Molecular Mechanisms of Antioxidant Action in Marine-Derived Peptides

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ABSTRACT

Marine ecosystems provide a rich and diverse source of bioactive proteins and peptides with notable health-promoting properties. Although the vast biodiversity of marine life is immense, research has predominantly concentrated on a limited group of organisms, including fish, mollusks, crustaceans, algae, sponges, and marine microorganisms. Fish stand out as a particularly valuable source of high-quality proteins, such as collagen, myoglobin, and actin, which are obtained not only from edible tissues but also from processing by-products. Additionally, other marine organisms like sea cucumbers, shellfish, and seaweeds have emerged as promising alternatives for collagen extraction, offering sustainable and versatile applications in health, cosmetics, and biomaterials. Marine-derived peptides exhibit a wide range of biological activities, including antioxidant, antihypertensive, antimicrobial, immunomodulatory, anticancer, and enzyme-inhibitory effects. For instance, peptides from shellfish possess angiotensin-converting enzyme (ACE) inhibitory activity that aids in blood pressure regulation, while antimicrobial peptides (AMPs) isolated from various marine species demonstrate broad-spectrum activity and excellent biocompatibility, making them attractive for pharmaceutical and food preservation uses. Among these bioactivities, antioxidant effects have been extensively studied. Marine peptides can neutralize reactive oxygen species (ROS), chelate metal ions, and enhance the body's antioxidant defense systems, thus mitigating oxidative stress linked to chronic diseases such as cardiovascular disorders, diabetes, and cancer. This antioxidant potential is attributed to specific amino acids that promote free radical scavenging and interaction with cellular membranes. Numerous studies report that protein hydrolysates derived from marine sources—including sea cucumbers, shrimp shells, krill, and fish muscle—exhibit strong antioxidant effects both in vitro and in biological models. These findings underscore the promising potential of marine-derived peptides as natural bioactive compounds for developing functional foods, nutraceuticals, and therapeutic agents aimed at improving human health.

Keywords:* : *Marine-derived peptides, Antioxidant activity, Bioactive compounds, Functional foods, Collagen extraction, Antihypertensive effects.

INTRODUCTION

Covering over 70% of the Earth's surface, marine environments represent one of the planet's most vast and diverse ecological systems. These habitats support an extraordinary array of life forms—from microscopic phytoplankton to the largest marine mammals—all of which have undergone remarkable evolutionary adaptations to survive and flourish in harsh and often extreme oceanic conditions. Such adaptations have led to the production of unique bioactive compounds, particularly peptides and proteins, characterized by exceptional structural complexity, functional diversity, and *biochemical properties that set them apart from their terrestrial plant-derived counterparts [1,2].Proteins are fundamental macronutrients that not only serve as a significant energy source, but also supply the essential amino acids required for key physiological processes, including tissue growth, cellular development, and overall maintenance of bodily functions, derived from these proteins, bioactive peptides—short chains of amino acids—are increasingly recognized for their diverse biological activities, these peptides are believed to exert important functional and metabolic effects, making them critical contributors to human health and homeostasis [1]. Marine-sourced bioactive peptides generally consist of between 3 and 40 amino acids, with the majority falling within the 2 to 20 range. These peptides remain biologically inactive within the structure of their parent proteins until they are released through processes such as enzymatic hydrolysis, gastrointestinal digestion, or microbial fermentation. Once liberated, their specific sequence, amino acid composition, and molecular weight determine their biological activity. These peptides engage with cellular receptors, enzymes, and ion channels, influencing a wide range of physiological pathways. Notably, marine-derived peptides can trigger programmed cell death through diverse mechanisms, including apoptosis, disruption of microtubule dynamics (anti-microtubule effects), inhibition of blood vessel formation, suppression of cell proliferation, and direct cytotoxic actions [3].Key marine organisms—including fish, seaweeds, shellfish, microalgae, crustaceans, cephalopods, mollusks, and their processing by-products—represent valuable reservoirs of proteins and bioactive peptides. These marine-derived compounds have attracted growing interest due to their wide-ranging biological activities, which include antioxidant, antihypertensive, antidiabetic, anticoagulant, antimicrobial, anti-inflammatory, anticancer, and immune-modulating effects. Owing to this broad spectrum of functionality, marine peptides and proteins are increasingly viewed as promising candidates for innovative uses across the food, pharmaceutical, and cosmetic industries [4]. What sets marine-derived peptides and proteins apart is their distinctive structural and functional profile, shaped by the extreme and variable conditions of their native environments. Unlike compounds from terrestrial sources, these marine molecules often exhibit enhanced stability, improved bioavailability, low toxicity, and environmentally sustainable production. Marine life forms must adapt to high salinity, intense pressure, limited oxygen, and shifting temperatures—factors that drive the evolution of uniquely potent biochemical systems. As a result, the bioactive compounds they produce possess exceptional functionality, making them particularly attractive for advanced applications in health and industry [5].In addition, recent progress in extraction technologies and analytical methodologies has significantly enhanced the discovery and detailed profiling of bioactive peptides and proteins derived from marine sources. These functional peptides are typically generated through the breakdown of proteins, using either traditional or advanced hydrolysis techniques. Methods range from acid and alkaline treatments to more sophisticated approaches such as solvent extraction, enzymatic hydrolysis, microbial fermentation, ultrasound-assisted processing, pulsed electric fields, pressurized liquid extraction, and subcritical water techniques. These innovations have greatly expanded the efficiency and precision of marine bioactive compound production [6]. Among the various techniques available, enzymatic hydrolysis is widely favored in both the food and pharmaceutical sectors due to its safety, specificity, and eco-friendly nature. In contrast to chemical and solvent-*

