motile and test positive for citrate utilisation, urease, and catalase, but they are oxidase-negative and do not reduce sulfur. In contrast, *Staphylococcus* species are non-motile, catalase-positive, and may have variable urease activity but generally test negative for citrate utilisation, oxidase, and sulfur reduction. *Aeromonas* species are motile and consistently positive for citrate, urease, catalase, oxidase, and sulfur reduction. *Bacillus* species are motile, catalase-positive, often utilise citrate, and exhibit variable urease and sulfur-reduction activity; they are oxidase-negative. *Pseudomonas* species are motile, oxidase-positive, and usually positive for citrate utilisation and catalase, although urease activity varies, and they do not reduce sulfur (Table 4).

Table 4: Biochemical characteristics of isolated bacterial strains

S. No.	Isolate Code	Biochemical Tests					
		Citrate Utilisation	Urease	Catalyst	Sulphur Reduction	Oxidase	Motility
1.	CC1	+	+	+	-	-	+
2.	CC2	-	+	+	-	-	-
3.	CC3	+	+	+	+	+	+
4.	CC4	+	-	+	-	-	+
5.	CC5	+	+	+	-	+	+

DISCUSSION

The study aims to examine the extensive bacterial populations in the foregut and hindgut regions of the alimentary tract of the Indian Major Carp (*Labeo catla*). In the present study, we isolated five bacterial strains from the digestive tract. Our study aligns with Dutta's 2018 study. The authors isolated several bacterial strains from the proximal and distal segments of *Labeo rohita* (18). Bacteria are generally abundant in aquatic environments, making it nearly impossible to avoid them in fish diets (19). These bacteria enter the fish's diet via ingestion and may colonise the gastrointestinal tract, forming a symbiotic relationship (4). Most of the available literature reports that the digestive tract of fish harbours a large number of microbes (see 20, 21, 22). Some study reported that the digestive tract of endothermic animals was colonised mainly by obligate anaerobes (23). However, some investigations reported that most fish guts contained aerobes or facultative anaerobes (24, 9).

The comparison of bacterial populations in the foregut and hindgut sections of the gastrointestinal tract revealed distinct microbial distributions, consistent with the known anatomical and functional differences between these areas. The highest colony-forming units (CFU) were observed at the 10^4 dilution, with a notable decrease at 10^6, indicating a dilution-dependent decline in viable bacteria. This pattern is consistent with typical microbiological expectations: higher dilutions yield fewer colonies because the bacterial concentration per unit volume is lower.

Furthermore, the analysis of viable bacterial counts in the gastrointestinal tract of *Catla catla* revealed a distinct regional variation, with the hindgut exhibiting significantly higher microbial populations than the foregut. This observation aligns with established patterns in fish gut microbiology, in which the hindgut serves as a more favourable niche for microbial colonisation due to its anaerobic environment, slower transit time, and greater substrate availability (25). In the present study, the mean viable bacterial count at a $10^{\wedge 4}$ dilution in the foregut was 12.72 ± 5.91 CFU/µL, compared to 20.32 ± 9.15 CFU/µL in the hindgut. However, at higher dilutions ($10^{\wedge 6}$), the foregut dropped to 4.21 ± 1.62 CFU/µL, while the hindgut

retained 8.31 ± 4.33 CFU/µL, indicating a more stable microbial load in the hindgut across dilutions. These findings align with the study made by Mukerjee and his colleagues, the authors reported that the total intestinal microflora in *L. catla* averaged 4.62×10^6 CFU/g, with higher densities typically found in the distal gut segments (26).

