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Fish Epidermal Mucus as a Source of Diverse Therapeutical Compounds

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Abstract

Microbes are helpful and destructive to human health and other living organisms. Microbes can be eliminated by using antibiotics against them, but their capability to resist regularly encountering antibiotics makes them more injurious. Microbes can adjust and adapt according to the chemicals used against them and become antibiotic resistant. Thus, the requirement for novel antimicrobial compounds increases with time to treat antibiotic-resistant microbes. Fish epidermal mucus encounters various pathogens present in their surrounding environment. It has become a rich source of novel antimicrobial compounds mainly antimicrobial peptides that can be used against various antibiotic-resistant pathogenic microbes. Compounds extracted from epidermal mucus can be used synergistically with other antibiotics or resistance modifying agents to inhibit the growth of resistant microbes. Fishes are consumed as a protein-rich food source worldwide and contribute to the world economy. Diseases in fish cause significant losses in the economic benefits exploited by fishermen and industries based on fisheries products. This paper will review compounds from fish epidermal mucus and their use to control the growth of antibiotic-resistant or non-resistant pathogenic microbes of humans and fishes. So, to increase fisheries' economic benefits and decrease infections involving resistant microbes.

Keywords Antimicrobial compounds · Antimicrobial peptides · Antibiotic-resistant · Epidermal Mucus · Synergy

Introduction

Antibiotic resistance occurs when microbes like bacteria and fungi are exposed to a specific antibiotic for an extended period. Microbes develop the potential to defeat the antibiotics used to kill them. Intensive use of antibiotics in veterinary and human healthcare sectors without proper knowledge of the dosage used and unnecessary antimicrobials in agricultural practices promote antibiotic resistance globally (Singhal 2022). Antibiotic-resistant bacteria have been a severe threat in the last three decades as many pathogenic and non-pathogenic microbes have become antibiotic-resistant (Ventola 2015). Nearly 1.3 million deaths were associated with bacterial antimicrobial resistance in 2019 only. 9,29,000 deaths were caused by six antibiotic resistance microbes (Murray et al. 2022). After the emergence of COVID-19, the use of antibiotics spikes abruptly, which is a severe cause of concern in terms of antibiotic resistance (Mohamad et al. 2022). Antibiotics inhibit the growth and development of microbes by interfering in the synthesis of cell walls and their central dogma pathways to obstruct the synthesis of protein, RNA, and DNA (Neu 1992). Antibiotics also interrupt the division of microbes to inhibit their growth (Bennett et al. 2014). Microbes develop intrinsic factors to cope with antibiotics' effects through mutation and DNA conjugation. Conjugation of DNA occurs with various microbes to get antibiotic-resistant genes or multiple drug-resistant genes (Munita et al. 2016). There are two ways to tackle resistance in microbes. First is modification in existing antibiotics and formation of different generations of antibiotics. Another way is to explore novel antimicrobial compounds from various biological sources. These compounds can act more effectively on the resistant microbes (Chen et al. 2020).

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Emergence of resistance in microbes increases the demand for new antimicrobials. Fishes in aquatic environments are in continuous contact with pathogenic microbes, enabling them to build an immune mechanism to protect themselves. Fishes possess many antimicrobial compounds in different parts of their body as a component of their immune system. The first line of defense against microbes present in water is epidermal mucus. The skin mucus is a physical and chemical barrier to invading microbes. Skin mucus is rich in mucin and other viscous colloids along with many enzymes providing antimicrobial nature to the mucus. Enzymes such as proteases, lectins, AMPs (Antimicrobial peptides), lysozymes, alkaline phosphatases and immunoglobulins are some of the components of mucus that provide antimicrobial nature to the mucus (Dash et al. 2018; Angeles Esteban 2012). Compounds extracted were reported to be effective against many pathogenic, opportunistic and resistant microbes. On further purification and characterization, the compounds could be used commercially in many forms for pharmaceutical uses (Rao et al. 2015).

This review highlights the concern for resistance in microbes and focuses on the way to tackle those resistant microbes using antimicrobial compounds present in fish mucus. Further, the role of compounds extracted from fish mucus in therapeutics against various pathogenic and opportunistic microbes of fishes and humans has also been discussed.