based methods, which may leave behind harmful residues or toxic by-products, enzymatic processes offer a cleaner and more controlled approach to producing bioactive peptides [7]. Marine-derived peptides, produced through protein hydrolysis, often contain unwanted impurities that can compromise their biological activity and therapeutic potential. To fully realize the benefits of these compounds, advanced separation and purification processes are essential. Purification plays a crucial role, particularly for peptides intended for consumer or clinical use, as it ensures high quality, efficacy, and safety. Cutting-edge techniques such as ultrafiltration, gel filtration chromatography, ion-exchange chromatography, and reversed-phase high-performance liquid chromatography (RP-HPLC) are widely utilized to isolate and refine these peptides. For example, immunomodulatory peptides extracted from Stolephorus chinensis were effectively purified using a combination of process optimization, ultrafiltration, ion-exchange chromatography, and RP-HPLC. These sophisticated methods not only enhance peptide purity but also boost their bioactivity, supporting their integration into pharmaceutical formulations, nutraceutical products, and functional food applications[8,9].

Marine-Derived Peptides and Proteins: Sources and Characteristics

*Although the marine ecosystem is incredibly diverse, research into its bioactive proteins and peptides has so far been limited to a relatively small group of organisms. These include certain invertebrates (such as mollusks, sponges, echinoderms, and cnidarians), various species of fish, different types of algae (including red, green, and brown varieties, as well as spirulina), crustaceans (like crabs, lobsters, crayfish, prawns, and shrimp), and marine microorganisms (Figure 1). Among these, fish are particularly notable as a rich and reliable source of high-quality proteins, containing essential amino acids vital for human metabolism. Fish proteins encompass a wide range of structural and functional types—including collagen, myoglobin, and actin—which are primarily located in tissues such as skin, bones, scales, and internal organs. Marine proteins and peptides derived from fish are obtained not only from edible fillets but also from processing by-products and waste materials [10,11]. Beyond fish, collagen has also been successfully isolated from a wide range of other marine organisms, including sea cucumbers, mollusks, sponges, crustaceans, seaweeds, and jellyfish. These alternative sources offer promising potential for sustainable and diverse applications in health, cosmetics, and biomaterials [12]. Shellfish—including shrimp, crabs, crayfish, and lobsters—are recognized as rich sources of high-quality proteins and biologically active peptides. These marine organisms produce a diverse array of functional peptides with various health-promoting properties. For example, mussels and clams contain opioid peptides, while blue mussels, oysters, scallops, and shrimp are known to generate antimicrobial peptides. Shrimp and crabs are sources of angiotensin-converting enzyme (ACE) inhibitory peptides, which are associated with blood pressure regulation. Immunomodulatory peptides have also been identified in fish, mussels, and scallops, and antioxidant peptides are notably present in scallops. Specific opioid peptides found in shellfish include dynorphins in blue mussels (*Mytilus edulis*), methionine-enkephalins in oysters and clams, and β-endorphins in shrimp, all of which contribute to their* *biofunctional value [13]. ACE inhibitory peptides derived from shellfish have demonstrated significant potential in managing hypertension by blocking the activity of the angiotensin-converting enzyme (ACE), thereby contributing to blood pressure reduction. Prominent examples include valyl-tyrosine (VY) extracted from shrimp (*Litopenaeus vannamei*) protein hydrolysates, isoleucyl-prolyl-proline identified in oysters (*Crassostrea gigas*), isoleucyl-leucine-proline from crab (*Portunus trituberculatus*) protein hydrolysates, and leucine-aspartic acid obtained from clams (*Meretrix meretrix*). In addition to their antihypertensive properties, shellfish also produce immunomodulatory peptides and proteins. Notable among these are hemocyanins found in mollusks and crustin in crustaceans, both of which play important roles in enhancing immune response by stimulating phagocytosis and promoting cytokine release [14]. Research involving spontaneously hypertensive rats has shown that gelatin hydrolysates obtained from marine organisms such as sea cucumbers, skate skin, jellyfish, and squid skin possess blood pressure-lowering effects—an important strategy in reducing the risk of cardiovascular diseases. Gelatin-derived peptides from fish have exhibited a wide range of bioactive properties, including antioxidant, anti-anemic, immunomodulatory, calcium-binding, mineral-chelating, antimicrobial, and enzyme-inhibitory activities, particularly against ACE and DPP-IV. These functional benefits have been documented in hydrolysates produced from gelatin extracted from a variety of fish species, including Atlantic salmon, cod, herring, hoki, Pacific whiting, pollock, snapper, and sole [15]. A wide range of antimicrobial peptides (AMPs) have been successfully identified and extracted from diverse marine species, including Atlantic cod (*Gadus morhua*), mud crab (*Scylla paramamosain*), oyster (*Crassostrea gigas*), yellow catfish (*Pelteobagrus fulvidraco*), marine sponge (*Trichoderma* sp.), sea snail (*Cenchritis muricatus*), marine bivalve (*Mytilus coruscus*), and octopus (*Octopus vulgaris*). These peptides are valued for their natural origin, strong antimicrobial potency, cost-efficiency, and excellent safety profile. Owing to their broad-spectrum activity and biocompatibility, marine-derived AMPs are emerging as highly attractive agents for use in pharmaceuticals, food preservation, and biomedical applications [16]. In addition, numerous antimicrobial peptides (AMPs) have been discovered in a variety of mollusk species, particularly within bivalves. Mussels, for instance, have been found to produce cysteine-rich peptides, while oysters and gastropods are known to contain defensins and proline-rich peptides with potent antimicrobial functions. Beyond peptides, mollusks—especially gastropods—also yield antimicrobial proteins such as egg case proteins (Sep-ECPs) and hemocyanin, further highlighting their potential as natural sources of bioactive compounds with broad-spectrum antimicrobial activity [17]. Recent research has intensified efforts to isolate bioactive peptides with anticancer potential from a wide array of marine organisms. These include sponges, tunicates, ascidians, mollusks, fish (along with their processing by-products), and various other aquatic species, all of which have emerged as promising sources of novel therapeutic agents [18].*

