

RESEARCH ARTICLE

Skin mucus of *Oncorhynchus mykiss* as a functional barrier: Quantitative immunological and antimicrobial profilingGokcenur Sanioglu Golen^{1,(*)}, Ali Uslu²¹Aksaray University, Faculty of Veterinary Medicine, Microbiology Department, 68100, Aksaray, Türkiye²Selcuk University, Faculty of Veterinary Medicine, Microbiology Department, 42130, Konya, Türkiye

Abstract

Fish are constantly exposed to various bacterial, fungal, and viral pathogens in both natural and artificial environments. As the first line of defense against these external threats, mucosal surfaces, particularly skin mucus, play a critical role. Skin mucus is not only a physical barrier but also an active immunological interface enriched with molecules such as lysozyme, immunoglobulins, complement proteins, and antimicrobial peptides. In aquaculture systems, infectious diseases represent a major threat to fish health and productivity. This study aimed to evaluate the immunological components and antimicrobial activity of skin mucus obtained from *Oncorhynchus mykiss* (*O. mykiss*). Levels of immunoglobulin M (IgM), complement components C3 and C4 were quantified using enzyme-linked immunosorbent assay (ELISA). Lysozyme activity was assessed spectrophotometrically. The antimicrobial properties of the mucus were tested against eight selected bacterial and fungal pathogens by determining their Minimum Inhibitory Concentration (MIC) and Minimum Lethal Concentration (MLC). Skin mucus samples contained significant levels of immune-related proteins. Mucus showed significant antimicrobial activity against Gram-positive and Gram-negative bacteria and some common fungal species. In particular, its bacteriostatic and bactericidal effects are quite high against Gram-negative bacteria. This study demonstrates that the skin mucus of *O. mykiss* is not only a passive barrier but also an active component of innate immunity, offering potential as a natural biological source in sustainable aquaculture practices and in the development of alternative antimicrobial agents.

Keywords: Antimicrobial peptides, Immunity, Lysozyme, Mucus, *Oncorhynchus mykiss*

(*) **Corresponding author:**Gokcenur Sanioglu Golen
gokcenursanioglu@aksaray.edu.tr**Received:** 26.06.2025**Accepted:** 08.10.2025**Published:** 23.10.2025**How to cite this article?**Sanioglu-Golen G and Uslu A 2025.
Skin mucus of *Oncorhynchus mykiss*
as a functional barrier:
Quantitative immunological and
antimicrobial profiling.
Eurasian J Vet Sci, 41, e0462.

INTRODUCTION

Fish are generally exposed to pathogen microorganisms in their aquatic environments and depend on innate immunity to mount rapid defensive responses (Ellis 2001, Işık et al 2016). An important component of this innate immune system is the mucus layer coating their skin. This mucus is not just a passive barrier; it also helps protect the fish by using its rich mix of active molecules (Hitit et al 2020, Sharba et al 2022). A recent study showed that dynamic physical and biochemical barrier, displaying numerous biological and ecological roles such as protection against abrasion environmental toxins, pathogens. Fish mucus also contains various immune components (such as lysozyme, immunoglobulins (IgM, IgD), complement proteins (eg., C3, C4), lectins, antimicrobial peptides) that play a crucial role in preventing pathogen adhesion and invasion. In addition to this barrier function, these components exhibit bacteriostatic or bactericidal

effects, contributing directly to the inhibition or elimination of microbial pathogens (Fiordelmondo et al 2023, Dik et al 2024).

Mucus surfaces are dynamic matrices. Mucosal surfaces are highly dynamic structures, and their composition can vary between fish species as well as in response to internal factors such as developmental stage, and external influences including stress, water temperature, pH, and infections (Hoare et al 2021, Fiordelmondo et al 2023, Reverter et al 2018).

Oncorhynchus mykiss (*O. mykiss*) generally farmed in the Black Sea region due to its high adaptability to the local environment and good growth performances (Akbulut et al 2002, Kaya Öztürk et al 2019). Under intensive aquaculture conditions the mucus layer. natural defence mechanisms have played a critical important role in enhancing our understanding of fish immunological mechanisms against pathogens and environmental changes (Esteban 2012, Ghafarifarsani et al 2021).