Antibiotics Mechanism of Action and Resistance

Antibiotics are chemicals from organisms like bacteria and fungi that inhibit other microbes' growth. Chemically these are polypeptides, amino sugars, macrolides and various other compounds (Waksman 1961). Antibiotics act not on the host but on the foreign cells, such as invading microbes. Antibiotic selectivity is based on the difference in the host cell's physiological and biochemical functions and pathogenic cells (Skold 2011).

Antibiotics have three significant ways to inhibit microbial growth (Neu 1992). Firstly, by inhibiting wall synthesis, antibiotics such as penicillin and cephalosporins act as a pseudo substrate and deacetylase the peptide linkage of the peptidoglycan layer of the microbial cell wall (Spratt and Cromie 1988). Another antibiotic family, vancomycin, ties up with the peptide substrate and prevents the microbial walls' peptidoglycan layer's crosslinking. The second mode of inhibition is by blocking the machinery of protein synthesis. Antibiotics such as erythromycin, tetracyclines, and kanamycin interrupt protein synthesis steps (initiation, elongation, and termination). All the steps are highly enzyme-regulated, antibiotics block the active sites of active enzymes and prevent the synthesis of proteins (Fourmy et al. 1996). The third mode of inhibition is by interrupting the DNA replication mechanism, and various enzymes are involved in the replication process. Antibiotics block a specific enzyme's functioning to prevent DNA replication. Fluoroquinolones are the synthetic antibiotic structure that inhibits topoisomerase's functioning, and topoisomerase prevents the supercoiling of DNA by cutting, uncoiling and ligating the two DNA strands. Fluoroquinolone blocks the ligating activity of topoisomerase and inhibits DNA replication (Rosen 1990).

Antibiotic resistance was reported after the discovery of antibiotics. Antibiotic resistance is observed in various microbes that resist a specific antibiotic drug commonly used against them. Antibiotic resistance occurs due to the constant exposure of an antibiotic medication to a microbe. Due to penicillin's intensive use, antibiotic resistance to penicillin was observed during the early 1940s, just a few years after its discovery. In the case of vancomycin, another antibiotic drug, resistance was observed after almost 3 to 4 decades of its discovery, as the use of vancomycin was not that frequent after it was discovered. When exposure to vancomycin increases, resistance against vancomycin is also observed. The number of antibiotics discovered so far was very few, and due to the lack of antibiotic discoveries from the 1960s to the 2020s, reliance on the discovered antibiotics has increased, and resistance to those antibiotics has been observed. This means that after prolonged exposure, microbes adapt and resist the drug that was effective against them earlier (Giedraitiene et al. 2011). In the past two years, after the emergence of the Coronavirus Disease of 2019 (COVID-19), antibiotics have caused a ripple in the antibiotic resistance problem (Rizvi and Ahammad 2022). The primary reasons for antibiotic overuse are the higher rate of infections due to poor healthcare discarding, poor water quality, low immunization, and malnutrition. Easy availability of antibiotics and poor prescriber knowledge or over-the-counter (OTC) purchase of antibiotics also trigger the overuse of antibiotics (Singhal 2022).

Resistance occurs to prolonged exposure and overuse of antibiotics in many ways, such as antibiotic modification, antibiotic efflux, conjugative transferase, and target site modification and hiding. Different antibiotics showed a specific method for the evolution of resistance in them, and resistance comes about by modification in three major classes of antibiotics β-lactam, aminoglycoside and chloramphenicol (Iovine 2013). Antibiotics such as macrolides, tetracyclines and fluoroquinolones are pumped out by efflux pumps on the membrane of resistant bacterial cells (Džidić et al. 2008).

