

Review:

Biological activity and zoochemical analysis of anchovies (*Stolephorus* spp.)

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Abstract. *Widowati D, Saputra EF, Hartono DAN, Wicaksono FR, Safira RN, Yap CK, Naim DMd, Setyawan AD. 2025. Review: Biological activity and zoochemical analysis of anchovies (Stolephorus spp.). Asian J Nat Prod Biochem 23: 70-92.* Anchovies (*Stolephorus* spp.) are small pelagic fish with broad distribution across tropical and subtropical marine ecosystems. They are ecologically significant as mid-trophic species and hold substantial value due to their rich nutritional and zoochemical composition. This review synthesizes current knowledge on anchovy-derived compounds, including high-quality proteins, essential amino acids, longchain omega-3 fatty acids (EPA and DHA), and bioavailable minerals such as calcium and selenium. These bioactive components exhibit diverse biological activities antioxidant, anti-inflammatory, antihypertensive, antidiabetic, and anticancer mediated through defined molecular mechanisms. Structure-function relationships and molecular interactions are examined in relation to their healthpromoting effects. The review also discusses technological innovations in extraction, processing, and formulation for functional foods and nutraceuticals. Environmental risks, including heavy metal bioaccumulation, microplastic exposure, and overfishing, are critically addressed along with strategies for sustainable harvesting and ecosystem-based management. Anchovies represent a promising marine bioresource for circular bioeconomy models, yet significant gaps remain in clinical validation, advanced compound characterization, and industrial-scale application. The multifunctional properties of anchovies support their integration into health-oriented and ecologically sustainable innovations, offering new opportunities in food, pharmaceutical, and environmental sectors.

Keywords: bioactive peptides, functional food, marine nutraceuticals, omega-3 fatty acids, *Stolephorus*, sustainable fisheries

INTRODUCTION

Anchovies (*Stolephorus* sp.) are small fish in the family Engraulidae. (Sosiawati 2019), widely distributed across tropical and subtropical marine ecosystems, including the Indo-Pacific region (Susanto et al. 2017; Sari et al. 2019). Characterized by their streamlined bodies, silvery coloration, and filter-feeding habits, anchovies play a vital role in marine food webs as both predator and prey (Checkley and Barth 2009; Pikitich et al. 2012). Their ecological importance is supported by their ability to regulate plankton populations while simultaneously serving as a food source for larger fish, marine mammals, and seabirds (Chaves and Umbria 2003; Tanaka et al. 2008). As such, anchovies contribute significantly to the sustainability and resilience of coastal marine ecosystems.

Beyond their ecological role, anchovies have long been recognized as a valuable component of traditional diets in many coastal communities. Rich in high-quality protein, essential fatty acids such as EPA and DHA, vitamins, and trace minerals like calcium, iron, and selenium, anchovies

represent a cost-effective source of animal-derived nutrition (Barros et al. 2014; Kari et al. 2022). These nutritional properties make anchovies not only important for addressing malnutrition and micronutrient deficiencies but also for supporting cardiovascular health, bone development, and immune function (Galasso et al. 2020a; Hu et al. 2020). The processing of anchovies into salted fish, fermented sauces, and dried snacks further enhances their accessibility and shelf life, especially in tropical developing nations.

Recent advances in marine nutraceutical research have highlighted the therapeutic potential of anchovies. Bioactive compounds derived from *Stolephorus* spp., including peptides, polyunsaturated fatty acids, and antioxidant enzymes, have shown promising pharmacological properties such as antihypertensive, antithrombotic, anti-inflammatory, antimicrobial, and antidiabetic effects (Mutalipassi et al. 2021; Shaik and Sarbon 2022b; Azfaralariff et al. 2023); these properties position anchovies as potential candidates for functional food development and novel marine-based health products.

In particular, fish-derived biopeptides have gained attention for their role in inhibiting free radicals and modulating key molecular pathways associated with chronic diseases (Hasari et al. 2021; Engwa et al. 2022).

While numerous studies have investigated the nutritional value of marine fishes in general, focused research on the zoochemical composition—defined as the spectrum of bioactive substances derived from animal tissues and biological activity of anchovies remains limited and fragmented. Zoochemical constituents in anchovies include not only macronutrients but also functional elements such as albumin, amino acids, marine enzymes, and trace hormones that may exert physiological effects in humans (Chen et al. 2022a; Nurmawati et al. 2022). A comprehensive synthesis of these components is needed to understand better their health implications, mechanisms of action, and potential applications in functional food and pharmaceutical development.

Moreover, global changes such as climate-driven shifts in marine productivity, increasing pollution from microplastics and heavy metals, and overexploitation of anchovy stocks pose challenges to the sustainability and safety of anchovy-derived products (Lima et al. 2022; Komala et al. 2024). However, understanding the interaction between anchovy biology, their zoochemical profiles, and environmental conditions is crucial for designing sustainable harvesting and processing strategies. Furthermore, evaluating the safety aspects, such as the potential accumulation of mercury or persistent organic pollutants, is essential for ensuring consumer health and regulatory compliance.

This review aims to bridge existing knowledge gaps by synthesizing current scientific literature on the biological activity and zoochemical analysis of anchovies (*Stolephorus* spp.). Specifically, it (i) explores the nutritional and bioactive profiles of anchovy tissues, (ii) summarizes known biological effects and underlying mechanisms of action, and (iii) evaluates environmental and health implications relevant to anchovy utilization. By integrating ecological, biochemical, and pharmacological perspectives, this review seeks to provide a holistic understanding of the role of anchovies in marine systems and human health.

This article also highlights the potential of anchovies as sustainable bioresources for future nutraceutical innovation, especially in the context of food security, marine conservation, and public health. The review concludes with directions for future research, including the need for standardized analytical methods, bioactivity-guided compound isolation, and toxicological assessments. It is hoped that this synthesis will serve as a valuable reference for marine biologists, food technologists, public health researchers, and policymakers working at the intersection of biodiversity conservation and human nutrition.

ECOLOGICAL AND TAXONOMIC OVERVIEW

Habitat and environmental adaptations

Anchovies (*Stolephorus* spp.) inhabit a range of tropical and subtropical marine environments, particularly warm and nutrient-rich coastal waters. These habitats are typically characterized by temperatures between 15-30°C and salinity levels close to 35 ppt, which are optimal for plankton production their primary food source (Pebruwanti and Fitriani 2018; Sari et al. 2019). Anchovies tend to dwell in the epipelagic zone up to 50 meters deep, where light penetration supports primary productivity and food availability.

They are commonly found in estuaries, coral reef fringes, bays, and river mouths, often aggregating in large schools to maximize feeding efficiency and predator avoidance (Prihantoko and Boesono 2019). Upwelling zones and coastal current systems are especially favorable due to the continuous supply of plankton and suspended nutrients. These physical conditions directly influence the distribution, growth, and reproductive success of anchovy populations.

A variety of physiological and morphological traits enables adaptation to these dynamic environments. Anchovies are capable of osmoregulation across varying salinity gradients, allowing them to survive in brackish and estuarine conditions (Falco et al. 2020). Their streamlined, hydrodynamic bodies facilitate fast and energy-efficient swimming. At the same time, their upward-facing mouths and elongated lower jaws enable rapid filtration of planktonic prey from the water column (Yusfiandayani et al. 2019). These morphological features are further complemented by reproductive strategies such as high fecundity and rapid larval development, allowing anchovy populations to recover quickly from environmental disturbances (Purnomo et al. 2017).

In addition to physical adaptations, anchovies exhibit behavioral and social traits that enhance survival. They often form synchronized schooling formations that provide hydrodynamic benefits and reduce individual predation risk through the confusion effect (Yoga 2019). Such schooling also facilitates efficient foraging in patchy plankton environments. Moreover, their migration patterns are influenced by seasonal temperature shifts and plankton blooms, ensuring continued access to food and suitable spawning grounds.

The ecological flexibility of anchovies is further demonstrated by their presence in various microhabitats and fluctuating coastal systems. Their abundance in highproductivity areas makes them a key trophic link in marine food webs, transferring energy from primary producers (phytoplankton and zooplankton) to higher-level predators, including commercial fish species, marine mammals, and seabirds (Pikitich et al. 2012). Consequently, anchovies are considered important ecological indicators of coastal ecosystem health. To support this description, spatial data on the environmental preferences of anchovies can be illustrated in Figure 1 and Table 1.



Figure 1. Typical habitats of *Stolephorus* spp. in tropical coastal ecosystems

These environmental parameters define anchovy habitat selection and underpin their seasonal migration, spawning success, and abundance across Indo-Pacific waters. Understanding these ecological preferences is crucial for effective management of anchovy fisheries, particularly in the face of climate-driven oceanographic changes and coastal habitat degradation (Costalago et al. 2011).

Species distribution and invasiveness

Anchovies (*Stolephorus* spp.) are widely distributed throughout tropical and subtropical waters, especially in the Indo-Pacific region, extending westward from the Indian Ocean to the western Pacific Ocean. Their primary habitats include coastal zones of Southeast Asia, northern Australia, the Bay of Bengal, and surrounding archipelagic waters (Cavraro et al. 2022; FishBase 2024). Their physiological tolerance to variable salinities and temperatures, as well as

their plankton-based diet, which is abundant in many coastal ecosystems, facilitates this broad distribution.

In Indonesia, anchovies are commonly found in the Java Sea, Sulawesi Sea, and waters surrounding the Lesser Sunda Islands (Andriyono 2018; Yonvitner et al. 2020). Their occurrence in shallow marine areas, estuaries, and reef-associated systems reflects their ecological plasticity. Seasonal spawning migrations enable them to exploit nutrient-rich areas, particularly during monsoonal-driven upwelling events. During such periods, anchovy populations can increase dramatically, forming large shoals that are both ecologically and commercially significant (Masyhudi 2024).

The dispersal potential of *Stolephorus* spp. also raises concern regarding their invasive tendencies in non-native environments. While not traditionally considered invasive, their high fecundity, broad salinity tolerance, and rapid growth rates make them capable of establishing populations in new areas if accidentally introduced (Lima et al. 2022). In some regions, such as the Caribbean and Gulf of Mexico, closely related anchovy species have been observed exhibiting non-native spread, potentially competing with local pelagic fish for food resources.

A major ecological concern is their ability to outcompete native plankton-feeding species due to their efficient foraging and reproductive capacities. In ecosystems where trophic balance is fragile, such introductions could result in shifts in plankton community structure, reduction in native species abundance, and alterations in nutrient cycling (Smith and Jones 2023). Moreover, anchovies may serve as vectors for pathogens or parasites if transported via ballast water or aquaculture effluents.

Despite their broad range, most *Stolephorus* species remain underrepresented in global conservation databases. According to the IUCN, many anchovy species are listed as Least Concern (LC) or Data Deficient (DD) due to limited population data and taxonomic resolution (Fricke et al. 2019; GBIF 2024). This underscores the need for increased taxonomic and ecological research, especially in regions where their populations are heavily exploited or where potential invasive pathways exist. To visualize their distribution and assess invasiveness risk, see Figure 2 and Table 2.

Table 1. Summary of habitat types and environmental adaptations of *Stolephorus* spp.