Microbial contamination and the presence of zoonotic pathogens in all animal-derived food products pose significant risks to both animal and human health (Baytaroglu and Kucukkagnici 2025, Ghafarifarsani et al 2021, Keskinoglu and Gulel 2022, Telli et al 2022). Significant pathogens in aquaculture such as *Salmonella* spp., *Yersinia* spp. and *Aeromonas* spp. can deal mucosal immunity. Zoonotic bacteria pose important risks to both fish and human health. Consequently, assessing the antimicrobial properties of fish mucus against these pathogens is vital for sustainable aquaculture and food safety (Ghafarifarsani et al 2021).

Recent studies also highlight the antifungal potential of fish mucus, particularly against *Aspergillus fumigatus* and various *Candida* species (Yadav and Mishra 2023). This suggests that fish mucus is a different immune component, giving defense against bacteria, fungi, and viruses (Díaz-Puertas et al 2023, Yadav and Mishra 2023, Dik et al 2024).

The aim of this study is to evaluate in vitro the immune-related components present in the mucus of *O. mykiss* and determine its antimicrobial efficacy against zoonotic bacterial pathogens, including *Escherichia coli* (*E. coli*), *Pasteurella multocida* (*P. multocida*), *Aeromonas* spp., *Yersinia* spp., and *Salmonella* spp. Insight into how this natural barrier interacts with pathogens can provide important understanding for fish health management and potential biomedical applications.

MATERIAL AND METHODS

Collection of Fish Mucus Samples

Mucus samples were collected from the abdominal and lateral surfaces of each fish using a sterile spatula and transferred to sterile 15 mL tubes. Mucus samples were obtained fresh from commercially sourced *O. mykiss*. To ensure homogeneity, fish of the same harvest batch were used, meeting standard freshness criteria such as firm texture, bright gills, and clear eyes, as described by the FDA (2024). The sampled fish represented a homogeneous group, with an average weight of approximately 1500 ± 100 g. A total of 20 fish mucus samples were taken from fish transported under the aseptic conditions and delivered to the laboratory under cold chain conditions. Mucus samples were centrifuged at 2000 g for 10 min, the resulting supernatant was subsequently passed through a $0.45 \mu\text{m}$ membrane filter (Sartorius), and all samples were stored at -20°C until use (Hitit et al 2020).

Determination of Lysozyme Activity

The *Micrococcus lysodeikticus* (ATCC No: 4698; Sigma-Aldrich) solution was prepared in 0.05 M sodium phosphate buffer to a final concentration of 0.2 mg/mL (pH 6.2) for lysosym activity. In each analysis, 100 μL of

the mucus sample was mixed with 2.5 mL of bacterial suspension, and the change in absorbance was measured at 450 nm. Lysozyme activity was calculated based on the rate of absorbance decrease observed over a specific time interval and expressed as activity units per unit volume (Levipan et al 2020, Sridhar et al 2021).

Determination of Immune Parameters

Immunoglobulin M (IgM), complement component 3 (C3), and complement component 4 (C4) levels were measured to evaluate the immune profile of mucus samples. Commercial ELISA kits from MyBioSource were used for these analyses: Fish Immunoglobulin M ELISA Kit (BAA12043.1) for IgM, Fish Complement Component 3 ELISA Kit (MBS005953) for C3, and Fish Complement 4 ELISA Kit (MBS281826) for C4.

All assays were carried out using 96-well microplates, following the manufacturers' instructions. Absorbance readings were taken at 450 nm with a microplate reader. The resulting optical density (OD) values were then used to calculate the concentrations of each component (in $\mu\text{g/mL}$) by referencing standard curves generated during the assay.