Antimicrobial Compounds from Fish Mucus

Fishes are an essential part of the ecosystem as well as of the economic market. Fishes are used as food, and their products such as, fish liver oil, gelatin, fish albumin, fish protein concentrates, glue, protamine etc., are widely procured worldwide. In 2018-19, the quantity of fish produced was twice that of poultry and three times that of cattle. In the last two decades, the fisheries sector contributed to employment mainly in Asian countries, and the products from fisheries increased 527% from 1990 to 2018 (FAO, 2020). Hence, there is an excellent contribution of fish and aquaculture products to the economy. The benefits of fish in the healthcare sector are well known as fish meat provides Omega 3 fatty acids, vital minerals, and proteins. Apart from this, an unfocused portion of fish is epidermal mucus. Epidermal mucus contains many antimicrobial compounds. Here, we pinpoint those compounds and their application in the healthcare sector.

Fish encounter many pathogens in the aquatic ecosystem, which causes various diseases, leading to a decrease in fish production and, hence, decreasing fish and aquaculture's economic contribution to the world economy (Béné et al. 2016). In the aquatic ecosystem, fishes' first line of defense against pathogens is the epidermal layer mucus. The epidermal layer's mucus composition varies from fish to fish as the mucus-producing cells differ in different fish species. The epidermal layer contains two types of cell stem lines: a proteinaceous cell stem line and a mucus cell stem line. The mucus cell stem line produces three different types of cells: mucus cell or goblet cell, sacchiform cell, and club cell. These three cells produce mucus in different fish species (Shepherd 1993; Zaccone et al. 2001). In studies, Malpighian cells or filament-containing pavement cells were also reportedly involved in epithelial mucus production (Whitear 1970). Mucus is a slippery and slimy layer that covers the fish and protects the fish from invading pathogens. It protects the fish from various pollutants such as surfactants, detergents and heavy metals (Arasu et al. 2013). Mucus also provides the function of respiration, ionic balance and conductance, osmotic regulation, reproduction and excretion (Ingram 1980). Mucus confronts the pathogen present in the aquatic environment and develops various immune responses against them. Mucus contains mucin, a macromolecular glycoprotein gel that reduces friction between fish and the aquatic environment and plays a vital role in immunity (Austin and McIntosh 1998). Mucin is rich in sialic acid, neutral glycoproteins, threonine and proline (Rose and Voynow 2006). Mucus antimicrobial compounds are one of the immune responses generated by the epidermal layer of fishes. Antimicrobial compounds extracted and purified from the mucus contain various classes of compounds such as antimicrobial peptides, pectinase, lysozyme, alkaline phosphatase, cathepsin, lectins and proteases are potent compounds used against diverse pathogens (Arockiaraj et al. 2013). Mucus from some fishes also possesses amphipathic helical peptides like dermaseptin, ceratotoxin, and magainin that act as detergent for the membrane and dissolves the membrane by binding with anionic phospholipids, which results in lysis of cells (Mat Jais et al. 2008; Pickering 1974).

The amount of mucus and biochemical compounds present in mucus is affected by the temperature, pH and salinity of water where the fish inhabit. As observed in gilt-head sea bream, the production of peptides in mucus changes with temperature ranging from (22 – 14) °C as the stress caused by the difference in temperature triggers the physiological system of fishes, such as the hypothalamus-pituitary interrenal axis, to produce hormones and excessive mucus (Sanahuja et al. 2019; Iger et al. 1995). Change in pH ranging from (5–9) and salinity stimulates rodlet cells to produce mucus and enzymes such as alkaline phosphatase. Alkaline pH stimulates the mucus cells to produce more mucus, but mucus peptides' deterioration occurs at alkaline pH. Comparatively, at acidic pH, mucus and alkaline phosphatase production in mucus increases without damage (Iger et al. 1997; Al-Arifa 2011). The variation in enzyme activities is also thought to be related to the thickness of the epidermis, and a thicker epidermis has a more significant frequency of mucus cells producing different enzymes such as alkaline phosphatase, proteases and lysozyme. The mucus composition and the antimicrobial component of mucus vary from species to species as the mucus-producing cell type, surrounding conditions (such as pH, temperature and salinity) tolerance changes as species differ (Iger et al. 1997; Fast 2002). The antimicrobial compounds found in the epidermal mucus of fish can be used against many pathogenic microbes resistant to various antibiotics. Synergism of compounds from mucus and old antibiotics or modifying agents would show effective results against resistive microbes (Gibbons et al. 2003). A description of the major immune components found in fish mucus is now enumerated.