Habitat type	Environmental characteristics	Anchovy adaptations
Coastal pelagic zones	Warm, shallow seas (20-30 °C), high light penetration	Streamlined body for fast swimming; filter-feeding on plankton
Estuaries and river mouths	Variable salinity (brackish), high nutrient input	Osmoregulatory capability; tolerance to salinity gradients
Coral reef fringes	Clear waters, structural complexity	Schooling behavior for predator avoidance; high maneuverability
Upwelling zones	Cool, nutrient-rich currents, high plankton concentration	High reproductive capacity; rapid growth linked to food abundance
Bays and lagoons	Sheltered, productive inshore environments	Opportunistic feeding; aggregation in large shoals for foraging efficiency
Epipelagic layer (0-50 m)	Oxygen-rich, photic zone with abundant zooplankton	Upward-facing mouth and elongated jaw for plankton capture

Sources: Prihantoko and Boesono (2019); Sari et al. (2019); Yusufiandayani et al. (2019); Falco et al. (2020)

Table 2. Major *Stolephorus* species and their distribution across ocean regions

Species	Native range	Reported non-native occurrence	Invasiveness risk	IUCN Status
<i>S. indicus</i>	Indian Ocean, Southeast Asia	Red Sea (occasional)	Low-Moderate	Least Concern
<i>S. commersonii</i>	Western Pacific, Indonesia, Australia	Caribbean Sea (introduced records)	Moderate	Data Deficient
<i>S. dubiosus</i>	Indo-Malay waters	No records	Low	Not Evaluated
<i>S. waitei</i>	Papua New Guinea, Northern Australia	No records	Low	Least Concern
<i>S. insularis</i>	Western Indian Ocean	Not documented	Unknown	Not Evaluated

Sources: Fricke et al. (2019); Lima et al. (2022); FishBase (2024); GBIF (2024);



Figure 2. Global distribution map of anchovy (*Stolephorus* spp.) species across Indo-Pacific regions

Understanding both the natural distribution and potential for ecological spread is critical for managing anchovy populations (Abdusysyahid 2024). In regions where fisheries are expanding or shipping intensity is high, monitoring programs are essential to detect possible introductions and assess ecological impacts. Moreover, accurate species identification is vital, as cryptic diversity within *Stolephorus* may obscure invasion dynamics and conservation priorities.

Taxonomic classification and morphology

Anchovies of the genus *Stolephorus* are members of the family Engraulidae, order Clupeiformes, class Actinopterygii, phylum Chordata, and kingdom Animalia (GBIF 2024). This taxonomic grouping places them among ray-finned fishes with a close evolutionary relationship to herrings and sardines. The genus *Stolephorus*, first described by Lacepède in 1803, comprises numerous species that are morphologically similar but differ in subtle anatomical features, meristic counts, and geographical distributions (Hata and Motomura 2022). Most species are small, typically ranging from 5 to 15 cm in length, and are easily recognized by their slender bodies, prominent silver lateral bands, and terminal or slightly upturned mouths.

Morphologically, *Stolephorus* spp. exhibit adaptations that support their pelagic lifestyle and plankton-feeding behavior. Their bodies are laterally compressed and covered in thin cycloid scales that reflect light, aiding in

camouflage in the epipelagic zone. The head is moderately large with a terminal to slightly superior mouth, suited for filter feeding on plankton. Notably, the lower jaw tends to protrude beyond the upper jaw, which enhances surface skimming efficiency during foraging. The dorsal fin is short and positioned near the middle of the body, while the anal fin is relatively longer, contributing to maneuverability. The caudal fin is deeply forked, enabling fast swimming and sudden directional changes an important trait for evading predators and maintaining schooling formation (Yusfiandayani et al. 2019).

Internally, anchovies possess a well-developed gill raker system used to trap fine planktonic particles. The vertebral column is flexible, providing agility in water, and the swim bladder is reduced or absent in many species, reflecting their active, near-surface lifestyle. Sexual dimorphism is minimal, although during the spawning season, females may exhibit slightly enlarged abdomens. Eggs are pelagic and spherical, typically hatching within 24-48 hours after external fertilization in warm, plankton-rich waters (Gopinath and Abdussamad 2024).

These morphological and anatomical traits not only support the ecological niche of anchovies but also serve as taxonomic markers in species identification. Due to the subtle differences between species, accurate identification often requires detailed examination of gill raker counts, scale patterns, and the shape of the supramaxilla bones. Advances in molecular genetics and morphometric analysis are increasingly used to resolve taxonomic ambiguities within the genus. Such precision is particularly important in ecological studies, fisheries management, and when evaluating the bioactivity or biochemical properties of specific *Stolephorus* species (Table 3; Figure 3).

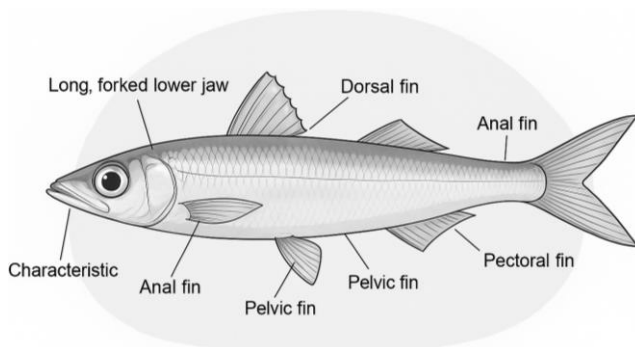
Ecological roles and conservation status

Anchovies (*Stolephorus* spp.) occupy a pivotal ecological niche in tropical and subtropical marine ecosystems as midtrophic-level organisms. Functioning both as consumers of plankton and as prey for higher-level predators, they serve as a crucial energy conduit linking primary producers with piscivorous fish, marine mammals, and seabirds (Tanaka et al. 2008; Pikitich et al. 2012). By intensively consuming phytoplankton and zooplankton, anchovies help regulate planktonic community dynamics and influence nutrient cycling through the excretion of nitrogen and phosphorus compounds (Checkley and Barth 2009). Their dense schooling behavior also plays a role in predator-prey interactions by shaping the foraging strategies of larger pelagic species such as tuna, mackerel, and dolphins (Figure 4).

Table 3. Morphological characteristics used in taxonomic identification of *Stolephorus* spp.

Morphological trait	Diagnostic relevance	Variation among species
Body shape and size	General body form (slender, compressed), total length	Typically 5-15 cm; some species reach >18 cm
Lateral silver band	Presence, width, and reflectivity of lateral stripe	Prominent in most species; varies in length and intensity
Mouth orientation	Terminal or slightly superior mouth; jaw projection	Lower jaw often protrudes beyond upper jaw
Gill raker counts	Number and density of gill rakers on the first branchial arch	Used to differentiate plankton-feeding adaptations
Scale type and arrangement	Type (cycloid), scale size, and pattern along the lateral line	Species-specific scale patterns and size
Supramaxilla bone shape	Subtle anatomical differences in the upper jaw bone structure	Key for species-level separation
Fin position and shape	Dorsal, anal, and caudal fin characteristics	Dorsal fin mid-body; anal fin relatively long
Swim bladder presence	Reduced or absent in many species; aids in pelagic adaptation	Absence supports near-surface swimming behavior
Egg morphology (when known)	Shape and buoyancy of pelagic eggs (for reproductive identification)	Spherical, transparent, planktonic

Sources: Yusfiandayani et al. (2019); Hata and Motomura (2022); Gopinath and Abdussamad (2024)

**Figure 3.** General morphology and anatomical features of anchovies (*Stolephorus* spp.), highlighting diagnostic structures

In many coastal areas, anchovies serve as ecological indicators of ocean productivity and environmental stability. Their population fluctuations are closely linked to oceanographic parameters such as sea surface temperature, upwelling intensity, and chlorophyll-a concentration, making them highly sensitive to climate variability and anthropogenic disturbance (Chaves and Umbria 2003; Lima et al. 2022). For example, El Niño events can drastically reduce anchovy biomass by disrupting upwelling systems and decreasing plankton abundance, as observed in the Pacific Ocean and parts of Southeast Asia. Consequently, long-term monitoring of anchovy populations can provide early warning signs of ecosystem stress or regime shifts in marine environments.

From a conservation perspective, most species within *Stolephorus* remain poorly evaluated, with many listed as Data Deficient (DD) or Least Concern (LC) under the IUCN Red List criteria (Fricke et al. 2019; GBIF 2024). This reflects both their ecological ubiquity and the lack of comprehensive population data, especially for lesser-known or cryptic species. Despite the absence of immediate global extinction risk, anchovy populations face localized threats such as overfishing, habitat degradation (e.g., mangrove and coral reef loss), and marine pollution including microplastics and heavy metals—that can bioaccumulate through trophic transfer (Hasari et al. 2021; Komala et al. 2024).

In response to these pressures, some countries have implemented seasonal fishing quotas, gear restrictions, and Marine Protected Areas (MPAs) to safeguard anchovy stocks and maintain ecosystem integrity. However, the effectiveness of such measures often depends on enforcement capacity and scientific input, including accurate stock assessments and life history data. Additionally, climate change-induced alterations in ocean currents and productivity may require adaptive management strategies that integrate environmental forecasting with fisheries regulation. The summary of their ecological roles and conservation status is presented in the following visual aids (Table 4).

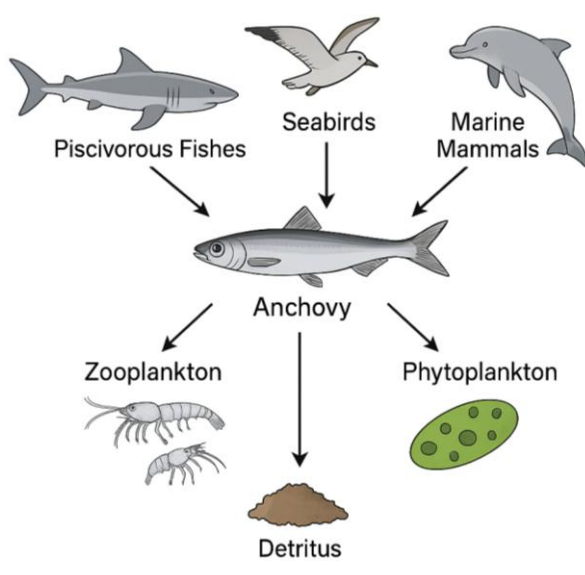
**Figure 4.** Schematic diagram of anchovy position in the marine trophic food web

Table 4. Reported conservation status of *Stolephorus* species by region

Species	Region	IUCN red list status	Known threats	Data availability
<i>S. indicus</i>	Indian Ocean, Southeast Asia	Least Concern	Overfishing, coastal habitat degradation	Moderate
<i>S. commersonnii</i>	Indo-Pacific, Australia	Data Deficient	Bycatch, marine pollution, and warming temperatures	Limited
<i>S. waitei</i>	Coral reef zones, Northern Australia	Least Concern	Coral degradation, artisanal fisheries	Sparse
<i>S. insularis</i>	Western Indian Ocean	Not Evaluated	Unknown	Very limited
<i>S. dubiosus</i>	Indonesia, Indo-Malay Archipelago	Not Evaluated	Habitat disturbance, taxonomic ambiguity	Very limited

Sources: Fricke et al. (2019); Lima et al. (2022); IUCN (2023); GBIF (2024)

Given their ecological importance and increasing utilization for human consumption, anchovies warrant more rigorous conservation attention. Comprehensive species-level monitoring, coupled with regional collaboration, is essential to prevent future overexploitation and ensure that anchovy populations continue to fulfill their ecological and economic roles in tropical marine systems (Appeltans et al. 2020).

ZOOCHEMICAL ANALYSIS OF ANCHOVIES

Nutritional profile (proteins, fats, minerals, vitamins)

Anchovies (*Stolephorus* spp.) are a nutrient-dense marine food source that offers a well-balanced profile of macronutrients and micronutrients essential for human health. Their small size belies their nutritional richness, as anchovy flesh is particularly high in protein, containing approximately 15-25 g of protein per 100 g of edible portion, depending on species, season, and processing method (Kari et al. 2022). This protein is of high biological value, with a complete amino acid profile that supports tissue maintenance and growth.

The fat content of anchovies varies between 2-10%. It is primarily composed of Polyunsaturated Fatty Acids (PUFAs), especially omega-3 fatty acids such as Eicosapentaenoic Acid (EPA) and Docosahexaenoic Acid (DHA). These bioactive lipids play crucial roles in reducing cardiovascular risk, regulating inflammation, and supporting neurodevelopment (Barros et al. 2014; Hu et al. 2020). Additionally, anchovies contain cholesterol-lowering

sterols and phospholipids that enhance lipid metabolism and membrane function.

