

Research Article

Comparative analysis of bioactive compounds and antibacterial activities of crude skin extracts from surma fish (*Rastrelliger kanagurta*) and Lal Poa Fish (*Chrysochir aureus*)

Md. Alomgir Hossain, Md. Moin Uddin¹, Md. Samsuzzaman¹, Sadia SultanaSunzida Akter Eva and Mohammad Nazir Hossain^{2*}

Department of Genetic Engineering and Marine Biotechnology, Bangladesh Maritime University,
Dhaka, Bangladesh

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ABSTRACT

Fish skin, considered a waste product in the fisheries industry, is a potential source of various bioactive compounds with antibacterial properties. In this study, we investigated the bioactive compounds and antibacterial activities of 50% ethanolic crude skin extracts from *Rastrelliger kanagurta* (Surma fish) and *Chrysochir aureus* (Lal poa fish). Qualitative screening showed the presence of alkaloids, terpenoids, saponins, and phenolic compounds with differences between the two fish species. Antibacterial activity was evaluated against *Staphylococcus aureus*, *Klebsiella* spp., and *E. coli*, using the agar well diffusion method at concentrations of 25 and 50 mg/mL. At higher concentration, Surma fish extract showed partially active antibacterial activity against *Klebsiella* spp., and *E. coli*, but Lal poa fish showed partially active against *E. coli* bacterial strain at both concentrations, possibly due to its higher phenolic content. These results indicated that fish skin could serve as a potential source of natural antibacterial agents.

Introduction

According to the Food and Agriculture Organization (FAO), forecasting total fisheries and aquaculture output to reach 197 million tons shows a 1.7% increase compared to 2024 (EFFOP, 2025). The fish processing is a major economic sector worldwide and an important source of protein and nutrients, especially in developing countries. This industry produces only 40% fish products for human consumption and more than 60% by-products as waste, such as heads, skins, trimmings, fins, frames, viscera, and roes. As a result, considerable amounts of waste are generated and contribute to environmental pollution. However, fish processing wastes are excellent rich sources of bioactive compounds,

gelatin, bioactive peptides, proteins, and collagen (Chalamaiah et al., 2012; Ideia et al., 2020). Among them, fish skin is an important part of fish body and essential component of the innate immune system and acts as a physical and chemical barrier against pathogenic microorganisms in aquatic environments (Abuine et al., 2019). It contains various secondary metabolites with significant pharmacological and biotechnological applications such as human health, nutraceuticals, food sector, and medicine (Waqar et al., 2025). Peptides and protein hydrolysates derived from fish frames, skins, viscera, and scales have demonstrated various biological significant activities, including antioxidant, antihypertensive, antimicrobial, and antidiabetic activities (Abuine et al., 2019).

*Corresponding author: <nazir@aiub.edu>

¹Department of Marine Fisheries & Aquaculture, Bangladesh Maritime University, Dhaka, Bangladesh²Department of Biochemistry and Molecular Biology, American International University-Bangladesh

In the fish processing industry, large amounts of waste biomass are discarded. Among them, a small amount of waste is processed into low-market-value products, such as animal feed, fish meal, and fertilizers, despite their high biological potential. It not only causes environmental concerns but also leads to the loss of potentially useful bioresources (Ideia et al., 2020). Therefore, the transformation of fish processing waste into high-value goods is a superior alternative. This would not only enhance the economic value of the catch but also reduce the volume of marine processing trash (Fang et al., 2017). Marine-derived natural compounds showed a remarkable antibacterial effect against drug-resistant pathogen (Bharathi and Lee, 2024). Consequently, fish by-products emerged as a vital field for the finding of new antibacterial agents.

Previous studies mainly focused on fish skin mucus extracts, fish skin collagen, fish protein hydrolysate, and fish skin gelatin (Arfat et al., 2014; Díaz-Puertas et al., 2023; Kumari et al., 2019; Baco et al., 2022). The Surma fish (*Rastrelliger kanagurta*) and Lal poa fish (*Chrysochir aureus*) are the most dominant and important marine fish species in the Bay of Bengal, Bangladesh. These fish significantly contribute to the food sector, local fish industry, and national economy.