Antimicrobial peptides

AMPs are generally positively charged molecules with a low molecular weight with variations in biochemical properties, chain length, structure and amino acid sequences. AMPs showed activity against all microbes, bacteria, fungi, parasites, and viruses. They were reported as inhibitors of central dogma processes and blocked the synthesis of DNA, RNA and proteins (Patrzykat et al. 2002). First AMP reported from fish epidermal mucus is a 33-residue paradaxin extracted from *Pardachirus marmoratus*.

Paradaxin is an α-helical peptide that effectively inhibits the growth of microbes (Oren and Shai 1996). A 51-residue peptide hipposin extracted from *Hippoglossus hippoglossus* skin mucus actively inhibits bacterial growth (Birkemo et al. 2003). Cystine-rich AMPs (cathelicidins, liver-expressed AMPs and defensins) were also reported from fish epidermal mucus, which effectively inhibits the growth of gram-negative and gram-positive bacteria (Ángeles Esteban 2012). Some ribosomal AMPs were also reported from fish mucus effective against gram-positive bacteria (Fernandes et al. 2002). H2A and H6 peptides of histone extracted from the epidermal mucus of *Oncorhynchus mykiss* effectively inhibited bacterial and fungal growth (Fernandes et al. 2002; 2003). Pleurocidin, which is a 25-residue peptide similar to dermaseptin and ceratotoxin, was extracted from *Pleuronectes americanus* epidermal mucus and tested against various pathogenic microbial strains (Cole et al. 1997). *Epinephelus coioides* mucus was found to contain epinecidin-1 a 67 amino acid multifunctional AMP reported as an antibacterial, antifungal and anti-tumor agent (Neshani et al. 2019). In a study by Patel et al. (2020), it was observed that different classes of bioactive molecules (small peptides, sphingolipids and fatty acids) having antibacterial properties were purified and characterized from the epidermal mucus of *Puntius sophore*. Epidermal mucus extract of *Takifugu pardalis* (puffer fish species) contains a novel 23 amino acid hepcidin type −2 like peptide that inhibits the growth of gram-negative and gram-positive microbes (Go et al. 2019). Pscidin-4 is a novel peptide of 44 residues in the pscidin family extracted from hybrid striped bass (*Morone chrysops* female x *M. saxatilis* male). Pscidin-4 showed activity against 11 fish pathogenic bacteria and 6 human pathogenic bacteria (Noga et al. 2009). So, AMPs from fish mucus could be developed into drugs and antibiotics for fish and human diseases (Rakers et al. 2013).

Lectins

Lectins are carbohydrate-binding proteins or glycoproteins that agglutinate cells and glycoconjugates present in cells. There are various forms of lectin identified from fish skin mucus, including congerins, galectins, intelectin, pufflectin and nattectin etc, showed effective inhibition of microbes. Some Ca^{+2} dependent mannose-binding lectins that bind to the surface of pathogens were also reported from *Silururs asotus* (Tsutsui et al. 2011). Pufflectins were extracted from pufferfish and reported binding with a parasitic trematode *Heterobothrium okamotoi*. AJL-2 a novel lectin from eel's skin mucus was extracted and tested against *E coli* by Suzuki et al. (2003).

Proteases

Proteases have many types in terms of catalytic activity, fish epidermal mucus possesses serine, cystine and aspartic proteases. All these proteases were reported effective in killing microbes by cleaving the peptide bonds of microbial walls. Proteases activate innate immune components like immunoglobulins, complements or AMPs (Aranishi 1999). A minute amount of anti-protease was also reported from the epidermal mucus of fish *Amphiprion clarkii* (Wang 2019).