Antibacterial and Antifungal Activity Tests

The antimicrobial activity of the fish skin mucus was assessed using a serial dilution method in 96-well microtiter plates. Minimum Inhibitory Concentration (MIC) and Minimum Lethal Concentration (MLC) values were determined according to CLSI (2024) and EUCAST (2020) guidelines. All bacterial and fungal microorganisms tested were obtained from the Culture Collection of the Faculty of Veterinary Medicine, Aksaray University. The tested microorganisms included *Staphylococcus aureus*, and Gram-negative bacteria such as *E. coli*, *P. multocida*, *Aeromonas* spp., *Yersinia* spp., and *Salmonella* spp., along with fungal pathogens *A. fumigatus* and *Candida* spp.

The total protein concentration of the crude mucus extract was quantified using the Bradford assay. This value was used as the starting concentration for the dilution assays. Two-fold serial dilutions were prepared in sterile conditions, and for each well, 100 μL of diluted mucus sample was added, followed by the addition of approximately 1.5×10^6 CFU/mL of bacterial or fungal suspension.

At the end of the incubation period, each well was visually inspected. The lowest mucus concentration at which no visible turbidity was observed was recorded as the MIC. To determine the MLC, 10 μL suspensions were aseptically taken only from the wells where MIC was observed and streaked onto Mueller–Hinton agar and Sabouraud dextrose agar plates. Plates were incubated at 37°C for 24 h and 28°C for 48 h) under aerobic conditions. The lowest concentration that produced no microbial growth on the

Table 1. ELISA Results for Mucosal Immune Parameters of skin mucus of *Oncorhynchus mykiss* (IgM, C3, C4)

Parameter	Mean Concentration (µg/mL)	Standard Deviation (±SD)
IgM	303.4	12.7
C3	102.8	10.2
C4	98.6	9.3

solid media was recorded as the MLC.

MIC and MLC determinations were performed following standard broth microdilution protocols recommended by the Clinical and Laboratory Standards Institute (CLSI, 2024; EUCAST, 2020) and previously described methods

Statistical Analysis

Positive controls contained only microorganisms, while negative controls contained only fish mucus. Antibiotics and antifungal agents were used for comparison. All experiments were performed in triplicate, and results were expressed as mean ± standard deviation (X ± SD). Statistical differences were analyzed using one-way ANOVA followed by Tukey's post hoc test ($p < 0.05$).

RESULTS

Lysozyme Activity Findings

The lysozyme activity of fish mucus analyzed using *M. lysodeikticus*, and the mean activity level was determined as $2.86 \pm 0.31 \text{ U mg}^{-1}$.

Mucosal Immune Parameters

The ELISA analyses performed on the mucus samples revealed that the concentrations of IgM, C3, and C4 were $303.4 \pm 12.7 \text{ µg/mL}$, $102.8 \pm 10.2 \text{ µg/mL}$, and 98.6 ± 9.3

Table 2. MIC and MLC Values of Fish Mucus Against Various Microorganisms of skin mucus of *Oncorhynchus mykiss*

Microorganism	MIC (µg/mL)	MLC (µg/mL)
<i>Staphylococcus aureus</i>	62.5	125
<i>Escherichia coli</i>	125	250
<i>Pasteurella multocida</i>	62.5	125
<i>Aeromonas</i> spp.	31.25	62.5
<i>Yersinia</i> spp.	62.5	125
<i>Salmonella</i> spp.	125	250
<i>Aspergillus fumigatus</i>	250	500
<i>Candida</i> spp.	125	250

µg/mL, respectively. All measurements were conducted in triplicate, and the data are presented as mean ± standard deviation (Table 1).

Antimicrobial Activity Findings (MIC and MLC Values)

The antimicrobial effects of fish mucus on eight different microorganisms were determined based on MIC and MLC values, which were expressed as protein equivalents (µg/mL) according to the Bradford-based protein quantification of crude mucus extract, whose initial concentration was approximately 4 mg/mL.

The analysis revealed significant differences in microbial susceptibility to fish mucus, with MIC values ranging from 31.25 to 250 µg/mL and MLC values from 62.5 to 500 µg/mL. The lowest MIC was observed against *Aeromonas* spp. (31.25 µg/mL), while the highest MIC was recorded for *A. fumigatus* (250 µg/mL). Similarly, the lowest MLC value was 62.5 µg/mL for *Aeromonas* spp., and the highest MLC was 500 µg/mL for *A. fumigatus* (Table 2).