In the fish processing industry, a large amount of byproduct is generated and discarded. However, it has potential biological importance. (Barua et al., 2026; Bhakta and Das, 2021). But, comparative bioactive chemical compound analysis and antibacterial activities of crude extracts from fish skin are tiny, especially for Surma fish and Lal poa fish. Therefore, the present study hypothesizes that crude skin extracts of these fish contain bioactive compounds with antibacterial properties. The objective of this study was to investigate and compare the bioactive compounds and antibacterial activities of crude extracts prepared from the skins of two commercially important marine fish species, Surma fish and Lal Poa fish, to evaluate their potential as natural antibacterial resources.

Materials and methods

Sample Collection Area

Fish samples of Surma fish and lal poa fish, 0.5 kg weight of each sample were collected from the local market of Muslim Bazar, Pallabi, Dhaka, Bangladesh in Fig. 1. Samples were carefully transported to the laboratory and then stored at -40°C until further use.

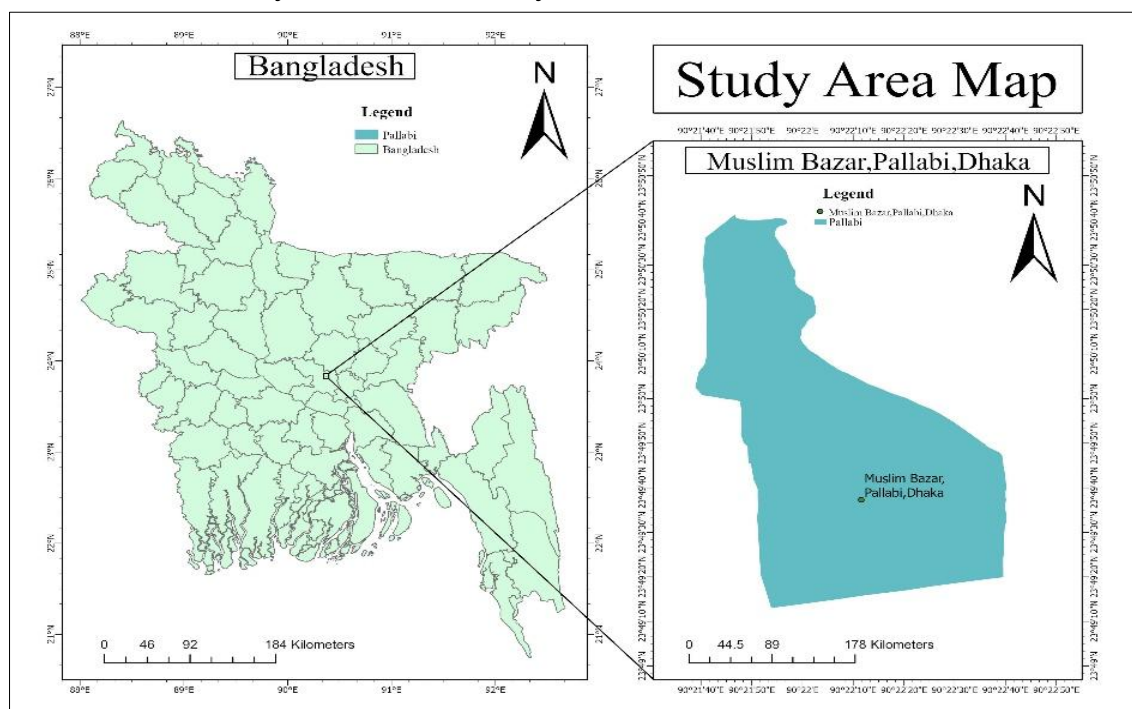


Fig. 1. Sample Collection Area.

Sample Processing

The samples were washed with distilled water and 70% ethanol to reduce microbial contamination, and epidermal tissue (skin) was then aseptically separated using sterile knife and forceps. The isolated skins were rewashed with 70% ethanol and dried in a dryer at 40°C for 72 hours until completely dry, then ground into a fine powder using a sterile mortar and pestle for further analysis (Hellio et al., 2022).

The powdered samples were weighed (Surma fish=3.64 g; Lal poa fish=1.8 g), soaked in 50% ethanol in 1:10 ratios for 4 days in a shaking incubator at 37°C, 150 rpm (Kim et al., 2019). The extract was filtered and then evaporated using a water bath at 40°C (Sapkota et al., 2020). The crude extracts were weighed and dissolved in 50% ethanol at an initial concentration of 100 mg/mL for further analysis.

Bioactive Compound Screening

The 50% ethanolic crude extracts of fish skin were qualitatively analyzed to identify key bioactive compounds, including Steroids, Terpenoids, Saponins, Flavonoids, Tannin, Phenol, Alkaloids and Carbohydrates, following established standard protocols for each assay as described below (Balamurugan et al., 2019).