This observed variation in bacterial load between the two regions may reflect the functional specialisation of the hindgut, which supports dense microbial communities through fermentation and nutrient recovery (8)**.** Most studies have also shown that the hindgut harbours a diverse array of anaerobic and facultatively anaerobic bacteria that contribute to the production of short-chain fatty acids, vitamin synthesis, and immune modulation (27, 28). However, region-specific variation in bacterial load has also been documented in other Indian Major Carps (*Labeo rohita* and *Cirrhinus mrigala*) (16). The authors reported that the hindgut microbiota play a vital role in host metabolism and health. The region-specific variation was influenced by factors such as diet, environmental conditions, host physiology, and hormonal regulation (29, 30). Additionally, studies in ruminants and hindgut fermenters have consistently demonstrated that hindgut regions, such as the cecum and colon, harbour more diverse and abundant microbiota than foregut regions, such as the stomach or crop (31, 32) The hindgut provides a stable, anaerobic environment that supports microbial proliferation, especially of obligate anaerobes involved in fibre degradation and short-chain fatty acid production (33).

CONCLUSION

In conclusion, the study underscores the importance of anatomical compartmentalisation in shaping microbial ecology within the gastrointestinal tract of the study animal. The hindgut's higher bacterial load and greater variation relative to the foregut support its role as a microbial fermentation chamber, with implications for host nutrition, immunity, and gut health.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

The author (s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

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REFERENCES

1. Gadhiya, A., Katariya, S., Khapandi, K., & Chhatrodiya, D. (2025). Probiotics as a sustainable alternative to antibiotics in aquaculture: A review of the current state of knowledge. The Microbe, 8, 100426. https://doi.org/10.1016/j.microb.2025.100426.

2. Nayak, S.K. (2010). Role of gastrointestinal microbiota in fish. Aquaculture Research, 20(3), 441–453. https://doi.org/10.1111/j.1365-2109.2010.02546.x.

3. Sun, Y.-Z., Zhang, Y., & Ringø, E. (2022). Composition, functions and modulation of gut microbiota in maricultural animals. Frontiers in Marine Science, 9, 985012. https://doi.org/10.3389/fmars.2022.985012.

4. Yukgehnaish, K., Kumar, P., Sivachandran, P., Marimuthu, K., Arshad, A., Paray, B. A., & Arockiaraj, J. (2020). Gut microbiota metagenomics in aquaculture: Factors influencing gut microbiome and its physiological role in fish. Reviews in Aquaculture, 12(3), 1903–1927. https://doi.org/10.1111/raq.12416.

5. Zhang, Z., Yang, Q., Liu, H., Jin, J., Yang, Y., Zhu, X., Han, D., Zhou, Z., & Xie, S. (2024). Potential functions of the gut microbiome and modulation strategies for improving aquatic animal growth. Reviews in Aquaculture, 16(1), 1–20. https://doi.org/10.1111/raq.12959.

6. Hampton, H. G., Watson, B. N., & Fineran, P. C. (2020). The arms race between bacteria and their phage foes. Nature, 577(7790), 327-336. https://doi.org/10.1038/s41586-019-18948.

7. Li, T., Long, M., Gatesoupe, F.-J., Zhang, Q., Li, A., & Gong, X. (2015). Comparative analysis of the intestinal bacterial communities in different species of carp by pyrosequencing. Microbial Ecology, 69(1), 25–36. https://doi.org/10.1007/s00248-014-0480-8.

8. Ringø, E. Z. Z. V., Zhou, Z., Vecino, J. G., Wadsworth, S., Romero, J., Krogdahl, Å., Olsen, R. E., Dimitroglou, A., Foey, A., Davies, S., & Owen, M. (2016). Effect of dietary components on the gut microbiota of aquatic animals: A never-ending story? Aquaculture Nutrition, 22(2), 219–282. https://doi.org/10.1111/anu.12346.

9. Egerton, S., Culloty, S., Whooley, J., Stanton, C., & Ross, R. P. (2018). The gut microbiota of marine fish. Frontiers in Microbiology, 9, 873. https://doi.org/10.3389/fmicb.2018.00873.

10. Kokou, F., Sasson, G., Friedman, J., Eyal, S., Ovadia, O., Harpaz, S., & Mizrahi, I. (2019). Core gut microbial communities are maintained by beneficial interactions and strain variability in fish. Nature Microbiology, 4(12), 2456–2465. https://doi.org/10.1038/s41564-019-0560-0.