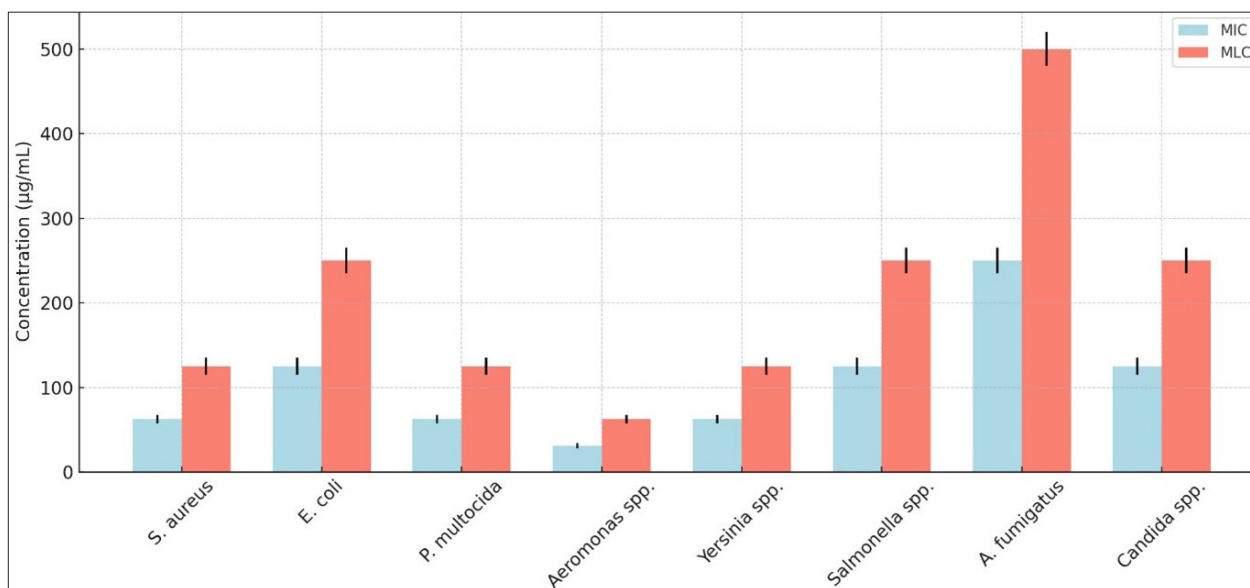


Figure 1. Comparative MIC and MLC concentrations (µg/mL) of fish mucus against tested microorganisms. Values are presented as mean ± standard deviation (SD) from three independent replicates. MIC: Minimum Inhibitory Concentration; MLC: Minimum Lethal Concentration.

The MIC and MLC values of fish mucus against all tested microorganisms are summarized in Figure 1, with standard deviations representing variation among replicates.

DISCUSSION

In this study, the immunological parameters present in the skin mucus of *O. mykiss* and the antimicrobial activity of this mucus against various bacterial and fungal pathogens were evaluated. The findings demonstrated that fish mucus is not merely a passive barrier, but a dynamic immune component that responds to environmental threats.

The measured IgM concentration in mucus samples was 303.4 ± 12.7 $\mu\text{g/mL}$, indicating an active mucosal immune response in *O. mykiss*. Although this value is lower than the 392 ± 16 $\mu\text{g/mL}$ reported by Dik et al (2024) in trout mucus, differences in sample extraction methods, dilution rates, and analytical techniques limit direct comparisons. Nevertheless, the IgM level observed here is significant as it reflects the physiological immune state of freshly collected mucus samples. This finding aligns with the mucosal IgM increases reported by Hoare et al (2021) and Ghafarifarsani et al (2021), supporting the notion that fish mucus plays an active role in humoral immune modulation, particularly at the initial site of pathogen contact. Hence, mucosal IgM may serve as a potential biomarker for monitoring fish health and optimizing aquaculture immunization strategies.