Micronutrient-wise, anchovies are an excellent source of dietary calcium, particularly when consumed with bones, as in dried or fried forms. The calcium content may range from 300 to 1000 mg per 100 g, which significantly contributes to bone health and prevention of osteoporosis, particularly in populations with limited dairy intake. This information is crucial for making informed dietary choices. Iron, zinc, magnesium, selenium, and phosphorus are also abundant, supporting immune response, oxygen transport, and antioxidant defense systems (Chen et al. 2022a). Selenium, in particular, acts as a cofactor for glutathione peroxidase, an important antioxidant enzyme, while zinc is essential for enzymatic activity and DNA repair.

In terms of vitamins, anchovies provide moderate to high levels of fat-soluble vitamins, especially vitamin D and vitamin A. These vitamins play a crucial role in calcium absorption, bone mineralization, and visual function. In addition, anchovies also provide appreciable amounts of water-soluble B vitamins, such as niacin (B3), riboflavin (B2), and vitamin B12, which are also present in appreciable amounts and contribute to energy metabolism and neurological health (Galasso et al. 2020a).

To illustrate the comprehensive nutrient composition of anchovies and their dietary value relative to other small pelagic fish (Table 5). Anchovies' nutritional profile affirms their potential as a low-cost, high-impact food for public health and food security strategies, particularly in coastal and low-resource settings. Their integration into local diets, either fresh or in processed forms, can help combat hidden hunger and contribute to sustainable nutrition solutions.

Table 5. Nutritional composition of anchovies (per 100 g fresh weight)

Nutrient	Anchovies (<i>Stolephorus</i> spp.)	Sardines (<i>Sardinella</i> spp.)	Mackerel (<i>Rastrelliger</i> spp.)	Notes
Protein (g)	18.2-25.0	20.0-24.0	19.5-22.0	High-quality, complete amino acid profile
Fat (g)	4.5-10.0	10.0-15.0	8.0-12.0	Mostly polyunsaturated (omega-3)
Omega-3 (EPA + DHA, g)	1.2-2.5	1.5-3.0	1.8-2.8	Cardioprotective and anti-inflammatory
Calcium (mg)	450-1000	300-500	150-250	Highest when consumed with bones
Iron (mg)	2.0-4.5	1.8-3.5	1.2-2.5	Important for hemoglobin synthesis
Vitamin B12 (µg)	8.5-12.0	9.0-13.0	8.0-11.0	Supports nerve function and metabolism
Vitamin D (IU)	250-600	300-550	200-500	Key for calcium absorption and immunity
Selenium (µg)	30-60	25-55	20-45	Antioxidant cofactor (GPx)

Sources: Barros et al. (2014); Galasso et al. (2020); Kari et al. (2022)

Amino acid composition

The high protein content of anchovies (*Stolephorus* spp.) is complemented by a well-balanced profile of amino acids, both essential and non-essential, which contributes to their high nutritional and functional value. These amino acids not only serve as the building blocks for protein biosynthesis but also participate in physiological processes such as neurotransmission, immune response, antioxidant defense, and metabolic regulation (Andriani and Muhdar 2021; Mutalipassi et al. 2021). The quality of anchovy protein is particularly high due to its digestibility and the presence of all essential amino acids in proportions suitable for human dietary needs.

Among the essential amino acids, anchovies are notably rich in lysine, leucine, isoleucine, methionine, and threonine (Kari et al. 2022). Lysine plays a key role in calcium absorption and collagen formation, while leucine and isoleucine are critical for muscle repair and metabolic signaling (Aggarwal and Bains 2022). Methionine serves as a precursor to sulfur-containing compounds and antioxidants like glutathione (Castro et al. 2023). These amino acids are indispensable in human diets, especially in populations that rely on cereal-based staples with limited amino acid profiles.

In addition, non-essential amino acids such as glutamic acid, alanine, serine, and arginine are abundant in anchovy tissue. Glutamic acid functions as an excitatory neurotransmitter and is a precursor for γ -Aminobutyric Acid (GABA), whereas alanine is involved in energy metabolism and glucose regulation. Arginine supports immune function and nitric oxide synthesis, contributing to vascular health. The interaction between these amino acids contributes to the functional versatility of anchovies as both a food and a potential therapeutic agent (Leke and Schousboe 2016).

The amino acid profile may vary based on environmental factors, including water temperature, food availability, and physiological maturity, as well as postharvest processing methods such as fermentation or hydrolysis. For example, fermented anchovy products have been reported to exhibit enhanced concentrations of free amino acids and bioactive peptides due to proteolysis by endogenous or microbial enzymes (Azfaralariff et al. 2023; Kan et al. 2025). To illustrate the comparative abundance of amino acids in anchovies, Table 6 summarizes typical concentrations reported per 100 g of raw anchovy protein.

This amino acid richness contributes to the growing interest in anchovies as a raw material for bioactive peptide production. Enzymatic hydrolysis of anchovy protein using proteases such as alcalase or papain can yield short-chain peptides with demonstrated antioxidant, antihypertensive, and antimicrobial properties (Shaik and Sarbon 2022a; Kan et al. 2025). Such functional applications further elevate anchovies from traditional protein sources to ingredients of high potential in nutraceutical and pharmaceutical formulations.

Albumin and functional proteins

Albumin is a major water-soluble protein found in anchovy muscle tissue, serving as a critical biomolecule for maintaining osmotic pressure, binding and transporting endogenous and exogenous substances, and preserving structural integrity in both fish and human physiology. In *Stolephorus* spp., albumin content contributes not only to nutritional value but also to several biological activities, including antioxidant, anti-inflammatory, and immunomodulatory functions (Quan et al. 2020; Park et al. 2022). The concentration of albumin in fresh anchovy meat has been reported to range between 1.5 and 3.0% of total crude protein, varying by species and freshness status.

In the context of human health, fish-derived albumin is of particular interest due to its high digestibility and capacity to bind essential micronutrients such as calcium, zinc, and fatty acids. Albumin acts as a carrier protein for poorly water-soluble compounds, including free fatty acids and hormones, making it a functional element in food and pharmaceutical formulations. In anchovies, the presence of albumin is also linked to meat texture and flavor, contributing to consumer acceptability and culinary versatility (Barros et al. 2014).

Extraction of albumin from anchovies can be performed using hot water, salt solutions, or proteolytic enzymes. Among these, thermal extraction (e.g., 60-70°C for 30-60 minutes) is commonly applied to recover albumin in aqueous form for use in the food industry (Park et al. 2022). The isolated albumin can be dried into powder form and utilized in dietary supplements, protein fortifiers, or pharmaceutical carriers. In several studies, albumin-rich extracts from anchovies have demonstrated stability, solubility, and potential bioactivity comparable to bovine serum albumin, suggesting their promise as alternative protein ingredients (Kan et al. 2025).

Table 6. Essential amino acid profile of anchovy protein hydrolysates

Amino acid	Content (mg/100 g edible portion)	Classification	Physiological role
Lysine	1,600-1,800	Essential	Calcium absorption, collagen synthesis
Leucine	1,700-1,900	Essential	Muscle repair, metabolic signaling
Isoleucine	900-1,100	Essential	Hemoglobin formation, energy production
Methionine	700-850	Essential	Antioxidant precursor (glutathione), liver health
Threonine	800-950	Essential	Immune regulation, protein balance
Valine	800-1,000 (estimated, not always reported)	Essential	Muscle metabolism, tissue repair
Phenylalanine	~950-1,050 (in some hydrolysates)	Essential	Precursor for neurotransmitters
Histidine	~600-700 (in growth stages)	Essential (semi)	Growth, tissue repair, and histamine synthesis
Tryptophan	~250-350 (low in fish, variable)	Essential	Serotonin precursor, mood regulation

Sources: Andriani and Muhdar (2021); Kari et al. (2022); Azfaralariff et al. (2023)

Aside from albumin, anchovies also contain other functional proteins such as myosin, actin, tropomyosin, and enzymatic peptides, which contribute to physiological and biochemical properties. These proteins are involved in muscle contraction, post-mortem degradation, and are precursors to bioactive peptides with diverse effects, including antihypertensive and antioxidant activities (Hortillosa et al. 2022; Shaik and Sarbon 2022a). During enzymatic hydrolysis, these structural proteins are broken down into smaller peptides that retain or even enhance biofunctionality, making anchovies an excellent raw material for functional ingredient development. A summary of the main functional proteins and their health-relevant roles is presented in Table 7 and Figure 5.

The functional protein profile of anchovies supports their growing use not only as a staple protein source but also as a platform for bioengineering novel bioactive compounds. Continued research into isolation, characterization, and standardization of these proteins is necessary to unlock their health and industrial potential fully (Musalipassi et al. 2021).

Mineral binding and bioavailability

Anchovies (*Stolephorus* spp.) are excellent natural sources of essential dietary minerals, including calcium, magnesium, iron, zinc, and selenium. Beyond their nutritional abundance, these minerals are bioavailable due to their binding to specific organic molecules such as peptides, proteins (e.g., albumin), and phospholipids within the fish tissue. This mineral-protein interaction facilitates the stability, solubility, and absorption of minerals during digestion, enhancing their functional value in human nutrition (Swastawati et al. 2020; Chen et al. 2022b).

Calcium is the most dominant mineral found in anchovies, especially when consumed with bones (Safitri et al. 2023). It is predominantly bound to collagen and phosphoproteins in the skeletal matrix, allowing for gradual release and absorption during gastrointestinal digestion. Anchovies can provide up to 1,000 mg of calcium per 100 g in dried or whole forms, covering close to the daily recommended intake in many populations with limited dairy consumption. Magnesium and phosphorus are similarly associated with bone and muscle proteins and play synergistic roles in skeletal development and energy metabolism (Lall and Kaushik 2021).

Iron in anchovies is present in both heme and non-heme forms and is typically bound to myoglobin and ferritin

proteins, which enhances its bioavailability relative to plant sources. This makes anchovies a useful dietary component for preventing iron deficiency anemia, particularly in women and children. The zinc content is bound to metalloproteins and enzymes, supporting catalytic activity, immune regulation, and tissue repair. Meanwhile, selenium, often found in the form of selenoproteins, contributes to antioxidant enzyme activity and thyroid function (Galasso et al. 2020a).

These mineral-binding mechanisms are significant because they reduce the likelihood of mineral precipitation in the digestive tract and enhance transport across intestinal membranes. The interaction between proteins and divalent metal ions is regulated by specific amino acid residues such as histidine, cysteine, and glutamic acid that form coordinated covalent bonds with metal ions, stabilizing them in bioavailable forms (Park et al. 2022). This makes anchovy-derived mineral complexes favorable for use in functional food and nutraceutical formulations. To better illustrate the role of anchovies in delivering essential minerals, Table 8 summarizes the key minerals found, their binding forms, and associated physiological functions. The high mineral bioavailability of anchovies, coupled with their affordability and widespread accessibility, supports their inclusion in public health nutrition strategies targeting micronutrient deficiencies. Furthermore, anchovy-based mineral-peptide complexes are being explored as ingredients in fortification programs and therapeutic food products for vulnerable populations.