11. Han, S., Liu, Y., Zhou, Z., He, S., Cao, Y., Shi, P., & Yu, Y. (2010). Analysis of bacterial diversity in the intestine of grass carp (Ctenopharyngodon idellus) based on 16S rDNA gene sequences. Aquaculture Research, 42(1), 47–56. https://doi.org/10.1111/j.1365-2109.2010.02543.x.

12. Meng, X., Li, S., Qin, C., Zhu, Z., Hu, W., Yang, L., & Chen, J. (2018). Intestinal microbiota and lipid metabolism responses in the common carp (Cyprinus carpio L.) following copper exposure. Ecotoxicology and Environmental Safety, 160, 257–264. https://doi.org/10.1016/j.ecoenv.2018.05.050.

13. Liu, C., Zhao, L.-P., & Shen, Y.-Q. (2021). A systematic review of advances in intestinal microflora of fish. Fish Physiology and Biochemistry, 47(6), 2041–2053. https://doi.org/10.1007/s10695-021-01027-3.

14. Hou, H., Li, L., Xu, C., Li, C., & Tang, W. (2025). Research progress on the functions of intestinal microbiota in freshwater fish and its applications in aquaculture. Feed Research, 48(2), 206–212. https://doi.org/10.13557/j.cnki.issn1002-2813.2025.02.037.

15. Mondal, S. (2022). Screening of probiotic bacteria from Rohu (Labeo rohita) gut for enzyme production. Journal of Applied Aquaculture, 34(2), 245–260. https://doi.org/10.1080/10454438.2022.2045678.

16. Sundaray, J. K., Mohapatra, M., Rasal, A., Udit, U. K., Mohanty, S., Mohanty, D., Das, I. I., Sahoo, L., & Sahoo, P. K. (2025). Sex-specific gut microbiome dynamics in Labeo catla: Links to reproductive hormones, metabolic dimorphism, and environmental factors. Frontiers in Microbiology, 16, 1651975. https://doi.org/10.3389/fmicb.2025.1651975.

17. Liu, J., Tan, Y., Cheng, H., Zhang, D., Feng, W., & Peng, C. (2022). Functions of gut microbiota metabolites, current status and future perspectives. Ageing and Disease, 13(4), 1106–1126. https://doi.org/10.14336/AD.2022.0104.

18. Dutta, D., Banerjee, S., Mukherjee, A., & Ghosh, K. (2018). Potential gut adherent probiotic bacteria isolated from rohu, Labeo rohita (Actinopterygii: Cypriniformes: Cyprinidae): Characterisation, exo-enzyme production, pathogen inhibition, cell surface hydrophobicity, and bio-film formation. Acta Ichthyologica et Piscatoria, 48(3), 221–223. https://doi.org/10.3750/AIEP/02251.

19. Kanika, N. H., Liaqat, N., Chen, H., Ke, J., Lu, G., Wang, J., & Wang, C. (2025). Fish gut microbiome and its application in aquaculture and biological conservation. Frontiers in Microbiology, 15, Article 1521048. https://doi.org/10.3389/fmicb.2024.1521048.

20. Fatima, S. K. T. (2025). Study of gut microbiota of major carp fishes of India. In S. Sudarshan, S. Pedge, & V. B. Kalyankar (Eds.), Recent trends in fisheries and aquaculture (pp. 1-55). [ISBN: 978-93-95369-50-3]. https://doi.org/10.5281/zenodo.16932412.

21. Kim, P. S., Shin, N. R., Lee, J. B., Kim, M.S., Whon, T.W., Hyun, D.W., Yun, J.H., Jung, M.J., Kim, J.Y., & Bae, J.W. (2021). Host habitat is the major determinant of the gut microbiome of fish. Microbiome, 9, 166. https://doi.org/10.1186/s40168-021-01113-x.