Immunological factors in fish mucus, particularly IgM and complement components, can vary with developmental stage and body size. Previous studies have shown that juveniles often exhibit weaker immune responses than adults due to incomplete development of innate and adaptive systems (Magnadóttir 2006; Uribe et al 2011). Similarly, complement activity and antibody levels differ across growth stages, reflecting ontogenetic changes in immune competence (Guardiola et al 2014; Saleh et al 2019). In this study, fish of relatively uniform size (~1500 g) were used, minimizing variability. Nevertheless, potential stage-dependent differences in mucus immune factors should be considered when interpreting and generalizing the findings.

The C3 (102.8 ± 10.2 $\mu\text{g/mL}$) and C4 (98.6 ± 9.3 $\mu\text{g/mL}$) levels detected in mucus indicate that the complement system is active at mucosal surfaces and that innate immunity is functioning effectively. These results are generally consistent with previous studies. Esteban (2012) pointed out that fish skin mucus contains a range of immune-related proteins, including key complement components such as C3 and C4. In a study, Saleh et al (2019) identified C3 and C4 in carp mucus and determined that the presence of these components may vary depending on the stage of immune activation. Guardiola et al (2014) detected measurable levels of C3 and lysozyme in mucus

samples obtained from various marine fish species and identified the relationship between these molecules and general immune function. Compared to the findings of Chen et al (2023) in *O. niloticus*, our results in *O. mykiss* confirm that although C3 levels are similar, host-parasite specificity strongly influences the killing efficiency.

Despite these findings, studies providing quantitative data on the concentrations of C3 and C4 in particular in fish mucus are still quite limited. The partially lower levels of these complement proteins in mucus compared to blood serum levels are explained by their limited diffusion to external mucosal surfaces, as also noted by Salinas et al (2011).

The lysozyme activity of fish mucus was measured as 2.86 ± 0.31 U mg^{-1} . This level of activity is in agreement with previous studies reporting lysozyme as a major component of the innate immune system in various fish species. This effect is attributed to its ability to degrade bacterial cell walls through enzymatic cleavage of peptidoglycan (Mozumder 2005, Levipan et al 2020).

This study thoroughly examined the antimicrobial effects of *O. mykiss* mucus components against bacterial species including *E. coli*, *Aeromonas* spp., *Yersinia* spp., *Salmonella* spp., and *P. multocida*, as well as fungal pathogens *Candida* spp. and *A. fumigatus*. The results confirmed that fish mucus functions as an active immune barrier beyond its physical properties.

Particularly noteworthy was the strong antibacterial activity observed against Gram-negative pathogens, with the lowest MIC (31.25 $\mu\text{g/mL}$) recorded for *Aeromonas* spp. This supports the role of mucus as a robust defense mechanism in aquaculture environments, where *Aeromonas* spp. are common pathogens. Diaz Pubertas et al (2023) similarly reported broad-spectrum inhibition by mucus extracts, indicating functionally conserved mucosal defenses across fish species.

Lee et al (2020) conducted a comprehensive review of mucus from 47 fish species and reported variable antibacterial activity against 46 bacterial strains. Factors such as extraction method (aqueous, organic, acidic), habitat (freshwater, marine, brackish), and pathogen structure all influenced mucus efficacy. The use of fresh, undiluted aqueous extracts in the present study preserved the natural immune profile, enhancing physiological relevance.

Moderate MIC values observed against pathogens such as *P. multocida*, *Yersinia* spp., and *E. coli* support the broad-spectrum potential of fish mucus. However, as Levipan and Avendaño-Herrera (2020) noted, pathogens like *Piscirickettsia salmonis* can bypass the mucus barrier and infect through skin or gills. Although the behavior of such pathogens in the presence of mucus remains unclear, mucus may still inhibit colonization or delay cellular

invasion. Strengthening mucosal immunity appears crucial for managing endemic aquaculture pathogens.

The antifungal activity observed against *Candida* spp. (MIC: 125 µg/mL) and *A. fumigatus* (MIC: 250 µg/mL) was less potent than against bacterial strains, suggesting relatively limited antifungal efficacy. Nonetheless, Yadav and Mishra (2023) showed that fish mucus from multiple species inhibited several fungi, including *Candida*, *Aspergillus*, and *Mucor*. Although Özil et al (2022) focused on antifungal effects of essential oil nanoemulsions, biological materials like mucus offer sustainable, resistance-free defenses due to their natural peptide content.