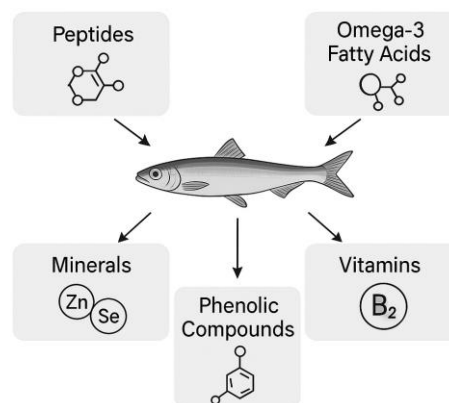


Figure 5. Overview of bioactive compounds identified in anchovy tissues

Table 7. Functional roles of albumin and other anchovy-derived proteins

Protein/peptide	Relative abundance	Functional role	Application potential
Albumin	Moderate (1.5-3%)	Osmoregulation, fatty acid, and micronutrient transport	Nutritional supplements, pharmaceutical carriers
Myosin	High	Muscle contraction, structural integrity	Source of bioactive peptides, texture enhancement
Actin	High	Cytoskeletal framework, elasticity	Functional protein source, peptide precursor
Tropomyosin	Medium	Regulatory protein in muscle contraction	Allergen marker, possible peptide hydrolysate precursor
Protein hydrolysates	Variable	Antioxidant, antihypertensive, and antimicrobial activities	Functional foods, nutraceuticals, therapeutic candidates

Sources: Park et al. (2022); Shaik and Sarbon (2022); Sun et al. (2022)

Enzymatic and hormonal components

Anchovies (*Stolephorus* spp.) contain a variety of endogenous enzymes and hormonal compounds that contribute to both their physiological function as living organisms and their potential value as bioactive food ingredients. These enzymatic components including proteases, lipases, and amylases play central roles in muscle metabolism, digestion, post-mortem autolysis, and preservation of fish freshness. In the context of food processing and biotechnology, such enzymes are also harnessed to produce bioactive peptides through hydrolysis, enhancing the nutraceutical potential of anchovy-derived products (Jeyashakila et al. 2022; Hortillosa et al. 2022).

Proteases, the most abundant among anchovy enzymes, facilitate the breakdown of muscle proteins into peptides and free amino acids. This proteolytic activity is crucial not only for muscle turnover in vivo but also for generating antioxidant, antihypertensive, and antimicrobial peptides during post-harvest fermentation or enzymatic hydrolysis (Shaik and Sarbon 2022a). Lipases, meanwhile, catalyze the hydrolysis of triglycerides into free fatty acids and glycerol, influencing flavor formation and lipid digestion. In anchovy fermentation products such as fish sauce, endogenous lipases contribute to the release of omega-3-rich lipid fractions with recognized health benefits (Kan et al. 2025).

Although less abundant in fish than in herbivorous species, amylases are present in trace amounts in anchovy gut tissue and assist in the partial digestion of glycogen or

starch residues from ingested plankton. These enzymes may also have applications in microbial enzyme production when anchovy tissues are used as substrates in fermentation media (Oktavia et al. 2018).

In addition to enzymes, anchovies possess hormonal substances that regulate metabolic and physiological processes, both within their own systems and as potentially bioactive components in human health. These include thyroid hormones (e.g., thyroxine/T4 and triiodothyronine/T3), steroid precursors, and small amounts of peptide hormones (Nurmawati et al. 2022). Although present in low concentrations, these hormones may exert biological effects after ingestion, particularly when derived from concentrated extracts or fermented preparations.

Thyroid hormones in fish regulate growth, metamorphosis, and basal metabolism. When consumed, these hormones-or their precursors, such as iodine-bound compounds may contribute to thyroid support in iodine-deficient populations. Some steroidal precursors, such as cholesterol derivatives found in anchovies, also serve as substrates for endogenous hormone biosynthesis in the human body. Table 9 summarizes the main enzymatic and hormonal compounds found in anchovies and their known or potential biological significance. Although the direct pharmacological use of anchovy-derived hormones remains limited, understanding their presence opens new avenues for research in marine-based bioactive compounds. Advances in extraction, purification, and delivery systems may enable future development of functional products utilizing these enzymatic and hormonal elements.

Table 8. Bioavailability of minerals and their binding affinity in anchovy tissues

Mineral	Average content (mg/100 g)	Primary binding form	Physiological function
Calcium	450-1000	Bone matrix (collagen, phosphoprotein)	Bone health, muscle contraction
Magnesium	60-120	Complexed with phospholipids	ATP metabolism, neuromuscular function
Iron	2.0-4.5	Ferritin, myoglobin	Oxygen transport, red blood cell formation
Zinc	1.5-3.0	Metalloproteins, enzymes	Enzymatic catalysis, immune regulation
Selenium	30-60 µg	Selenoproteins (e.g., GPx)	Antioxidant defense, thyroid hormone activation
Phosphorus	200-300	Hydroxyapatite, nucleotides	Energy storage (ATP), bone mineralization

Sources: Swastawati et al. (2020); Chen et al. (2022); Kari et al. (2022)

Table 9. Enzymes and hormones identified in anchovy extracts and their physiological roles

Compound type	Specific molecule	Biological role in fish	Functional or health relevance
Protease	Trypsin, pepsin-like enzymes	Muscle turnover, digestion	Generation of antioxidant and ACE-inhibitory peptides
Lipase	Pancreatic-type lipase	Lipid mobilization, flavor development	Facilitates lipid metabolism, improves digestibility
Amylase	α-Amylase (low abundance)	Digestion of glycogen/starch in the gut	Potential in prebiotic fermentation processes
Thyroid hormone	T3, T4	Growth regulation, metabolic rate control	May support thyroid function in iodine-deficient populations
Steroidal precursor	Cholesterol	Hormone biosynthesis	Substrate for steroid and vitamin D synthesis
Peptide hormone	Insulin-like peptides	Glucose regulation	Potential anti-diabetic activity through glycemic control

Sources: Oktavia et al. (2018); Hortillosa et al. (2022); Nurmawati et al. (2022)

BIOLOGICAL ACTIVITIES OF ANCHOVIES

Antioxidant mechanisms

Oxidative stress defined as an imbalance between the production of reactive oxygen species (ROS) and the capacity of biological systems to neutralize them—is a central contributor to cellular damage, aging, and chronic diseases such as cancer, cardiovascular disorders, and neurodegenerative conditions. Anchovies (*Stolephorus* spp.), as marine food resources, offer a promising source of antioxidant compounds that counteract oxidative stress through multiple molecular pathways (Galasso et al. 2020a; Engwa et al. 2022).

The antioxidant potential of anchovies is primarily attributed to three groups of bioactive components: (i) omega-3 polyunsaturated fatty acids (PUFAs), particularly EPA and DHA; (ii) antioxidant peptides generated through enzymatic hydrolysis of anchovy protein; and (iii) trace minerals and vitamins with redox-regulating functions, including selenium, zinc, vitamin E, and vitamin D (Hasari et al. 2021; Sun et al. 2022). These compounds operate both independently and synergistically to neutralize free radicals, chelate pro-oxidant metals, and modulate intracellular antioxidant enzyme systems such as glutathione peroxidase (GPx), superoxide dismutase (SOD), and catalase (CAT).

Antioxidant peptides, typically consisting of 2-20 amino acids, are liberated from anchovy muscle proteins through enzymatic hydrolysis using enzymes such as alcalase, papain, or pepsin. These peptides contain amino acid residues such as histidine, tyrosine, cysteine, or methionine that donate electrons or hydrogen atoms to neutralize ROS (Shaik and Sarbon 2022a) (Figure 6). In vitro studies using DPPH and ABTS assays have demonstrated that anchovy protein hydrolysates exhibit high radical-scavenging activity, comparable to that of synthetic antioxidants like BHA or tocopherol under certain conditions (Kan et al. 2025).

Omega-3 fatty acids, while not classical antioxidants, exert indirect antioxidant effects by modulating inflammatory signaling and enhancing the activity of cellular antioxidant defenses. EPA and DHA have been shown to activate Nrf2 signaling pathways, which induce the expression of endogenous antioxidant enzymes. They also inhibit the production of pro-inflammatory cytokines and lipid peroxidation products that would otherwise promote oxidative tissue damage (Yang et al. 2020) (Figure 7).

The contribution of micronutrients is also notable. Selenium, present in anchovies as selenoproteins, plays an essential role in the catalytic function of GPx, which reduces hydrogen peroxide and lipid hydroperoxides. Zinc stabilizes cell membranes and modulates metallothionein expression, which scavenges hydroxyl radicals. Vitamin E (tocopherols) interrupts lipid peroxidation chains in membranes, while vitamin D may contribute to redox homeostasis through immunomodulatory functions (Barros et al. 2014). The mechanisms and sources of antioxidant activity in anchovies are summarized in Table 10.

The multifunctionality of anchovy-derived antioxidants makes them suitable candidates for inclusion in functional foods, dietary supplements, and pharmaceutical preparations targeting oxidative stress-related disorders. Future research should focus on standardizing hydrolysis protocols, identifying specific antioxidant peptide sequences, and conducting clinical trials to validate efficacy in vivo.

Antimicrobial and anti-inflammatory effects

Anchovies (*Stolephorus* spp.) exhibit significant antimicrobial and anti-inflammatory activities, primarily due to the presence of bioactive peptides, unsaturated fatty acids, and trace elements that interfere with microbial viability and inflammatory signaling pathways. These bioactivities are particularly evident in anchovy protein hydrolysates and fermented anchovy products, which contain concentrated peptides with specific biological functions (Shaik and Sarbon 2022a; Kan et al. 2025).

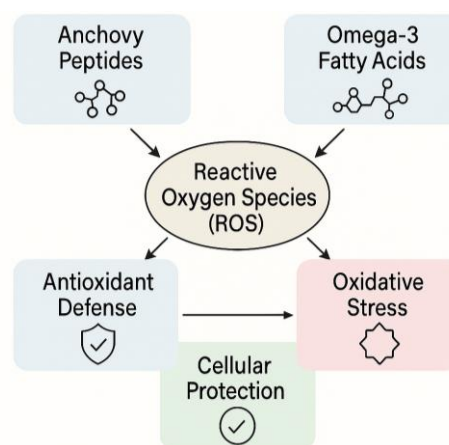


Figure 6. Mechanism of antioxidant activity of anchovy peptides and omega-3 fatty acids

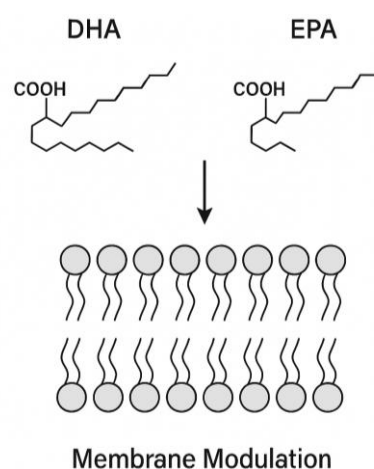


Figure 7. Molecular structure of DHA and EPA and their role in membrane modulation

Table 10. Antioxidant assays and efficacy values of anchovy-derived compounds

Component type	Source in anchovy	Assay method	Reported activity / efficacy	Mechanism of action
Antioxidant peptides	Protein hydrolysates (enzymatic)	DPPH, ABTS	70-90% radical scavenging at 1-5 mg/mL	Electron donation, metal chelation
Omega-3 fatty acids	Muscle lipids (EPA, DHA)	Lipid peroxidation assay (TBARS)	40-60% inhibition of MDA formation in vitro	Membrane stabilization, Nrf2 activation
Selenium	Selenoproteins (GPx precursor)	GPx enzyme activity	Increased GPx activity in supplemented models	Reduces hydroperoxides, supports redox balance
Zinc	Metalloproteins	SOD activity assay	Upregulated SOD expression in tissue cultures	Free radical scavenging, enzyme cofactor
Vitamin E	Lipid fractions	FRAP, ORAC	Comparable to α -tocopherol in lipid-soluble extracts	Breaks the lipid peroxidation chain
Vitamin D	Fat-soluble matrix	Cellular antioxidant assay	Modulates oxidative stress indirectly via immune regulation	Hormone-mediated antioxidant defense

Sources: Galasso et al. (2020); Hasari et al. (2021); Shaik and Sarbon (2022); Sun et al. (2022);

Antimicrobial activity is primarily attributed to small peptides (<10 kDa) released during enzymatic hydrolysis using proteases such as pepsin, trypsin, or alcalase. These peptides often possess amphipathic structures and cationic charges that enable them to bind to negatively charged bacterial membranes, causing disruption, pore formation, and eventual cell lysis (Hortillosa et al. 2022). Studies have shown that anchovy-derived peptides exhibit inhibitory effects against a diverse range of Gram-positive and Gram-negative bacteria, including *Escherichia coli*, *Staphylococcus aureus*, *Salmonella enterica*, and *Listeria monocytogenes*. This broad-spectrum antimicrobial activity is a testament to the potential of these peptides as natural food preservatives or as leads for antimicrobial drug development.