Lysozymes

Lysozymes are polypeptides and a well-known innate immune component reported from many sources like humans, animals, plants, bacteria and viruses. Lysozymes are the most common compound reported in the epidermal mucus of fish by many researchers all over the globe (Ángeles Esteban 2012). The ability of lysozymes to degrade the bond between N-acetyl glucosamine and N-acetylmuramic acid enables them to degrade microbial walls. Lysozymes were also reported to act on chitin of fungal walls (Wu 1999) and on viruses (Lee-Huang et al. 1999). Fish mucus contains c-type and g-type lysozymes, which effectively eliminate the pathogenic and opportunistic microbes (Qasba et al. 1997). Secretions of lysozymes are linked with various factors like stress, wounds and diseases. So, the amount of lysozyme secretion may indicate the health status of the fishes (Dash et al. 2018). Along with mentioned four major classes of antimicrobial compounds present in mucus, some other components such as metabolites, amino acids and fatty acids were also found in the mucus. These components also contribute to antifungal and antibacterial activity of mucus (Uthayakumar et al. 2012).

Fish mucus is a rich source of various novel bioactive molecules and antimicrobials such as AMPs, proteases, fatty acids, lysozymes and galectins. So, fish mucus could be served as a potent source of novel antimicrobial compounds that can help combat antibiotic resistance in microbes.

Use of Antimicrobial Compounds from Fish to Resist the Growth of Antibiotic-resistant Microbes and Aquatic Pathogenic Microbes

In various studies, mucus antimicrobial activity was observed due to the presence of one or more than one antimicrobial compounds in the mucus of fish. All the compounds present in the mucus can be used against many bacteria, "gram-positive and gram-negative," as well as against some fungi. Growth inhibition was observed in studies using mucus of different fish species against various microbes. Three novel antibacterial proteins showed pore-forming activity against microbes such as *Staphylococcus aureus* and *Aeromonas hydrophilla.* The proteins were purified from three separate fish species trench, eel and rainbow trout, and the masses of novel antibacterial proteins are 49kDa, 45kDa and 65kDa, respectively. The pore-forming activity was observed using a planar lipid bilayer (PLB) under the influence of applied voltage, when proteins pass through the PLB, their action was measured in terms of conductance (Ebran et al. 2000). Antimicrobial peptides (AMPs) from fish epidermal mucus were reported effective against many pathogens of fish and humans. It was also reported that AMPs from *Channa striatus* effectively treat wound healing by clotting blood at the wound to prevent bleeding and tissue formation using amino acids and fatty acids from epidermal mucus (Zakaria et al. 2007). AMPs were observed to be useful against several viral infections in fishes, such as viral hemorrhagic septicemia virus in Rainbow trout. It shows effective inhibition by beta-defensins and nervous necrosis virus in Medaka epinecidin-1 (Falco et al. 2007). Another AMP hipposin extracted from Atlantic halibut epidermal mucus showed the inhibition of *Enterococcus fecalis, Listeria ivanovii* and *Staphylococcus epidermis*, degrading the microbial walls (Birkemo et al. 2003). Proteases such as cathepsins observed in the epidermal mucus of Japanese eel showed bactericidal activity against pathogenic microbes like *Listonella anguillarum, Edwardsiella tarda and Flavobacterium columnare* (Aranishi 1999). In *Pardachirus marmoratus*, 33 residues paradaxin were extracted from skin mucus, these peptides have a helical structure similar to mellitin and showed pore-forming activity against metazoan membranes (Dash et al. 2018; Oren and Shai 1996). A unique epidermal lectin of 35KDa was extracted from *Silurus asotus* skin mucus and showed bacterial agglutination of *Aeromonas salmonicida* (Tsutsui et al. 2011).

Galectins like congerin were also reported from fish epidermal mucus, which exhibits haemagglutination properties in rabbits, sheep and horses (Shiomi et al. 1989). Lysozymes are the most commonly found antimicrobial compound in fish epidermal mucus. Lysozymes show effective inhibition of many microbes by hydrolyzing the bond between different sugar residues of N-acetyl glucose amine (NAG) and N-acetyl muramic acid (NAM) of microbial walls (Qasba et al. 1997). In some fishes, the compounds in mucus responsible for antimicrobial activity were purified, whereas some compounds must be purified. As in *Channa punctatus* and *Cirrhinus mirgala*, antimicrobial activity was observed against nine different bacterial genera, but the compounds responsible for the antimicrobial activity are still unknown (Kuppulakshmi et al. 2008). On the other hand, many compounds were purified from fishes' mucus, which showed effective microbial growth inhibition, as shown in (Table 1).