22. Luan, Y., Li, M., Zhou, W., Yao, Y., Yang, Y., Zhang, Z., Ringø, E., Olsen, R. E., Liu Clarke, J., Xie, S., Mai, K., Ran, C., & Zhou, Z. (2023). The fish microbiota: Research progress and

potential applications. Engineering, 29, 137–146. https://doi.org/10.1016/j.eng.2022.12.011.

23. Ou, W., Yu, G., Zhang, Y., & Mai, K. (2021). Recent progress in the understanding of the gut microbiota of marine fishes. Marine Life Science & Technology, 3(4), 434–448. https://doi.org/10.1007/s42995-021-00094-y.

24. 24.Uma, A., Palaniappan, S., & Abraham, T.J. (2020). Importance of gut microbiota in fish: A review. Indian Journal of Animal Health, 59(2-Spl):181-194. http://doi.org/10.36062/ijah.59.2SPL.2020.181-194.

25. Baliyan, R., Saini, N., Kumar, N., & Bhadula, S. (2025). Feeding behaviour of fish Catla catla (Hamilton, 1822) with special reference to gut content analysis. International Journal of Zoology Studies, 10(3), 67–71. https://www.zoologyjournals.com.

26. Mukherjee, A., Rodiles, A., Merrifield, D. L., Chandra, G., & Ghosh, K. (2020). Exploring intestinal microbiome composition in three Indian major carps under polyculture system: A high-throughput sequencing-based approach. Aquaculture, 524, 735206. https://doi.org/10.1016/j.aquaculture.2020.735206.

27. Mukhopadhya, I., & Louis, P. (2025). Gut microbiota-derived short-chain fatty acids and their role in human health and disease. Nature Reviews Microbiology, 23(10), 635–651. https://doi.org/10.1038/s41579-025-01183-w.

28. Park, I., & Mannaa, M. (2025). Fermented foods as functional systems: Microbial communities and metabolites influencing gut health and systemic outcomes. Foods, 14(13), 2292. https://doi.org/10.3390/foods14132292.

29. Clever, F., Sourisse, J. M., Preziosi, R. F., Eisen, J. A., Rodriguez Guerra, E. C., Scott, J. J., Wilkins, L. G. E., Altieri, A. H., McMillan, W. O., & Leray, M. (2022). The gut microbiome variability of a butterflyfish increases on severely degraded Caribbean reefs. Communications Biology, 5, 770. https://doi.org/10.1038/s42003-022-03679-0.

30. Cai, J., Yin, B., Wang, Y., Pan, K., Xiao, Y., & Wang, X. (2023). Gut microbiome play a crucial role in geographical and interspecies variations in mercury accumulation by fish. Science of the Total Environment, 869, 169381. https://doi.org/10.1016/j.scitotenv.2023.169381.

31. Godoy-Vitorino, F., Goldfarb, K. C., Karaoz, U., Leal, S., Garcia-Amado, M. A., Hugenholtz, P., Tringe, S. G., Brodie, E. L., & Dominguez-Bello, M. G. (2012). Comparative analyses of foregut and hindgut bacterial communities in hoatzins and cows. The ISME Journal, 6(3), 531–541. https://doi.org/10.1038/ismej.2011.131.

32. O'Donnell, M. M., Harris, H. M. B., Ross, R. P., & O'Toole, P. W. (2017). Core faecal microbiota of domesticated herbivorous ruminant, hindgut fermenters, and monogastric animals. Microbiology Open, 6(5), e00509. https://doi.org/10.1002/mbo3.509.

33. Ma, Y., Deng, X., Yang, X., Wang, J., Li, T., Hua, G., Han, D., Da, L., Li, R., Rong, W., & Deng, X. (2022). Characteristics of bacterial microbiota in different intestinal segments of Aohan fine-wool sheep. Frontiers in Microbiology, 13, 874536. https://doi.org/10.3389/fmicb.2022.874536.