Fermented anchovy products, such as fish sauce, have also demonstrated significant antimicrobial effects. This is due to the presence of low-molecular-weight peptides, organic acids, and high salt concentrations that effectively inhibit microbial growth. Additionally, anchovy lipids—particularly omega-3 fatty acids—are known to exert bacteriostatic or bactericidal effects by disrupting microbial membranes and suppressing the synthesis of proinflammatory lipopolysaccharides in pathogenic strains (Park et al. 2022).

On the anti-inflammatory front, anchovy-derived compounds modulate inflammatory responses through both direct and indirect mechanisms. Omega-3 fatty acids (EPA and DHA) are precursors to specialized pro-resolving mediators (SPMs) such as resolvins and protectins, which actively terminate inflammation by inhibiting neutrophil infiltration, reducing cytokine production (e.g., TNF- α , IL6), and promoting macrophage-mediated clearance of inflammatory debris (Yang et al. 2020) (Figure 8). These effects are further enhanced by selenium and zinc, which stabilize cellular redox status and downregulate NF- κ B-mediated pro-inflammatory gene expression.

Protein hydrolysates from anchovies have also been shown to reduce nitric oxide (NO) production and prostaglandin E2 (PGE2) synthesis in lipopolysaccharide-stimulated macrophage models. These actions reflect the immunomodulatory potential of specific peptide sequences

that interfere with inducible Nitric Oxide Synthase (iNOS) and Cyclooxygenase-2 (COX-2) expression, two key mediators in chronic inflammatory conditions (Shaik and Sarbon 2022a). A summary of these effects is presented in Table 11.

The dual antimicrobial and anti-inflammatory actions of anchovy-derived compounds highlight their relevance in developing functional foods, nutraceuticals, and even natural therapeutics. Their potential to reduce reliance on synthetic antibiotics and anti-inflammatory drugs warrants further exploration, including clinical validation and formulation into safe, bioavailable products.

Antithrombotic and cardiovascular benefits

Cardiovascular Diseases (CVDs) remain the leading cause of global mortality, largely driven by a combination of thrombosis, dyslipidemia, inflammation, and oxidative stress. Anchovies (*Stolephorus* spp.) contain a suite of bioactive compounds most notably omega-3 fatty acids, peptides, and minerals such as selenium that collectively contribute to cardioprotective effects, including antithrombotic, anti-inflammatory, and lipid-modulating properties (Galasso et al. 2020a; Yang et al. 2020).

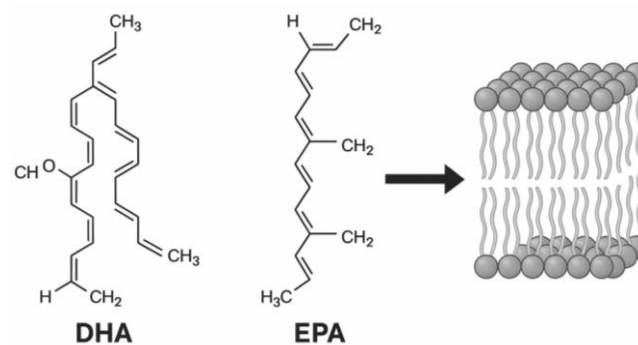


Figure 8. Pathways of antimicrobial and anti-inflammatory effects of anchovy peptides

The antithrombotic potential of anchovies is primarily linked to the high content of long-chain omega-3 polyunsaturated fatty acids (PUFAs), especially eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). These fatty acids competitively inhibit arachidonic acid metabolism, leading to the production of thromboxane A3 (TXA3) a weak platelet aggregator and vasoconstrictor instead of thromboxane A2 (TXA2), which strongly promotes platelet aggregation and vasoconstriction. By shifting the eicosanoid balance, omega-3s from anchovies reduce platelet activation and blood clot formation (Barros et al. 2014).

Additionally, EPA and DHA are precursors of resolvins, specialized pro-resolving lipid mediators that not only limit neutrophil infiltration in vascular tissue but also promote endothelial healing and vasodilation. These compounds work synergistically with selenium, which functions as a cofactor for Glutathione Peroxidase (GPx), reducing lipid peroxidation in vascular endothelium and thus preserving vessel integrity (Chen et al. 2022a).

Protein hydrolysates from anchovies have also demonstrated promising cardiovascular effects. Certain peptides exhibit Angiotensin-Converting Enzyme (ACE) inhibitory activity, thus contributing to blood pressure

reduction. These peptides mimic the action of pharmacological ACE inhibitors, preventing the conversion of angiotensin I to angiotensin II, a potent vasoconstrictor, and promoting vasodilation through bradykinin preservation (Shaik and Sarbon 2022a).

Moreover, anchovy consumption is associated with improved lipid profiles. Studies indicate that regular intake of small pelagic fish rich in omega-3s can lower serum triglycerides, increase HDL cholesterol, and reduce LDL oxidation all of which contribute to reduced risk of atherosclerosis and myocardial infarction. Anchovy-derived phospholipids also support lipid transport and membrane fluidity, important for maintaining cardiovascular homeostasis (Park et al. 2022).

Table 12 summarizes the cardiovascular and antithrombotic benefits of anchovy-derived bioactive compounds. The multifaceted cardiovascular benefits of anchovies make them a valuable component in heart-healthy diets. Their bioactive compounds act on multiple levels from endothelial protection to platelet function regulation offering both preventive and adjunctive potential for CVD management. Future work should focus on peptide isolation, clinical trials, and formulation into targeted nutraceuticals.

Table 11. Antimicrobial and anti-inflammatory bioactivities of anchovy peptides

Bioactive component	Source or derivation	Target / mechanism	Observed bioactivity	Application potential
Antimicrobial peptides	Enzymatic protein hydrolysates	Disrupt bacterial membranes, inhibit metabolism	Inhibitory effect against <i>E. coli</i> , <i>S. aureus</i> , <i>Salmonella</i>	Natural preservatives, antimicrobial nutraceuticals
Omega-3 fatty acids	Muscle lipids (EPA, DHA)	Bacterial membrane disruption, LPS suppression	Bacteriostatic effects; synergy with peptides	Immunonutrition, infection control
Organic acids	Fermented anchovy products	Low pH, microbial metabolic inhibition	Inhibits spoilage and pathogenic bacteria	Traditional food preservation
Selenium & zinc	Muscle-bound trace minerals	NF-κB inhibition, redox stabilization	Downregulates pro-inflammatory cytokines (TNF-α, IL-6)	Anti-inflammatory support
Anti-inflammatory peptides	Peptide fractions from hydrolysis	Suppress iNOS and COX-2, inhibit PGE2 and NO production	Reduced inflammation in macrophages and LPS-induced models	Functional food for inflammatory diseases
Resolvin precursors	EPA and DHA	Formation of SPMs (resolvins, protectins)	Promotes resolution of inflammation and tissue repair	Chronic inflammation management

Sources: Yang et al. (2020); Hortillosa et al. (2022); Park et al. (2022); Shaik and Sarbon (2022)

Table 12. Cardiovascular and metabolic regulatory effects of anchovy omega-3s

Compound / component	Mechanism of action	Health effect	Target pathway or marker
EPA and DHA (omega-3 PUFAs)	Inhibit thromboxane A2 (TXA2) synthesis, produce TXA3	Antithrombotic, vasodilation	Platelet aggregation, eicosanoid profile
EPA and DHA	Precursor of resolvins and protectins	Resolution of inflammation, endothelial protection	SPM signaling, inflammation markers
ACE-inhibitory peptides	Block conversion of angiotensin I to angiotensin II	Antihypertensive effect	Renin-angiotensin system
Selenium	Cofactor of glutathione peroxidase (GPx)	Endothelial antioxidant protection	Lipid peroxidation (MDA), GPx activity
Phospholipids	Improve lipid transport and membrane fluidity	Elevate HDL, reduce LDL oxidation	Lipid profile (HDL, LDL, TG)
Bioactive peptides	Inhibit DPP-IV and α-glucosidase	Improve insulin secretion, delay glucose absorption	Blood glucose, incretin activity
Zinc	Stabilizes insulin, supports β-cell activity	Enhance insulin synthesis and storage	Pancreatic function
Adiponectin modulation	Upregulated by omega-3s	Improves insulin sensitivity	Adipokine profile

Sources: Galasso et al. (2020); Yang et al. (2020); Chen et al. (2022); Park et al. (2022); Shaik and Sarbon (2022)

Anti-diabetic and metabolic regulation

Metabolic disorders, particularly type 2 diabetes mellitus (T2DM), are characterized by insulin resistance, hyperglycemia, and chronic low-grade inflammation. Increasing attention has been directed toward dietary interventions involving bioactive compounds from marine resources, including anchovies (*Stolephorus* spp.), which offer promising antidiabetic and metabolic regulatory effects through multiple biochemical pathways (Galasso et al. 2020a; Shaik and Sarbon 2022a).

One of the primary antidiabetic components in anchovies is omega-3 polyunsaturated fatty acids (PUFAs), especially EPA and DHA. These fatty acids enhance insulin sensitivity by modulating membrane fluidity, activating peroxisome proliferator-activated receptors (PPARs), and reducing adipose tissue inflammation. EPA and DHA have been shown to downregulate proinflammatory cytokines (e.g., TNF- α , IL-1 β) and upregulate adiponectin a hormone that promotes glucose uptake and fatty acid oxidation thus improving systemic glucose homeostasis (Yang et al. 2020).

Protein hydrolysates from anchovy muscle also exhibit antidiabetic activity, particularly by inhibiting α glucosidase and dipeptidyl peptidase-IV (DPP-IV). α Glucosidase inhibitors delay carbohydrate breakdown and glucose absorption in the intestine, thereby reducing postprandial blood glucose spikes. DPP-IV inhibitors, on the other hand, prolong the half-life of incretin hormones (GLP-1 and GIP), which stimulate insulin secretion and suppress glucagon release (Shaik and Sarbon 2022a). These effects have been validated in vitro and animal models, suggesting anchovy peptides as functional ingredients for glycemic control.

Minerals such as zinc and selenium, abundantly present in anchovies, are critical for insulin synthesis, storage, and secretion. Zinc stabilizes insulin hexamers and contributes to β -cell function, while selenium exerts antioxidant protection for pancreatic tissues through glutathione peroxidase activity (Chen et al. 2022a). The combined effect of these micronutrients supports metabolic regulation and reduces oxidative damage associated with diabetic complications.

Additionally, anchovy intake has been linked to improved lipid metabolism, a crucial factor in managing metabolic syndrome. EPA and DHA reduce hepatic lipogenesis and increase fatty acid β -oxidation via PPAR- α activation. These changes contribute to reduced serum triglycerides and improved lipid profiles, indirectly supporting insulin sensitivity and lowering cardiovascular

risk in diabetic patients (Park et al. 2022). The key metabolic regulatory effects of anchovy-derived components are summarized in Table 13 and Figure 9. These findings highlight anchovies as a functional dietary option for metabolic health, with integrated effects on glucose regulation, insulin signaling, and lipid metabolism. Future research should focus on identifying and purifying the most active peptide fractions and on evaluating their efficacy through human clinical trials.