Antibiotic-resistant microbe such as *Staphylococcus aureus* shows resistance against many antibiotics such as Vancomycin, Penicillin, Daptomycin, Tetracyclines, Chloramphenicol, Florfenicol, Methicillin and Oxacillin (Chen et al. 2020; Foster 2017). Fish mucus and antimicrobial components of mucus show effective inhibition of *Staphylococcus aureus* growth in many studies (Ramesh 2013; Kumari et al. 2019). Inhibition of Methicillin-Resistant *Staphylococcus aureus* (MRSA) was observed in a study on crude extract, organic extract, and acidic extract of four different freshwater fish species. Acidic extract of Bagrid catfish and Tilapia showed effectual inhibition of MRSA. The other two fishes, Giant snakehead and Stripped snakehead showed poor antimicrobial activity (Rao et al. 2015). In another study, antibiotic resistance of penicillin and chloramphenicol in *Streptococcus pneumoniae* was observed in 1965 and could be treated through antimicrobial mucus components (Appelbaum 1992). Resistant methicillin was observed in *Staphylococcus epidermidis* isolated from dental plaque (Tang et al. 2020). The mucus of carp species was found to treat Methicillin-Resistant *Staphylococcus epidermidis* (MRSE) (Kumari et al. 2019).

Several microbes are pathogenic, which causes the loss of aquaculture products and affects the health of fishes. It was detected that *Cocci*, *Enterobacteriaceae*, *Vibrio* and *Aeromonas* proved to be pathogenic to *Oncorhynchus mykiss* (Rainbow trout). Rainbow trout is native to the Pacific Ocean and edible in around 45 countries worldwide (Diler et al. 2000). Mucus of *Hippoglossus hippoglossus* showed effective inhibition of these microbes by using a histone H2A derived AMP hipposin. Hipposin disrupts the lipid bilayer glycoproteins, leading to microbial cells' death (Birkemo et al. 2003). In another study, it was observed that *Aeromonas*, *Pseudomonas*, *Bacillus* and *Enterobacteria* were pathogenic in *Coregonus Albula* (staple food of Finland), which were then treated by using mucus of fishes such as *Hypophthalmichthys nobilis*, *Ctenopharyngodon idella* and *Cyprinus* (Kumari et al. 2019; Zmysłowska et al. 2001). *Salmonella*, *Escherichia coli*, *Klebsiella*, *Pseudomonas aeruginosa* and *micrococcus* were also found to be pathogenic to *Tilapia nilotica* (Youssef et al. 1992). These pathogenic microbes present in *Tilapia nilotica* can be treated using mucus of *Hippoglossus* fish, *Oncorhynchus mykiss* and *Channa striatus* (Birkemo et al. 2003; Ramesh 2013). So, it is proposed that these compounds present in the mucus of fishes could fulfill the requirement of new antimicrobials and can also be considered necessary in fighting antibiotic resistance (Fig. 1).

Table 1 Activity of extracted compounds from mucus against gram-positive and gram-negative bacteria