Production of Peptides from Marine Organisms

Marine organisms, including fish, algae, crustaceans, microbes, and invertebrates, are considered prominent sources of bioactive proteins and peptides, these marine-derived compounds offer several advantages, such as compatibility with human metabolism, *absence of religious restrictions, and freedom from pathogens commonly found in terrestrial animals, these biomolecules exhibit a wide range of biological activities, including antioxidant, antihypertensive, antidiabetic, anticoagulant, antimicrobial, anti-inflammatory, antithrombotic, anticancer, and immunomodulatory effects, the bioactive potential of these marine proteins and peptides is illustrated in Figure 1*

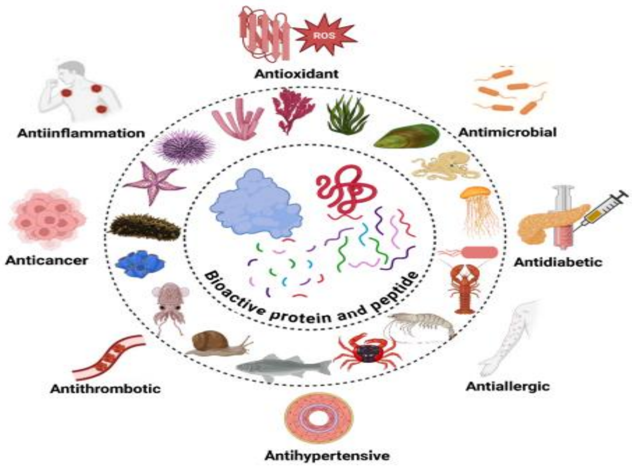

Figure 1. *Bioactive potential of marine-derived proteins and peptides*

Antioxidant Activity

Oxidation is an essential metabolic process in the bodies of vertebrates and humans. However, it also generates reactive oxygen species (ROS) and free radicals, which can disrupt cellular homeostasis and lead to oxidative stress. This stress can be highly damaging to cells and, over time, may contribute to the development of various diseases, including cardiovascular disease, stroke, arteriosclerosis, diabetes, and cancer [19]. In addition, the food industry and consumers alike are seriously concerned about the oxidation of lipids triggered by reactive oxygen species (ROS), as it leads to spoilage, unpleasant flavors, and the formation of harmful substances. Lipid peroxidation not only affects the quality and safety of food products but also poses significant risks to human health [20]. Proteins and peptides derived from marine sources demonstrate strong antioxidant effects, primarily due to their ability to neutralize free radicals, bind metal ions, and support the body's natural antioxidant systems. Their antioxidant capacity has been extensively studied using a range of in vitro methods, including tests that measure their ability to scavenge radicals like DPPH, hydroxyl, superoxide anion, and ABTS, as well as their metal-chelating activity. The core mechanism behind these effects involves the donation of hydrogen atoms or electrons from the antioxidant molecules, which helps stabilize and deactivate harmful free radicals [21]. Hydrophobic amino acids, including phenylalanine (Phe, F), tryptophan (Trp, W), tyrosine (Tyr, Y), alanine (Ala, A), valine (Val, V), and leucine (Leu, L), contribute to the enhanced free radical scavenging activity of peptides by facilitating their interaction with and penetration into the lipid bilayers of target organ membranes through hydrophobic interactions. Meanwhile, essential and acidic amino acids, such as lysine