Anti-cancer and anti-carcinogenic properties

Marine-derived foods are increasingly recognized for their chemopreventive potential, owing to a diversity of bioactive compounds capable of modulating cellular proliferation, oxidative stress, apoptosis, and inflammation. Anchovies (*Stolephorus* spp.) contain multiple classes of such compounds including omega-3 fatty acids, antioxidant peptides, and trace elements that may contribute to anticancer and oral health-promoting (anti-carcinogenic) effects (Galasso et al. 2020a; Shaik and Sarbon 2022a).

Anti-cancer effects

The anticancer potential of anchovy-derived compounds is primarily mediated through apoptosis induction, inhibition of inflammatory signaling, and oxidative stress reduction. EPA and DHA, the dominant omega-3 fatty acids in anchovies, modulate membrane lipid composition and reduce the activity of pro-inflammatory transcription factors such as NF- κ B and STAT3, which are commonly overexpressed in cancer cells. These effects lead to decreased expression of genes involved in cell survival and angiogenesis (e.g., Bcl-2, VEGF) and increased proapoptotic signaling (e.g., Bax, caspases) (Yang et al. 2020) (Figure 10).

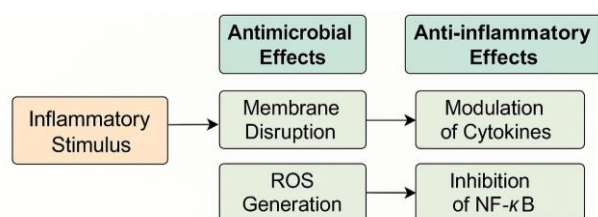


Figure 9. Proposed mechanisms of anchovy-derived compounds in metabolic regulation

Table 13. Anti-diabetic and metabolic regulatory compounds in anchovies (*Stolephorus* spp.)

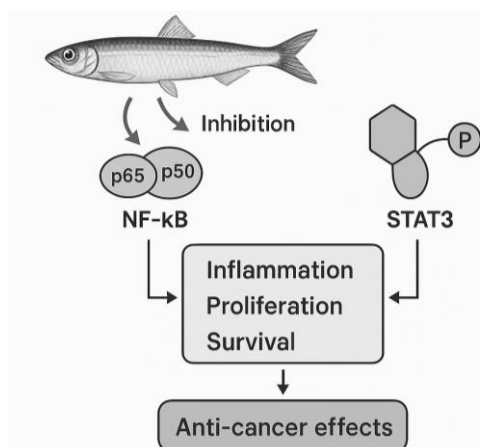
Compound / component	Mechanism of action	Metabolic effect
EPA and DHA (omega-3 PUFAs)	Activate PPARs, reduce inflammation, and increase adiponectin	Enhance insulin sensitivity, reduce glucose levels
Bioactive peptides	Inhibit α -glucosidase and DPP-IV	Delay glucose absorption, increase insulin release
Zinc	Stabilizes insulin, supports β -cell function	Promotes insulin secretion
Selenium	Antioxidant via GPx activity	Protects pancreatic islets
Phospholipids	Improve cell signaling and fat transport	Regulate lipid and glucose metabolism

Sources: Galasso et al. (2020a); Yang et al. (2020); Chen et al. (2022a); Shaik and Sarbon (2022a)

Table 14. Anti-cancer and anti-carcinogenic compounds in anchovies (*Stolephorus* spp.)

Bioactive component	Mechanism of action	Target / effect	Health relevance
EPA and DHA (omega-3)	Inhibit NF- κ B and STAT3 signaling; induce apoptosis	↓ Bcl-2, ↑ caspase activity, ↓ VEGF	Suppress tumor growth, angiogenesis
Antioxidant peptides	Scavenge ROS; disrupt mitochondrial function	↑ Bax/Bak, ↓ cell proliferation	Cytotoxic to cancer cells
Selenium	Incorporated in selenoproteins; modulates apoptosis	↑ DNA repair, ↑ immune surveillance	Reduces oxidative DNA damage, tumor risk
Zinc	Cofactor in DNA repair enzymes and tumor suppressor proteins	↓ Mutation rate, ↑ cell cycle regulation	Maintains genomic stability
Antimicrobial peptides	Disrupt <i>Streptococcus mutans</i> membrane integrity	↓ Plaque formation, ↓ acid production	Prevent dental caries
Protein hydrolysates	Inhibit bacterial metabolism and biofilm formation	↓ <i>S. mutans</i> adhesion, ↓ lactic acid production	Promote oral hygiene and cariostatic effects
Omega-3 fatty acids	Anti-inflammatory action in periodontal tissues	↓ Gingival inflammation, ↑ healing	Support oral and gum health

Sources: Yang et al. (2020); Hortillosa et al. (2022); Sun et al. (2022); Shaik and Sarbon (2022)

**Figure 10.** Anti-cancer mechanisms via NF- κ B/STAT3 inhibition by anchovy bioactives

In vitro studies have shown that anchovy protein hydrolysates can inhibit the proliferation of human colon, breast, and liver cancer cells by arresting the cell cycle and inducing mitochondrial dysfunction. These effects are often linked to specific low-molecular-weight peptides rich in hydrophobic or aromatic amino acids that interact with cancer cell membranes or intracellular targets (Kan et al. 2025).

Additionally, selenium and zinc in anchovies support DNA repair, inhibit tumor growth, and enhance immune surveillance. Selenium, in particular, has been implicated in reducing cancer risk via its incorporation into selenoproteins, which act as antioxidants and modulators of apoptosis (Table 14).

Anti-carcinogenic effects

Anchovies also show promise as functional foods in promoting oral health, especially by exerting anticarcinogenic effects. Antimicrobial peptides and enzymes derived from anchovy muscle or fermented anchovy products (e.g., fish sauce) inhibit the growth of

Streptococcus mutans, a key bacterium responsible for dental plaque formation and tooth decay (Hortillosa et al. 2022).

Omega-3 fatty acids contribute to oral health by reducing gingival inflammation and oxidative stress, while zinc plays a direct role in inhibiting plaque formation and reducing halitosis (Rajaram et al. 2021). Moreover, anchovy-derived protein hydrolysates have been shown to decrease biofilm production in vitro, suggesting their potential application in functional oral care products.

Taken together, the anticancer and anti-carcinogenic properties of anchovies reflect their potential as biofunctional ingredients for both preventive healthcare and oral hygiene applications. These dual benefits underscore the importance of expanding research into specific compound isolation, bioactivity validation, and clinical formulation development (Shaik and Sarbon 2022a).

Structure-function relationship of bioactive compounds

The biological activities exhibited by anchovy-derived compounds are closely related to their molecular structure, particularly the presence of specific functional groups, chain lengths, and conformational properties that determine their interaction with biological targets. Understanding this structure-function relationship is essential for elucidating the mechanisms underlying the observed antioxidant, antimicrobial, anti-inflammatory, and metabolic effects of anchovy-based compounds.

Bioactive peptides, generated through the enzymatic hydrolysis of anchovy proteins, are typically composed of 2 to 20 amino acid residues with distinct physicochemical characteristics (Kari et al. 2022). Their activity is largely governed by the presence of hydrophobic (e.g., leucine, valine), aromatic (e.g., tyrosine, phenylalanine), and sulfur-containing residues (e.g., cysteine, methionine). These side chains facilitate membrane interactions, radical scavenging, or enzyme inhibition depending on their orientation and charge. For instance, the ability of a peptide to scavenge free radicals correlates with the electron-donating potential

of phenolic and sulfur groups. In comparison, peptides with basic residues (e.g., lysine, arginine) are more effective at binding to bacterial membranes due to electrostatic attraction.

The structure of omega-3 fatty acids, especially eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), is also critical to their function. These long-chain polyunsaturated fatty acids contain multiple cis-double bonds that create kinks in the carbon backbone, enhancing their fluidity and enabling their integration into cellular membranes (Hishikawa et al. 2017). This structural property modulates membrane microdomains, affecting receptor function, signal transduction, and lipid mediator production. Moreover, the position and number of double bonds influence their metabolic conversion into bioactive lipid mediators such as resolvins and protectins, which are responsible for anti-inflammatory and pro-resolving effects (Bannenberg 2009).

Trace elements, such as selenium and zinc, are highly dependent on coordination chemistry and protein-binding specificity for their bioactivity. Selenium exerts its function when incorporated as selenocysteine into the active sites of enzymes like glutathione peroxidase, where the Se-H group serves as a redox-active center. Zinc, meanwhile, stabilizes protein domains (e.g., zinc fingers) or acts as a cofactor for enzymes like Superoxide Dismutase (SOD), with its tetrahedral coordination geometry enabling electron transfer reactions essential for antioxidative processes (Klotz et al. 2003; Wołonciej et al. 2016).

Finally, the spatial conformation of anchovy-derived enzymes and hormones plays a key role in determining their biological function. Structural domains such as the active sites of proteases or the receptor-binding motifs of peptide hormones must maintain precise folding to interact with substrates or cellular receptors. Even slight modifications in tertiary or quaternary structure due to pH, temperature, or hydrolysis can significantly alter bioavailability and efficacy (Kari et al. 2023).

Collectively, the functional diversity of anchovy bioactives stems from a high degree of structural specialization, which enables specific interactions with molecular targets in human physiology. Advances in proteomics, lipidomics, and molecular modeling continue to reveal how these structural attributes translate into potent biological effects, guiding the development of targeted nutraceuticals and functional foods based on anchovy compounds (Giannetto et al. 2016).

ENVIRONMENTAL AND PUBLIC HEALTH CONSIDERATIONS

Heavy metal and microplastic contamination risks

While anchovies (*Stolephorus* spp.) are widely regarded as a nutritious and bioactive-rich food resource, increasing concerns have emerged regarding their contamination by heavy metals and microplastics, particularly in coastal and estuarine environments subject to anthropogenic pollution. As small pelagic fish with high metabolic rates and wide foraging behavior, anchovies may accumulate contaminants both through trophic transfer and direct environmental exposure (Karami et al. 2017; Barhoumi et al. 2022).

Heavy metal accumulation in anchovies is primarily influenced by local water quality, sediment characteristics, and bioavailability of toxic elements such as mercury (Hg), cadmium (Cd), lead (Pb), and arsenic (As) (Table 15). Although anchovies are relatively low on the food chain and thus expected to exhibit lower biomagnification than apex predators, recent studies have detected variable levels of these metals in anchovy tissues depending on the region. For example, anchovy samples from industrialized coastal zones in the Mediterranean, South China Sea, and Indonesian estuaries have shown detectable concentrations of Pb and Cd exceeding WHO/FAO recommended safety thresholds (Syakti et al. 2019; Riani et al. 2024). Chronic exposure to such metals through frequent consumption may pose risks, including neurotoxicity, renal impairment, and carcinogenicity, especially among vulnerable populations such as pregnant women and children.

In parallel, microplastic contamination has emerged as a global threat to marine food safety. Anchovies are known to ingest microplastics defined as plastic fragments <5 μm either directly by mistaking them for plankton or indirectly through contaminated prey. Once ingested, these particles can accumulate in the gastrointestinal tract and, in some cases, translocate to edible tissues. The concern is not only the physical presence of the plastic particles but also their role as vectors for adsorbed persistent organic pollutants (POPs), including polychlorinated biphenyls (PCBs), dioxins, and polycyclic aromatic hydrocarbons (PAHs), which may leach into surrounding tissues and elicit endocrine-disrupting, mutagenic, or immune-suppressive effects (Rochman et al. 2015) (Figure 11).