Fish species	Extracted compound type	Activity against		Reference
		Gram +	Gram -	
Rita rita	Proteases		*E coli* *P aeruginosa* *S typhi*	Hjelmeland 1983
Trout	Pore-forming antibacterial protein	*Micrococcus luteus* *M varians* *S capitis* *S xylosus*	*A hydrophilla*	Ebran et al. 2000
Anguilla japonica	Amino proteases And Cathepsin D	*Micrococcus lysodeikticus*		Aranishi and Mano 2000
Hippoglossus hippoglossus Atlantic halibut	Peptides hipposin	*B subtilis*	*E coli* *V anguillarum*	Birkemo et al. 2003
Sobastes schlegeli Rabbit fish	Antimicrobial proteins		*A hydrophilla*, *A salmonicidia*, *P damselae* *V parahaemolyticus* *Shewanella putreficans*	Nagashima et al. 2001
Salmo salar (Atlantic salmon)	SAMPH1	*B subtilis*, *Listeria ivanovii*	*Aeromonas*, *E coli*, *V anguillarum*, *S enteric*	Luders 2005
Catla catla *Labeo rohita*	Antimicrobial peptide		*P Aeuroginosa* *S typhi* *K pneumonia* *E coli* *V cholera*	Balasubramanian et al. 2012
Oncorhynchus mykiss (Rainbow trout)	Antimicrobial peptide	*S aureus*	*Proteus mirabilis* *Salmonella* *Vibrio*	Ramesh 2013
Three Carp species *H nobilis* *C idella* *C caprio*		*S aureus* *B cereus* *S epidermidis*	*P Aeuroginosa* *K pneumonia* *E coli* *A hydrophilla*	Kumari et al. 2019
Puntius sophore	Fatty acids, lipids, amino sugars, amino alcohols, small peptides, etc.,	*S aureus* *B subtilis*	*P aeruginosa* *E coli*	Patel et al. 2020
Catla catla *Channa striatus*		*B coagulans* *S aureus*	*P aeruginosa* *K pneumonia* *E coli* *A hydrophilla* *Proteus vulgaris*	Ranjini et al. 2020
Anabas testudineus	AtMP2 peptide	*B cereus* *B subtilis* *S aureus*	*P aeruginosa*	Najm et al. 2021
Monopterus albus		*S aureus* *Streptococcus pyogenes*		Hilles et al. 2022
Cyprinus carpio	Defensin-like protein 1	*B cereus*	*A sobria* *Enterobacter kobei* *Leclercia adecarboxylata* *K pneumonia*	Shabir et al. 2022
Heteropneustes fossilis *Clarias batrachus*		*B cereus* *S aureus* *S epidermidis*	*P aeruginosa* *K pneumonia* *E coli* *A hydrophilla*	Bhatnagar et al. 2023