Dik et al (2024) detected high levels of galectin and hepcidin in *O. mykiss* mucus, with well-defined antiviral, antibacterial, and immunomodulatory properties. These peptides disrupt microbial membranes by forming pores, increasing permeability, and leading to cell death (Hussain and Sachan 2024). The low MIC values observed here support the presence and functional activity of such peptides. Mucus has attracted attention due to their antiviral and anticancer properties, as reported in studies conducted in recent years (Dik et al 2024, Hussain and Sachan 2024).

Beyond pathogen inhibition, the bioactive compounds in mucus also exhibit immunomodulatory functions. Immune components are known to contribute in inflammation and regulate cytokine responses (Hitit et al 2020). These features position mucus not only as a tool for aquaculture biosecurity but also as a promising source for natural therapeutic agents.

CONCLUSION

This study emphasized that the mucus of *O. mykiss* contains important components related to the innate immune system and significant antimicrobial activity. The presence of measurable levels of IgM, C3, and C4 indicates an active immune response on the mucosal surface and an effective defense mechanism against environmental pathogens.

Additionally, the mucus exhibited significant bacteriostatic and bacteriocidal effects against bacteria and fungal pathogens. In particular, strong antimicrobial activity was observed against pathogens commonly encountered in aquaculture, such as *Aeromonas* spp. and *Staphylococcus aureus*.

These findings suggest that fish mucus is not only a natural defense layer but also a source with biotherapeutic potential. It is considered necessary to conduct more detailed molecular-level research on the immune-related protein and peptide content of mucus. Overall, this study provides a valuable scientific foundation for the development of natural antimicrobial agents and the identification of alternative treatment approaches.

DECLARATIONS

Competing Interests

Authors declare that there are no conflicts of interest related to the publication of this article.

Availability of Data and Materials

The data that support the findings of this study are available on request from the corresponding author.

Funding

No external funding was received for this study.

Ethical Statement

Aksaray University Local Ethics Committee for Animal Experiments, Decision No: 25/5/38.

Author Contributions

Motivation/Concept: GSG; Design: GSG, AU; Control/Supervision: GSG, AU; Data Collection and Processing: GSG; Analysis and Interpretation: GSG, AU; Literature Review: GSG; Writing the Article: GSG, AU; Critical Review: AU

ORCID

GSG: <https://orcid.org/0000-0001-9950-330X> 

AU: <https://orcid.org/0000-0002-8319-831X> 

REFERENCES

- Akbulut B, Şahin T, Aksungur N, Aksungur M, 2002. Effect of initial size on growth rate of rainbow trout, *Oncorhynchus mykiss*, reared in cages on the Turkish Black Sea Coast. Turk J Fish Aquat Sci, 2(2).
- Baytaroglu ES, Kucukkagnici O, 2025. Screening of *Aeromonas* spp. and their antimicrobial resistance profiles in wild fish and seawater from the Gulf of Antalya, Mediterranean Sea. Eurasian J Vet Sci, 41:e0443, <https://doi.org/10.15312/EurasianJVetSci.2025.443>
- Chen J, Liu C, Yang T, 2023. Killing effect of fish serum and mucus on *Gyrodactylus cichlidarum* (Platyhelminthes, Monogenea) mediated by complement. Aquaculture, 567, 739248.
- Clinical and Laboratory Standards Institute (CLSI), 2024. Methods for dilution antimicrobial susceptibility tests for bacteria that grow aerobically. 12th ed. CLSI standard M07. Clinical and Laboratory Standards Institute, Wayne, PA. <https://clsi.org/shop/standards/m07>
- Díaz-Puertas R, Adamek M, Mallavia R, Falco A, 2023. Fish skin mucus extracts: an underexplored source of antimicrobial agents. Mar Drugs, 21(6), 350, <https://doi.org/10.3390/md21060350>
- Dik I, Dik B, Tufan Ö, Er A, 2024. Evaluation of potential antiviral activities of antimicrobial peptides in fish mucus. Fundam Clin Pharmacol, 38(3), 695–702, <https://doi.org/10.1111/fcp.12996>
- Ellis AE, 2001. Innate host defense mechanisms of fish against viruses and bacteria. Dev Comp Immunol, 25(8-9), 827-839
- Esteban AM, 2012. An overview of the immunological defenses in fish skin. Int Sch Res Notices, 2012(1), 853470, <https://doi.org/10.5402/2012/853470>
- European Committee on Antimicrobial Susceptibility Testing (EUCAST), 2020. Breakpoint tables for interpretation of MICs and zone diameters, Version 10.0, 1–90.
- Fiordelmondo E, Magi GE, Friedl A, El-Matbouli M, et al.,