(Lys, K), aspartic acid (Asp, D), and glutamic acid (Glu, E), act as efficient metal ion chelators. Mar. Drugs 2025, 23, 157 10 of 49 Additionally, aromatic amino acids like phenylalanine (Phe, F), tryptophan (Trp, W), and tyrosine (Tyr, Y) scavenge free radicals via direct electron transfer mechanism[22]. Multiple studies have indicated that proteins and peptides derived from marine organisms exhibit strong antioxidant capabilities. According to findings by Safari and Yaghoubzadeh [23]. Research has shown that antioxidant peptides extracted from the sea cucumber Holothuria leucospilota have promising potential as natural antioxidants for use in both pharmaceutical and food applications. In another study, protein hydrolysates were produced from different parts of the North Atlantic sea cucumber (Cucumaria frondosa)—including the body wall, internal organs, and processing by-products—using various endopeptidase enzymes: Alcalase (A), Corolase (C), and Flavourzyme (F). Among the enzyme combinations tested, the blend of Flavourzyme and Corolase (F + C) yielded hydrolysates with the strongest ability to neutralize DPPH and ABTS free radicals. Additionally, the highest metal ion chelation activity was observed in hydrolysates treated with enzyme pairs (C + F) and (A + F) [24]. Moreover, protein hydrolysates derived from Stichopus japonicus have demonstrated strong protective effects against oxidative stress. In experiments using Vero cells exposed to hydrogen peroxide (H_2O_2), treatment with the hydrolysate led to a significant reduction in reactive oxygen species (ROS), improved cell survival, and a decrease in apoptosis-related damage. Likewise, when tested on zebrafish embryos subjected to H_2O_2-induced oxidative stress, the hydrolysate effectively reduced both cell death and levels of ROS and lipid peroxidation [25]. Protein hydrolysates obtained from discarded raw shrimp shells and shrimp shell proteins, prepared using the enzymes trypsin, chymotrypsin, and pepsin, demonstrated strong antioxidant capabilities. These hydrolysates effectively scavenged free radicals such as ABTS, DPPH, and hydroxyl radicals, while also showing significant reducing power and the ability to chelate ferrous ions. Moreover, their antioxidant effects extended to biological systems, where they inhibited the oxidation of low-density lipoprotein (LDL) and provided protection to DNA molecules [26]. Antioxidant peptides extracted from Antarctic krill (Euphausia superba) demonstrated significant reducing power, effectively protected plasmid DNA from damage caused by hydrogen peroxide (H_2O_2), and inhibited lipid peroxidation [27]. Three new peptides were isolated from the myofibrillar protein hydrolysates of patin (Pangasius sutchi). After purification and testing, these peptides showed remarkable antioxidant properties, indicating their potential use as functional ingredients with antioxidative effects. Similarly,[28]. identified novel antioxidant peptides in muscle protein hydrolysates of mackerel (Scomber japonicus). Among these, the peptide ALSTWTLQLGSTSFSASPM demonstrated the highest free radical scavenging activity against DPPH radicals, while the peptide LGTLLFIAIPI exhibited the strongest superoxide dismutase (SOD)-like activity. These findings highlight the promising potential of fish protein hydrolysates as valuable sources of bioactive peptides with functional benefits [29]. also contributed to this field by...

Conclusion

Marine-derived proteins and peptides exhibit significant antioxidant activities through multiple mechanisms, including free radical scavenging, metal ion chelation, and enhancement of the body's natural antioxidant defenses. Their rich composition of hydrophobic, essential, acidic, and aromatic amino acids facilitates effective interaction *with oxidative agents and cellular membranes, contributing to their potent bioactivity. Various studies have demonstrated that protein hydrolysates from diverse marine sources such as sea cucumbers, shrimp shells, krill, and fish possess strong antioxidant capabilities in both in vitro and biological systems. These findings underscore the promising potential of marine protein hydrolysates as natural antioxidants for application in food preservation, pharmaceuticals, and functional foods, offering a natural strategy to combat oxidative stress and related diseases.*

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