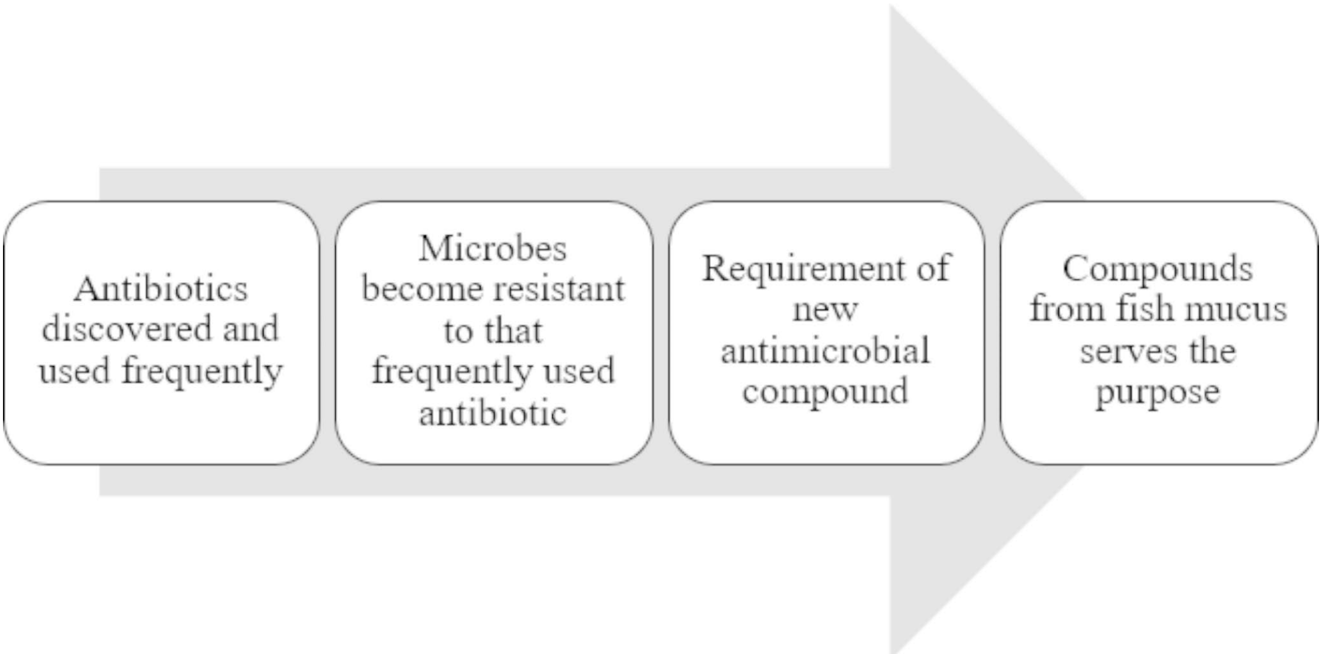

Fig. 1 Importance of naturally occurring antimicrobial compounds from fish mucus

Synergy of Antimicrobial Compounds to Overcome Resistance

The combination of antibiotics showed synergistic benefits. Newly discovered antimicrobial compounds combined with primitive ones to unleash the true potential of that freshly found antimicrobial compound. Microbes found to be resistive to an antibiotic show effective synergy with an antimicrobial compound, which is also not that practical without the synergistic effect of that resistive antibiotic. Combinations of β-lactams such as penicillin G or ampicillin with aminoglycosidase such as streptomycin or gentamycin showed efficient inhibition of β-lactam-resistant lactobacilli (Bayer et al. 1980). The mechanism behind the effect was not so precise in the case of β -Lactam resistant lactobacilli, but resistance in *Staphylococcus aureus* against quinolones and antiseptics was well explained by Stermitz et al. (2000). *Staphylococcus aureus* possesses a multidrug-resistant (MDR) pump NorA which acts as an efflux pump for cationic antimicrobials or quinolones. Several *Berberis* medicinal plants produce 5-methoxyhydnocarpin (5-MHC), which inhibits the efflux of berberine by binding with NorA MDR pump. Berberine alone hardly showed any inhibition, but in synergy, with (5-MHC) minimum bacterial resistance (MBR) was reduced, and the inhibition by berberine was satisfactory (Stermitz et al. 2000). In the case of Methicillin-Resistant *Staphylococcus aureus* (MRSA), extract of *Lycopus europaeus* yielded two new isopimarane diterpenes, namely methyl-1α-acetoxy-7α 14α-dihydroxy-8,15-isopimaradien-18-oate and methyl-1α,14α-diacetoxy-7α-hydroxy-8,15-isopimaradien-18-oate that blocks the MDR pumps of MRSA such as TetK, MsrA and NorA. Tetracycline and erythromycin synergism with these isopimarane diterpenes blocking the efflux pumps show a two-fold reduction in both the antibiotics' minimum inhibitory concentrations (MICs) (Gibbons et al. 2003).

Conclusion

Various antimicrobial compounds in the fish mucus can be purified and used against pathogenic microbes that adapt or modify to resist antibiotics of any type. Antibiotics are modified slightly (structurally or chemically) when the microbes become resistant to them. For example, generations 1, 2, 3, and 4 antibiotics (Penicillin: Penicillin G, Amoxicillin, Ticarcillin, and Piperacillin, respectively); all these generations of antibiotics have slight differences in their structure. When microbes become resistant to a specific antibiotic compound and its various generations, a new compound with antimicrobial properties is the best-known option. We can observe that in the last 3–4 decades, only 2–3 new antibiotics were discovered, and lack of discovery ultimately promotes resistivity in microbes. So, the resistivity of microbes increases as the availability of antibiotics is more petite. Modifications in antibiotics have been used so far, but it's not a permanent solution for antibiotic-resistant microbes. Antimicrobial compounds from fish mucus can act as an ideal replacement for antibiotics and can provide novel antimicrobial compounds that effectively prevent resistant microbes' action. Antimicrobial compounds from fishes are reported to be effective against aquatic pathogens, human pathogens and resistant microbes like MRSA and MRSE. Compounds extracted from mucus can also be used in synergism with old antibiotics and with resistance-modifying agents. Compounds extracted from mucus can also be used in therapeutics and aquaculture to prevent aquatic organisms' loss due to pathogenic microbes, ultimately enhancing aquaculture products.

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Declarations

Conflict of Interest The authors declared that they have no conflict of interest.

Ethical Approval and Consent to Participate This article does not contain any studies with human participants or animals performed by any of the authors.

Consent for Publication Not applicable.

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