Table 15. Heavy metal concentrations reported in anchovies from selected regions

Region / location	Mercury (Hg) (mg/kg)	Cadmium (Cd) (mg/kg)	Lead (Pb) (mg/kg)	Exceeds FAO/WHO (Limits)	Potential sources
North Coast of Java, Indonesia	0.03-0.11	0.02-0.09	0.15-0.35	Pb often exceeds limit	Urban runoff, industrial effluents
South China Sea	0.05-0.12	0.01-0.06	0.10-0.25	No or occasional exceedance	Shipping, aquaculture discharge
Mediterranean Sea (coastal)	0.07-0.18	0.03-0.10	0.20-0.40	Cd and Pb exceed limits	Industrial zones, historical contamination
Eastern Gulf of Thailand	0.01-0.06	0.01-0.04	0.08-0.20	Within safe limits	Low anthropogenic pressure
Estuaries of Sumatra	0.02-0.08	0.01-0.05	0.12-0.30	Pb near or above limit	Agriculture runoff, mining residue

Note: FAO/WHO Max Limits: Hg = 0.5 mg/kg; Cd = 0.1 mg/kg; Pb = 0.3 mg/kg (wet weight basis). Sources: Syakti et al. (2019); Barhoumi et al. (2022); Riani et al. (2024)

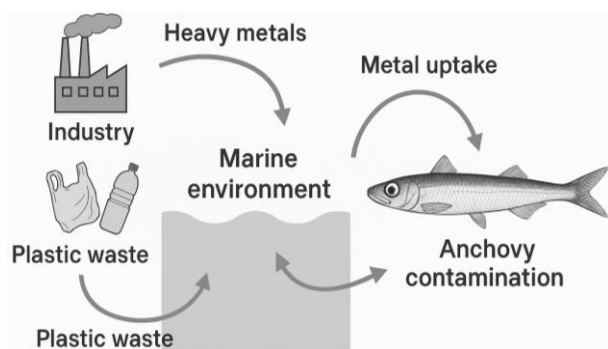


Figure 11. Sources and pathways of heavy metal and microplastic contamination in anchovies

Despite evisceration and cooking reducing some risk, certain forms of anchovy consumption such as whole dried fish, fermented paste, or powder supplements may retain higher contamination potential. These contaminants could include heavy metals like mercury or persistent organic pollutants like dioxins and PCBs. Moreover, bioaccumulated toxins may not be eliminated by traditional preparation methods, necessitating greater attention to source origin and monitoring protocols.

These concerns highlight the urgent need for routine surveillance of contaminant levels in anchovy stocks, especially in fisheries near densely populated or industrialized coastlines. Policy recommendations include tighter regulation of marine pollution sources, certification of seafood safety, and public advisories on maximum intake levels. However, it is crucial to remember that these are not the only solutions. Further studies are required to assess the bioaccessibility and toxicokinetics of contaminants in processed anchovy products and to explore mitigation strategies, such as depuration techniques or bioremediation.

Overexploitation and ecological impacts

As one of the most heavily harvested marine fish groups globally, anchovies (*Stolephorus* spp.) face increasing pressure from overfishing, driven by high demand for both human consumption and use as fishmeal in aquaculture and poultry feed industries. Their biological characteristics such as short lifespan, early maturation, and high fecundity make anchovies resilient to some extent. However, recent trends suggest that intensive and unregulated harvesting is outpacing their natural replenishment, particularly in coastal regions of Southeast Asia, West Africa, and the eastern Mediterranean (FAO 2022; Ali et al. 2025).

The ecological consequences of anchovy overexploitation extend beyond population declines. Anchovies occupy a key trophic position in marine food webs, acting as energy conduits between primary producers (phytoplankton, zooplankton) and higher trophic levels, including commercially important predatory fish, seabirds, and marine mammals. Reductions in anchovy biomass can trigger cascading effects disrupting predator foraging success, altering species interactions, and shifting

ecosystem structure and resilience (Smith et al. 2011). In areas like the Benguela and Humboldt Current systems, where anchovies dominate pelagic fish assemblages, such imbalances have led to regime shifts, with long-term impacts on biodiversity and fishery yields. Compounding these pressures, anchovy habitats particularly estuaries, coral reef edges, and nearshore upwelling zones are increasingly affected by climate change, pollution, and habitat degradation. Temperature anomalies, acidification, and coastal development influence spawning grounds and larval survival, exacerbating recruitment variability. This makes *Stolephorus* spp. particularly vulnerable to multistressor environments, where overfishing interacts with environmental stressors in complex and often unpredictable ways.

Addressing these challenges requires an EcosystemBased Fishery Management (EBFM) approach that integrates anchovy population dynamics, environmental variability, and multi-species interactions. Establishing spatial-temporal fishing regulations, catch limits, and community-based monitoring systems can help maintain sustainable stocks. Additionally, the inclusion of anchovy biomass trends in ecosystem health indicators is vital for marine spatial planning and conservation. Despite their economic and nutritional importance, anchovies are often undervalued in formal fisheries governance frameworks. This underrepresentation leads to data gaps in stock assessment and undermines the enforcement of conservation measures. Strengthening fisheries data collection especially for small-scale and artisanal fisheries along with regional collaboration, is critical to ensuring long-term sustainability.

Sustainable harvesting and management strategies

To safeguard the ecological and economic roles of anchovies (*Stolephorus* spp.) amid increasing exploitation pressures, the implementation of sustainable harvesting and fisheries management strategies is imperative. Given their pivotal role in tropical food webs and coastal economies, these strategies should be informed by biological stock assessments, ecosystem dynamics, and socioeconomic considerations, ensuring both the long-term viability of anchovy populations and continued benefits for coastal communities that depend on them (Salim et al. 2020; FAO 2022) (Figure 12). A foundational approach is the application of science-based catch limits, derived from Maximum Sustainable Yield (MSY) estimates and precautionary reference points. Regular stock assessments incorporating age-structured models, recruitment variability, and environmental indices are essential to prevent overexploitation. In regions with limited data availability, data-poor methodologies such as length-based indicators, Productivity-Susceptibility Analysis (PSA), or empirical harvest control rules can offer interim solutions for management (Froese et al. 2012). Seasonal and spatial fishing closures are effective in protecting anchovy spawning aggregations and juvenile recruitment zones. For instance, several countries have instituted moratoriums during peak breeding months or established Marine Protected Areas (MPAs) along critical nursery habitats.

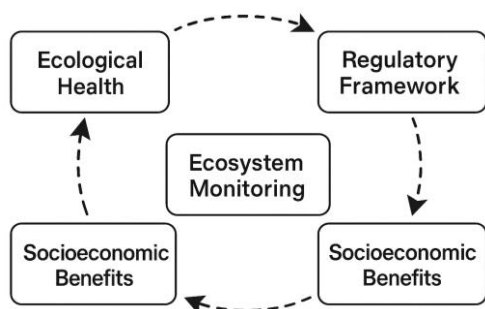


Figure 12. Conceptual framework of ecosystem-based anchovy fishery management, showing a cyclical interaction between ecosystem monitoring, regulatory enforcement, and ecological health. The first Socioeconomic Benefits node (right) refers to direct outcomes such as improved fishery income and local employment following regulatory compliance, while the second Socioeconomic Benefits node (bottom left) denotes long-term benefits including livelihood sustainability, food security, and social equity, driven by enhanced ecosystem and policy feedback

These measures support population recovery while minimizing ecological disruption, especially when designed with local fisher participation and compliance monitoring. The success of such interventions often depends on community trust, socio-cultural alignment, and enforceability. The integration of Ecosystem-Based Fisheries Management (EBFM) principles is also key. EBFM acknowledges the role of anchovies as mid-trophic connectors and seeks to balance their exploitation with the needs of dependent predators and ecosystem services. Such approaches could include multi-species stock modeling, habitat preservation, and food-web-based catch allocation strategies, especially in ecologically sensitive systems like coral reef margins and coastal upwelling zones (Pikitch et al. 2004). Moreover, cross-sector collaboration linking fisheries, conservation, and coastal development agendas is needed to operationalize EBFM effectively.

In many regions, anchovy fisheries are dominated by small-scale and artisanal operations, which pose both challenges and opportunities for sustainability. While these fisheries may have lower environmental footprints, they often lack institutional support, data recording, and access to sustainable technologies. Participatory governance and inclusive decision-making platforms, along with capacity building, co-management frameworks, and inclusion of Traditional Ecological Knowledge (TEK), can enhance stewardship and accountability in these sectors.

Emerging innovations such as selective fishing gear, low-impact harvest methods, and real-time monitoring systems can further reduce bycatch, habitat disturbance, and Illegal, Unreported, and Unregulated (IUU) fishing. Technological tools like electronic logbooks, machine learning for stock prediction, mobile applications for catch reporting, and satellite-based vessel tracking are gaining traction in community-based fisheries governance.

Lastly, promoting market-based incentives such as ecolabeling (e.g., MSC certification), fair trade agreements, and value-added product development can align economic interests with sustainability goals. By fostering traceable

supply chains and rewarding responsible practices, these mechanisms encourage fishers and processors to adopt conservation-minded approaches. Consumer education campaigns and retailer partnerships can amplify these incentives and drive demand for sustainably sourced anchovies. Collectively, these strategies underscore the need for integrated, adaptive, and participatory management systems that reflect ecological realities and social contexts. Anchovies, as fast-growing and high-value species, offer an ideal test case for advancing holistic marine resource management in the face of global change. Their management must remain dynamic, evidence-driven, and inclusive to ensure long-term ecological and socioeconomic resilience.

APPLICATIONS AND FUTURE PERSPECTIVES

Functional food and nutraceutical potentials

The increasing recognition of the health-promoting properties of marine bioresources has propelled anchovies (*Stolephorus* spp.) into the spotlight as a promising candidate for functional food and nutraceutical development. Rich in bioactive peptides, omega-3 fatty acids, essential minerals, and antioxidant compounds, anchovies offer multifunctional health benefits that align with current consumer demands for natural, preventive, and therapeutic dietary products (Galasso et al. 2020a; Shaik and Sarbon 2022a).

Anchovies, with their high content of EPA and DHA, are versatile ingredients for a range of functional foods, which are defined as conventional food products that provide additional physiological benefits beyond basic nutrition. These bioactive compounds contribute to cardiovascular protection, neurodevelopment, and anti-inflammatory regulation, making anchovies a key ingredient in formulations aimed at reducing noncommunicable disease risk. The potential applications of anchovies in products such as anchovy-enriched spreads, soups, sauces, crackers, and extruded snacks have been explored, with promising results in terms of sensory properties and improved lipid profiles (Park et al. 2022).

Simultaneously, the development of nutraceuticals bioactive compounds delivered in concentrated form (e.g., capsules, tablets, powders) is rapidly gaining momentum. The increasing consumer demand for health-promoting products drives this trend. Anchovy protein hydrolysates, with their peptide-based bioactivities, are particularly attractive for this market. These bioactivities include antioxidant, antihypertensive, and glucose-regulatory effects. These hydrolysates can be obtained through enzymatic processing using food-grade proteases, yielding fractions with low allergenicity and high digestibility. Their potential applications include anti-fatigue supplements, blood sugar regulators, and muscle recovery agents, especially for aging and physically active populations (Chalamaiah et al. 2012).

Fish oils extracted from anchovies are already well-established in commercial omega-3 supplement markets. However, innovations now focus on microencapsulation

techniques, which improve oxidative stability, bioavailability, and controlled release of fatty acids in the gastrointestinal tract. Additionally, fermented anchovy products such as fish sauce and pastes offer natural alternatives with added value as probiotic carriers or mineral supplements. Anchovies also serve as a sustainable and scalable source of functional biomolecules, given their high reproductive rate and availability in tropical and subtropical marine waters. This makes them suitable for long-term supply in health-based product pipelines, potentially leading to economic benefits for the industry. Moreover, the low trophic level and short lifespan of anchovies reduce ecological risks and contamination compared to large pelagic species, offering a safer raw material base (Alfio et al. 2021).

Challenges in this domain include ensuring standardization, quality control, and the utmost regulatory compliance for anchovy-derived products. Variability in bioactive content due to species, habitat, and processing methods must be addressed through rigorous characterization and labeling. Collaboration between fisheries, food technologists, and health scientists is needed to fully exploit the functional potential of anchovies while ensuring safety and consumer confidence. As the global trend toward personalized nutrition and marine-based therapeutics accelerates, *Stolephorus* spp. stand out as a biocompatible, accessible, and multifunctional marine resource for the next generation of health-supporting products (Mutalipassi et al. 2024).