2023. Effects of stress conditions on plasma parameters and gene expression in the skin mucus of farmed rainbow trout (*Oncorhynchus mykiss*). *Front Vet Sci*, 10, 1–8, <https://doi.org/10.3389/fvets.2023.1183246>
- Ghafariarsani H, Rashidian G, Sheikhlari A, Naderi Farsani M, et al., 2021. The use of dietary oak acorn extract to improve haematological parameters, mucosal and serum immunity, skin mucus bactericidal activity, and disease resistance in rainbow trout (*Oncorhynchus mykiss*). *Aquac Res*, 52(5), 2518–2527, <https://doi.org/10.1111/are.15101>
- Guardiola FA, Cuesta A, Arizcun M, Meseguer J, et al., 2014. Comparative skin mucus and serum humoral defence mechanisms in the teleost gilthead seabream (*Sparus aurata*). *Fish Shellfish Immunol*, 36(2), 545–551, <https://doi.org/10.1016/j.fsi.2014.01.001>
- Hitit M, Corum O, Ozbek M, Uney K, et al., 2020. Mucus from different fish species alleviates carrageenan-induced inflammatory paw edema in rats. *Asian Pac J Trop Biomed*, 10(10), 452–459, <https://doi.org/10.4103/2221-1691.290870>
- Hoare R, Shahin K, McLean K, Adams A, et al., 2021. Skin mucus proteins of rainbow trout (*Oncorhynchus mykiss*) in response to mucosal vaccination and challenge with *Flavobacterium psychrophilum*. *J Fish Dis*, 45(3), 491, <https://doi.org/10.1111/jfd.13562>
- Hussain A, Sachan SG, 2024. Evaluation of the antimicrobial efficacy of epidermal mucus extract from air-breathing fish (*Channa punctatus*) and identification of the peptides serving as immune components. *Int J Pept Res Ther* 30(4), <https://doi.org/10.1007/s10989-023-10581-8>
- Işık N, Güçlü F, Ceylan O, 2016. Parasites detected in Discus fishes (*Symphysodon discus*). *Eurasian J Vet Sci*, 32(3), 200–203. <https://doi.org/10.15312/EurasianJVetSci.2016318401>
- Kaya Öztürk D, Baki B, Öztürk R, Karayücel S, et al., 2019. Determination of growth performance, meat quality and colour attributes of large rainbow trout (*Oncorhynchus mykiss*) in the southern Black Sea coasts of Turkey. *Aquaculture Research*, 50(12), 3763–3775, <https://doi.org/10.1111/are.14339>
- Keskinoglu H, Gülel G, 2022. Determination of *Salmonella* spp. presence and antibiotic resistance in egg and egg products. *Eurasian J Vet Sci*, 38(1), <https://doi.org/10.15312/EurasianJVetSci.2022.361>
- Lee Y, Bilung LM, Sulaiman B, Chong YL, 2020. The antibacterial activity of fish skin mucus with various extraction solvents and their in vitro evaluation methods. *Int Aquat Res*, 12(1), 1–21, [https://doi.org/10.22034/IAR\(20\).2020.670998](https://doi.org/10.22034/IAR(20).2020.670998)
- Levipan HA, Avendaño-Herrera R, 2020. Assessing the impacts of skin mucus from *Salmo salar* and *Oncorhynchus mykiss* on the growth and in vitro infectivity of the fish pathogen *Piscirickettsia salmonis*. *J Fish Dis*, 44(2), 181–190, <https://doi.org/10.1111/jfd.13275>
- Levipan HA, Irgang R, Yáñez A, Avendaño-Herrera R, 2020. Improved understanding of biofilm development by *Piscirickettsia salmonis* reveals potential risks for the persistence and dissemination of piscirickettsiosis. *Sci Rep*, 10(1), 12224.