Test of Steroids

To identify steroids, 200 µL of the extract was mixed with 200 µL of chloroform, then 200 µL of concentrated H₂SO₄ was carefully added. The display of red color and yellowish-green fluorescence indicates the availability of steroids.

Test of Terpenoids

300 µL of the extract was mixed with 100 µL of chloroform, and then 150 µL of concentrated H₂SO₄ was introduced along the side of the test tube. A reddish-brown color at the interface indicates the presence of terpenoids.

Test of Saponins

200 µL of extract was violently mixed with 200 µL of distilled water. The development of foam is positive for saponins.

Test of Flavonoids

- I. 100 µL of extract was aliquoted, and 10% lead acetate was included. The yellow precipitate indicates the positive result of flavonoids.
- II. About 100 µL of extract with a few drops of concentrated H₂SO₄ yields an orange color, indicates the presence of flavonoids

Test of Tanin

To 100 µL of extract, a few drops of neutral 5% ferric chloride (FeCl₃) solution were added. The formation of dark green color indicates the availability of tannins.

Test of Phenol (Lead acetate test)

To 100 µL of extract, 60 µL of a 10% lead acetate solution was added, and the mixture was gently mixed. The production of a significant white precipitate indicates the positive result of phenols.

Test of Alkaloids

- I. Mayer's test: To 100 µL of extract, a few drops of Mayer's reagent were used; the presence of a creamy or white precipitate indicates a positive result for alkaloids.
- II. Wagner's test: To approximately 100 µL of extract, a few drops of Wagner's reagent were applied. A reddish-brown precipitate indicates the presence of alkaloids.

Test of Carbohydrates

Molisch's test: To about 100 µL of the extract, two drops of an alcoholic solution of α-naphthol were added, and the mixture was subsequently mixed gently. A few drops of concentrated H₂SO₄ were added along the side of the test tube. A violet ring signifies the presence of sugars.

Antibacterial Activities

The test bacterial strains used in this study were collected from the Department of Genetic Engineering and Marine Biotechnology at Bangladesh Maritime University. Bacterial strains are *Escherichia coli*, *klebsiella* spp., and *Staphylococcus aureus*. Bacterial stains were grown in Luria Bertani (LB) agar media and standardized at 0.5 McFarland standard, and then 100 µL of the bacterial suspension was spread on Mueller Hinton (MH) agar media using a sterile cotton

swab. 50 µL of crude skin extract was added to each well (7 mm) at two concentrations, 25 mg/mL and 50 mg/mL. Here, a used negative control (50% ethanol) and positive controls (gentamicin (10 µg) and ciprofloxacin (5 µg)). Plates were incubated at 37 °C for 24 hours. The experiment was performed in triplicate, and the results were documented as the mean zone of inhibition. (Purbowatiningrum, 2022).

Statistical Analysis

Experimental data were analyzed using Microsoft Excel, and the results are expressed as mean ± standard deviation (SD) from triplicate determinations.

Result

Bioactive Compound Screening

Bioactive compound screening results of the crude skin extracts of Surma fish and Lal Poa indicated the presence of several important secondary metabolites. Table1 presents qualitative confirmation of various bioactive compounds, indicating their presence or absence using standard methods.

Table 1. Biochemical compound screening of secondary metabolites.

Secondary Metabolites	Test Name	Result	
		Surma Fish	Lal Poa Fish
Steroids	H ₂ SO ₄ Test	-	+
Terpenoids	Salkowski test	+++	Not tested
Saponins	Foam Test	+	-
Flavonoids	Acetate Test	-	-
	H ₂ SO ₄ Test	-	-
Tannins	Ferric chloride test	-	-
Phenol	Lead acetate test	-	+++
Alkaloids	Mayer's test	+++	+
	Wagner's test	+++	+
Carbohydrates	Molisch's test	-	-

Note: (-): Not Detectable; (+): Low Quantities; (++) Moderate Quantities; (+++): High Quantities.