Technological innovations in extraction and processing

The valorization of anchovies (*Stolephorus* spp.) as a source of high-value bioactive compounds has driven significant progress in technological innovations related to extraction and processing methods. Advancements in this area are essential not only to increase yield and purity of target compounds but also to preserve their bioactivity, ensure safety, and enable their application in food, pharmaceutical, and nutraceutical industries (Shaik and Sarbon 2022a; Kan et al. 2025) (Table 16, Figure 13).

Conventional extraction techniques, such as solvent extraction or mechanical pressing, are limited by low

selectivity, thermal degradation, and the use of potentially toxic solvents. To overcome these limitations, modern techniques have been adopted, with enzymatic hydrolysis emerging as the most promising method for isolating bioactive peptides from anchovy proteins. This technique employs food-grade proteases (e.g., alcalase, papain, flavourzyme) under controlled pH and temperature conditions, allowing for the release of functional peptides with antioxidant, antihypertensive, and anti-diabetic properties while maintaining nutritional quality (Galasso et al. 2020a).

Further refinement is achieved through ultrafiltration and membrane fractionation, which enable size-based separation of peptide fractions (<3 kDa, 3-10 kDa, etc.), each with distinct bioactivities. These technologies enhance bioactive concentration, reduce allergenic potential, and allow for precise formulation of nutraceutical blends. In the lipid domain, Supercritical Fluid Extraction (SFE) using CO₂ has attracted attention as an environmentally friendly, solvent-free method for isolating omega-3-rich oils from anchovy tissues. SFE offers advantages in purity, oxidation resistance, and scalability, producing EPA/DHA-rich extracts suitable for encapsulation or direct inclusion in functional foods (Zhou and Wei 2023).

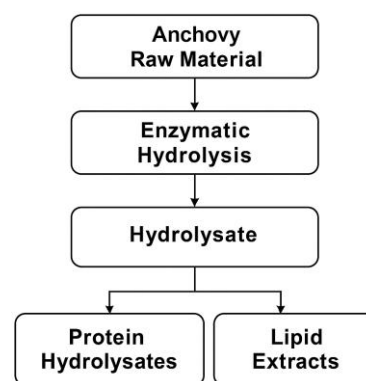


Figure 13. Processing flow of anchovy protein hydrolysates and lipid extracts

Table 16. Technological methods for anchovy bioactive extraction and their advantages

Method	Target compound	Key advantages	Limitations
Enzymatic hydrolysis	Bioactive peptides	High specificity, low temperature, retains bioactivity	Cost of enzymes, time-intensive
Ultrafiltration/membrane tech	Peptide fractionation	Size-selective separation improves bioactivity concentration	Membrane fouling, operational complexity
Supercritical CO ₂ extraction	Omega-3 fatty acids (EPA, DHA)	Solvent-free, high purity, low oxidation risk	High equipment cost, limited for hydrophilic
Spray drying (microencapsulation)	Omega-3s, peptides	Enhances stability, shelf life, and controlled release	May reduce yield or encapsulation efficiency
Controlled fermentation	Peptides, organic acids	Traditional method, natural flavor, microbial enzyme action	Variability in composition, long fermentation
Solvent extraction (conventional)	Total lipids, fatty acids	Simple and widely used	Risk of solvent residues, low selectivity

Note: Galasso et al. (2020); Shaik and Sarbon (2022); Sun et al. (2022)

Additionally, microencapsulation technologies such as spray drying, coacervation, and liposomal entrapment are increasingly being applied to protect sensitive fatty acids and peptides during storage and digestion, improving bioavailability and consumer acceptability. Fermentation-based approaches, traditionally used in Southeast Asian anchovy products like fish sauce and *bagoong*, have been optimized using starter cultures and controlled fermentation systems. These innovations improve consistency, enhance flavor profiles, and may enrich products with bioactive amino acids, peptides, and beneficial microbes. Moreover, integrated biorefinery models are being explored to utilize anchovy by-products such as heads, bones, and viscera for the recovery of gelatin, collagen peptides, enzymes (e.g., trypsin), and calcium-rich materials. These by-products can be processed into functional ingredients for the cosmetic, pharmaceutical, or animal feed industries, supporting circular economy principles and reducing waste (Perry and McClements 2020).

Emerging technologies such as Pulsed Electric Field (PEF) and Ultrasound-Assisted Extraction (UAE) are also under investigation to improve cell disruption and mass transfer efficiency. These technologies could potentially reduce processing time and energy consumption while preserving thermolabile compounds. These innovations collectively contribute to the sustainable utilization of anchovies as a multifunctional marine resource, enabling the development of high-quality, bioactive-rich products for diverse markets. Ongoing research is needed to scale these technologies economically and to assess their environmental impacts, particularly in small-scale and artisanal fishery contexts where infrastructure remains limited (Figure 14).

Research gaps and recommended directions

Despite growing interest in the health benefits and bioactive potential of anchovies (*Stolephorus* spp.), several critical research gaps remain that limit their full scientific validation and commercial exploitation. One of the most pressing needs is for more in vivo or clinical trials to confirm the efficacy and safety of anchovy-derived compounds in human systems. This is crucial for translating laboratory findings into standardized functional food or pharmaceutical products, making the research highly relevant and necessary.

A second major gap concerns the species-specific characterization of bioactives. This is a crucial area of studies that fail to distinguish between different *Stolephorus* species, despite likely variation in biochemical profiles due to genetics, habitat, diet, and seasonal dynamics. Establishing detailed comparative analyses across multiple species and geographic origins is not just essential for quality control, regulatory approval, and targeted product development, underscoring the significance and impact of the research.

The mechanistic understanding of how specific anchovy-derived compounds exert their biological effects particularly antioxidant, anti-inflammatory, anti-diabetic,

and anti-cancer actions also remains limited. While general pathways such as inhibition of oxidative stress or modulation of signaling cascades (e.g., NF- κ B, PPAR) have been proposed, more refined studies using molecular docking, omics-based platforms (proteomics, metabolomics), and systems biology are needed to clarify compound-target interactions.

Additionally, extraction, hydrolysis, and bioactivity testing protocols lack standardization, making cross-study comparison difficult. The development of validated methods for measuring peptide composition, antioxidant activity, enzymatic inhibition, and toxicity is critical for building a coherent body of evidence and ensuring replicability. Environmental and safety assessments also warrant further investigation. Anchovies are vulnerable to heavy metal and microplastic contamination in polluted coastal waters, yet few studies assess the bioaccessibility, accumulation, and toxicokinetics of such contaminants in processed anchovy products. Integrated approaches combining food science with environmental toxicology will be necessary to ensure consumer safety and guide source selection.

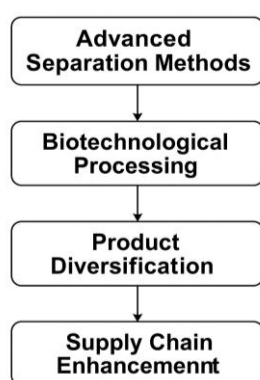
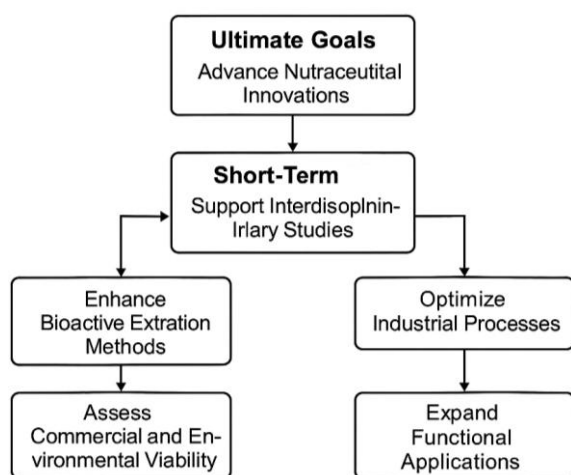
Moreover, the valorization of anchovy processing by-products (e.g., heads, viscera, bones) for bioactive extraction remains underexplored. If processing technologies and market frameworks are adequately developed, these fractions could provide low-cost, sustainable inputs for gelatin, collagen, mineral, or enzyme production, supporting circular bioeconomy goals. Lastly, the integration of local and indigenous knowledge, particularly from regions with long traditions of anchovy use (e.g., Southeast Asia, the Mediterranean), offers valuable insights into preparation methods, consumption patterns, and ethnonutritional relevance areas currently underrepresented in scientific literature. To address these gaps, future research should prioritize clinical trials that validate the health claims of anchovy-derived nutraceuticals, alongside species-specific chemoprofiling using multi-omics platforms to capture biochemical diversity across different *Stolephorus* taxa.

Emphasis should also be placed on mechanistic studies that elucidate the links between molecular structure and physiological function, as well as the development of standardized, scalable, and environmentally friendly extraction methods to ensure consistency and sustainability. In parallel, environmental monitoring and toxicological assessments are needed to evaluate contamination risks associated with heavy metals and microplastics in anchovy-based products.

Finally, greater attention should be given to the valorization of by-products and integration of traditional ecological knowledge, particularly in regions with long-standing cultural uses of anchovies. Collectively, these research directions will enhance the scientific credibility, safety, and real-world applicability of anchovies as a strategic marine bioresource for sustainable nutrition and health promotion (Table 17, Figure 15).

Table 17. Summary of research gaps and proposed scientific directions

Identified research gap	Recommended scientific direction	Expected impact
Lack of clinical validation of health claims	Conduct human trials on anchovy-derived nutraceuticals	Evidence-based support for therapeutic applications
Limited chemoprofiling at the species level	Apply omics-based techniques (e.g., proteomics, lipidomics, metabolomics)	Improved specificity and bioactive standardization
Poor understanding of molecular mechanisms	Perform mechanistic studies linking compound structure to physiological effect	Rational design of functional formulations
Non-standardized extraction and processing methods	Develop green, scalable, and standardized protocols	Increased industrial applicability and reproducibility
Potential environmental contamination in anchovy products	Integrate contaminant monitoring and risk assessment in product development	Enhanced safety and consumer confidence
Underutilized anchovy by-products and traditional knowledge	Explore bioactive recovery from waste and local processing wisdom	Circular bioeconomy and cultural value preservation

**Figure 14.** Technological innovations in anchovy extraction and the value chain development scheme**Figure 15.** Research roadmap for future development of anchovy-based bioresources

CONCLUDING REMARKS

Anchovies (*Stolephorus* spp.) represent a highly valuable marine resource at the intersection of biodiversity, nutrition, and biomedicine. This review has highlighted the broad spectrum of zoochemical compounds present in

anchovies including omega-3 fatty acids, bioactive peptides, essential minerals, and functional proteins—that contribute to various biological activities such as antioxidant, antimicrobial, anti-inflammatory, metabolic, and anti-cancer effects. These bioactives exhibit promising potential for development into functional foods, nutraceuticals, and pharmaceutical agents, supported by emerging evidence from both in vitro and in vivo studies. Ecologically, anchovies play a pivotal role in marine food webs, yet they are increasingly threatened by overexploitation, habitat degradation, and contamination from heavy metals and microplastics. These environmental challenges necessitate integrated strategies for sustainable harvesting, technological innovation, and contamination risk mitigation.

Advances in enzymatic extraction, microencapsulation, and biorefinery approaches have begun to unlock the commercial and therapeutic potential of anchovy-derived products, although further standardization and validation are urgently needed. Despite significant progress, key research gaps remain, particularly regarding species-specific biochemical profiling, mechanistic elucidation, clinical efficacy, and safe consumption thresholds. Addressing these issues through interdisciplinary research will strengthen the foundation for future applications and policy development. Anchovies, due to their global availability, nutritional richness, and functional versatility, are uniquely positioned to contribute to marine-based solutions for health, sustainability, and food security in the coming decades.

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