- Magnadóttir B, 2006. Innate immunity of fish (overview). *Fish Shellfish Immunol*, 20(2), 137–151. <https://doi.org/10.1016/j.fsi.2004.09.006>
- Mozumder MMH, 2005. Antibacterial activity in fish mucus from farmed fish (Master's thesis, Universitetet i Tromsø).
- Özil Ö, Diler Ö, Nazıroğlu M, 2022. Antifungal activity of some essential oil nanoemulsions against Saprolegniasis in rainbow trout (*Oncorhynchus mykiss*) eggs: Antifungal activity of essential oil nanoemulsions. *Aquaculture International*, 30(5), 2201–2212, <https://doi.org/10.1007/s10499-022-00897-5>
- Reverter M, Tapissier-Bontemps N, Lecchini D, Banaigs B, et al., 2018. Biological and ecological roles of external fish mucus: A review. *Fishes*, 3(4), 41, <https://doi.org/10.3390/fishes3040041>
- Saleh M, Kumar G, Abdel-Baki AAS, Dkhil MA, et al., 2019. Quantitative proteomic profiling of immune responses to *Ichthyophthirius multifiliis* in common carp skin mucus. *Fish Shellfish Immunol*, 84, 834–842, <https://doi.org/10.1016/j.fsi.2018.10.078>
- Saleh AA, Mohamed AZ, Elnesr SS, Khafaga AF, Elwan H, Abdel-Aziz MF, Hafez EE, 2024. Expression and immune response profiles in Nile tilapia (*Oreochromis niloticus*) and European sea bass (*Dicentrarchus labrax*) during pathogen challenge and infection. *Int J Mol Sci*, 25(23), 12829. <https://doi.org/10.3390/ijms252312829>
- Salinas I, Zhang YA, Sunyer JO, 2011. Mucosal immunoglobulins and B cells of teleost fish. *Dev Comp Immunol*, 35(12), 1346–1365, <https://doi.org/10.1016/j.dci.2011.11.009>
- Sharba S, Sundh H, Sundell K, Benktander J, et al., 2022. Rainbow trout gastrointestinal mucus, mucin production, mucin glycosylation and response to lipopolysaccharide. *Fish Shellfish Immunol*, 122, 181–190, <https://doi.org/10.1016/j.fsi.2022.01.031>
- Sridhar A, Krishnasamy Sekar R, Manikandan DB, Arumugam M, et al., 2021. Activity profile of innate immune-related enzymes and bactericidal of freshwater fish epidermal mucus extract at different pH. *Environ Sci Pollut Res*, 28, 33914–33926, <https://doi.org/10.1007/s11356-020-11173-5>
- Telli N, Telli AE, Biçer Y, Turkal G, et al., 2022. Occurrence and antibiotic susceptibility of *Vibrio* spp., *Aeromonas* spp. and *Listeria* spp. in seafoods. *Eurasian J Vet Sci*, 38(1), <https://doi.org/10.15312/EurasianJVetSci.2022.359>
- Uribe C, Folch H, Enriquez R, Moran G, 2011. Innate and adaptive immunity in teleost fish: a review. *Vet Med – Czech*, 56(10), 486–503. <https://doi.org/10.17221/3294-VETMED>
- U.S. Food and Drug Administration (FDA), 2024. Selecting and serving fresh and frozen seafood safely. U.S. Food and Drug Administration. Available at: <https://www.fda.gov/food/buy-store-serve-safe-food/selecting-and-serving-fresh-and-frozen-seafood-safely>
- Yadav R, Mishra A, 2023. A study on characteristics of fish mucus and their antifungal activity. *Res J Recent Sci*, 2277, 2502.