Antibacterial Activities

The crude skin extracts of Surma and Lal poa fish showed various antibacterial activities against tested bacterial strains, including *Staphylococcus aureus*, *Klebsiella* spp., and *E. coli*. At concentration of 25mg/mL, the Surma fish extract showed inactive antibacterial activities against *Staphylococcus aureus* (8.1±0.2), *Klebsiella* spp. (9.1±0.2) and *E.coli* (9.3±0.2) indicating completely inactive antibacterial activities and Lal poa fish crude extract exhibited inactive activities against *Staphylococcus aureus* (8.0±0.0) and *Klebsiella* spp. (8.1±0.2), while showing partial activity against *E. coli* (11.3±0.2).

At a concentration of 50mg/mL, the Surma fish extract showed inactive activities against *Staphylococcus aureus* (8.6±0.5) but partially active antibacterial activity against *E.coli* (11.0±0.5) and *Klebsiella* spp. (11.0±0.0) and Lal poa crude extract showed inactive against *Staphylococcus aureus* (9.1±0.2) and *Klebsiella* spp. (8.0±0.0), while showing partially active antibacterial activity against *E.coli* (11.1±0.2). The positive control exhibited active antibacterial activity against all tested bacterial strains, and the negative control showed no inhibitory effect, thereby confirming the reliability and validity of the assay (Figs. 2 and 3).

Discussion

In this Study, evaluated the bioactive compound and antibacterial activities of crude skin extract obtained from Surma fish and Lal poa fish. Bioactive compound screening indicate that both fish species contained different significant bioactive secondary metabolites, while compound variations have both species. Surma fish extract showed the presence of alkaloids, terpenoids and Saponins, while Lal poa fish extract showed the presence of Steroids, Phenol and alkaloids. Saponins were detected only in Surma fish, whereas flavonoids, carbohydrates and tannins were absents in both fish extracts. The variation of bioactive compounds, which May influence their biological activities.

The presence of secondary metabolites such as alkaloids, terpenoids, phenols, steroids, and saponins

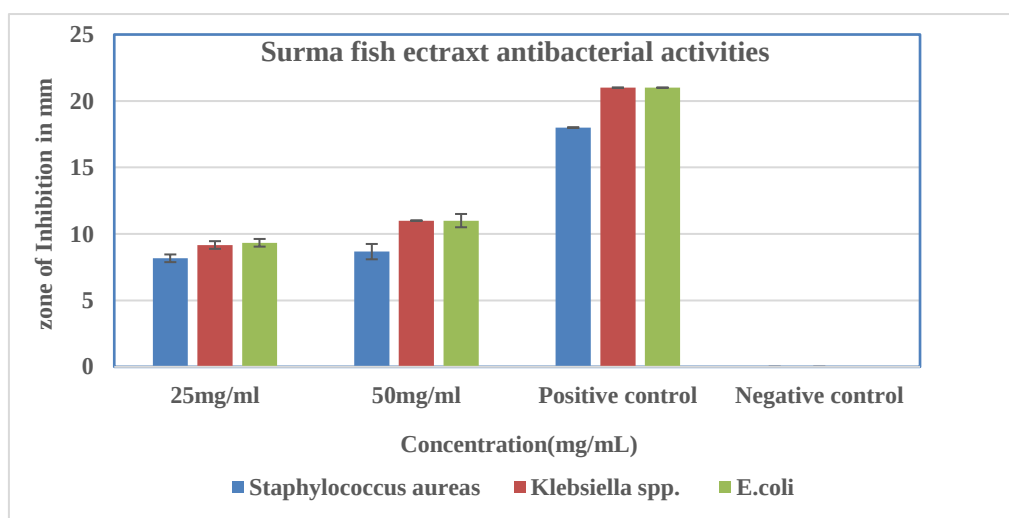


Fig. 2. Antibacterial activity of Surma fish skin crude extract against selected bacterial strains. Values are expressed as Mean±SD (n=3).

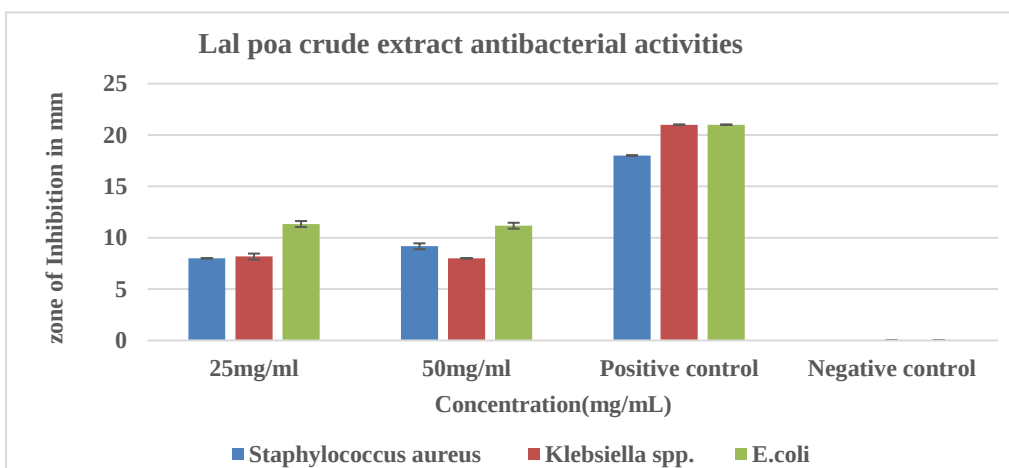


Fig. 3. Antibacterial activity of Lal poa fish skin crude extract against selected bacterial strains. Values are expressed as Mean±SD (n=3). Note: Legend: <10 mm-inactive; 10-13mm -partially active; 14-19 mm-active; >19mm- very active (Buniel, 2024).

is considered a potential bioactive chemical compound because they are usually linked with antibacterial characteristics and can demonstrate potential antimicrobial activities against harmful pathogenic microbes (Alemu et al., 2024). In this study, the presence of strong alkaloid and terpenoid compounds in Surma fish extract may explain its comparatively higher antibacterial activities, while phenolic compounds in Lal poa fish may contribute to its partial antibacterial activities. Similar studies have reported that fish-derived bioactive chemical compounds, such as protein hydrolysates and peptides,

can exhibit antibacterial, antihypertensive, antioxidative, anticancer, and anticoagulant properties due to their secondary metabolite composition (Baco et al., 2022).

According to (Buniel, 2024), both crude extract showed inactive to partially active antibacterial activities depending on concentration and bacterial strain. Antibacterial activities slightly increased at 50 mg/mL in Surma fish, particularly against *klebsiella* spp. and *E. coli*, suggesting a concentration dependent activities of the bioactive compound present in the crude extract, but overall activity was very low, especially against *S. aureus*, where both extracts were

inactive. This may arise from the structural properties of Gram-positive bacteria or an inadequate concentration of active chemicals in the crude extracts. Conversely, partial activity was observed against *E. coli* and *Klebsiella* spp. bacterial strain, indicating that Gram-negative bacteria may have a greater reaction to specific bioactive compounds found in fish skin extracts. Similarly, Alkaloids and terpenoid compounds have antibacterial activities against pathogenic bacteria (Al-Marzoqi et al., 2015). Lal poa fish extract showed comparatively more effective activity against *E. coli* than Surma fish, likely because of its higher phenolic content. At both concentrations, *E. coli* was partially active, but *S. aureus* and *Klebsiella* spp. showed inactive activity. A previous study reported that the presence of phenolic compounds has antibacterial activities and can disrupt bacterial proteins and cell membranes (Maddox et al., 2010).

Despite the weak antibacterial activity, the study suggests that the fish crude skin extract from Surma fish and Lal poa fish contains various secondary metabolites with antibacterial properties and provides preliminary comparative information on the bioactive compounds profile and antibacterial activities of crude skin extracts from these commercially important fish species.

Conclusion

Fish skin, commonly observed as a waste product and crude extract from Surma fish and Lal poa fish, contains different type of bioactive compounds such as saponins, phenolics, alkaloids and terpenoids etc. both fish extracts showed antibacterial activities against various pathogenic bacterial strains, indicating that fish skin, may be a potential bioresource of natural antibacterial agents for medical and biotechnological applications. Further research focused on the isolation and characterization of bioactive compounds is essential to properly evaluate their medicinal potential.

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Authors contribution

Md. Alomgir Hossain: Data curation, Formal analysis, Investigation, Methodology, Validation, Visualization, Writing – original draft, Writing – review & editing; Md. Moin Uddin: Investigation, Methodology, Validation, Visualization, Writing – review & editing; Md. Samsuzzaman: Investigation, Methodology, Validation, Visualization, Writing – review & editing; Sadia Sultana: Investigation, Writing – review & editing; Sunzida Akter Eva: Investigation, Writing – review & editing; Mohammad Nazir Hossain: Conceptualization, Funding Acquisition, Methodology, Project Administration, Resources, Supervision, Writing – review & editing.

Conflict of interest

The authors declare no conflict of interest.

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