# Different Gelatins Used in the Encapsulation of Fish Oil

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## Abstract

Fish oil, containing omega-3 polyunsaturated fatty acids such as eicosapentaenoic acid and docosahexaenoic acid, has attracted considerable interest for its health advantages, such as reducing inflammation and decreasing triglyceride levels. Nonetheless, fish oil is highly susceptible to oxidation, resulting in rancidity, reduced effectiveness, and unwanted smells, creating difficulties in its application as a dietary supplement. Using gelatin, encapsulation is commonly used to safeguard fish oil from oxidation and enhance its stability. This method involves enveloping the oil in a biopolymer to create a shield that guards against elements such as oxygen and light. Gelatin, obtained from different animal origins, is important in encapsulation. Various forms of gelatin, such as porcine skin gelatin (PSG), bovine skin gelatin (BSG), and fish gelatin (FG), have different characteristics that impact encapsulation efficiency and the stability of the enclosed fish oil. PSG and BSG are well-liked because they are readily accessible, cost-efficient, and have outstanding gel-forming capabilities. Nevertheless, their use may be restricted in specific markets due to dietary and religious guidelines. FG, obtained from fish skin and bones, is a better option that meets these limitations and fits well with human biology. A thorough search plan was created to locate pertinent papers from electronic databases such as PubMed, Google Scholar, Web of Science, or Scopus. This review examines the utilization of various kinds of gelatin to encapsulate fish oil, studying how their physical and chemical properties disturb the stability and efficacy of the encapsulated fish oil. The research also emphasizes the difficulties presented by oxidation and the significance of encapsulation in preserving the nutritional value and longevity of fish oil supplements. This review aims to offer insights into using different types of gelatins for encapsulating fish oil to enhance its oxidative stability and health advantages through a comparative analysis.

Key words: Encapsulation, fish gelatin, fish oil, gelatin, Omega 3 fatty acid

## INTRODUCTION

sish oil had been traditionally consumed to ensure sufficient Vitamin A and D intake; for many years, these micronutrients have been recognized as essential for the body's normal functions (homeostasis). It was not before the well-known publication by Dyerberg and Bang and their work on the Inuit diet in the mid-1970s that the interest and awareness of the significance of marine omega-3 polyunsaturated fatty acids (PUFAs) in human health increased.[1] Fish and other sea creatures are abundant in a distinct type of polyunsaturated fats called omega-3 or n-3 fatty acids. Adding omega-3 fatty acids to the diet lowers triglyceride levels, an effect pronounced in those with marked hypertriglyceridemia.<sup>[2]</sup> Omega 3, a PUFA, provides numerous health advantages. Omega 3 is a vital nutrient necessary for the proper functioning of the brain, cardiovascular system, and growth development system.<sup>[3]</sup> Omega 3 can decrease blood pressure, lower triglycerides in the blood, decrease joint inflammation in rheumatoid disease, support brain and eye functions, aid in preventing dementia, depression, asthma, migraine, and diabetes, and reduce the risk of heart disease and ischemic stroke. Foods such as marine fish, walnuts, soybeans, flax seed oil, and canola oil are high in Omega 3.<sup>[4]</sup> Fish oil is a significant dietary source of PUFAs, such as eicosapentaenoic

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**Received:** 13-10-2024 **Revised:** 14-12-2024 **Accepted:** 22-12-2024 acid (EPA) and docosahexaenoic acid (DHA), and is known for its strong biological activities and health benefits. The complete PUFA composition of fish oils is influenced by various factors such as the type of fish, gender, stage of development, food intake, and surroundings, which could affect their possible advantages for health.<sup>[5]</sup> The benefits of fish oil seem to come from its omega-3 fatty acid content. Mackerel, herring, tuna, and salmon are examples of fish rich in these oils. The body doesn't produce many omega-3 fatty acids on its own. Omega-3 fatty acids decrease pain and inflammation while also inhibiting excessive blood clotting.<sup>[6]</sup> To improve the oral absorption of fish oil and test its antiinflammatory effect, a fish oil nanoemulsion was developed using cis-4,7,10,13,16,19-docosahexaenoic fatty acid as a biomarker for oral administration.<sup>[7]</sup> Consistent consumption of adequate amounts of PUFAs is essential in decreasing the occurrences of various illnesses and chronic conditions such as psoriasis, bowel diseases, mental disorders, cancer, rheumatoid arthritis, heart diseases, diabetes, respiratory disorders, coordination issues, movement disorders, obesity, and fragile bones. As a result, health agencies around the globe strongly encourage the consumption of foods rich in PUFAs.<sup>[5]</sup> While the European Food Safety Authority Panel on Dietetic Products, Nutrition, and Allergies recommends consuming 250-500 mg/day of EPA and DHA for adults with cardiovascular risk, scientific authorities recommend the consumption of fish twice a week to benefit from the nutritional effects of omega-3 fatty acids.[8] Aside from the distinct EPA and DHA composition found in fish oils, omega-3 fatty acids are the most vulnerable to oxidation. Once the fish oil supplement bottles are opened, they are subjected to oxidation, beginning with oxygen in the air, leading to rancidity and deterioration issues. Fish oil is highly susceptible to oxidation, which leads to degradation, reduced efficacy, and the formation of unpleasant odors and tastes.<sup>[8]</sup> Oxidation significantly affects the nutritional value of oils. The formation of harmful compounds such as 4-hydroxy-2-alkenal and 4-hydroxy-2-hexanalcan negatively impact health. In addition, oxidation causes unpleasant odors and tastes, shortens shelf life, and reduces the content of essential fatty acids.<sup>[9]</sup> To ensure optimal nutritional benefits and maintain acceptable quality, omega-3 fatty acid oils must have good sensory properties and strong oxidation resistance. This instability presents challenges for the long-term storage and delivery of nutritional supplements.<sup>[5]</sup> Encapsulation is a method that entails enclosing a substance or a blend of materials within another material to shield delicate functional materials from undesirable conditions. The trapped substance is referred to as the core material or active agent, whereas the material that encloses it is called the shell or coating material.[10]

The end-product of this technique is known as core-shell microcapsules or microspheres, which have a round shape and a variety of sizes of about  $0.1-10 \,\mu\text{m}$ . Encapsulation can be utilized in various applications, including the encapsulation of food ingredients such as flavors and

vitamins. It can be achieved through spray drying, fluid-bed coating, freeze-drying, and emulsification.[11] Encapsulation is a promising solution to protect fish oil from oxidation and improve its stability. Compared to single-component encapsulation materials, protective coatings formed by combining polysaccharides and proteins can improve products' encapsulation efficiency (EE) and oxidative stability. This combination of proteins and polysaccharides to encapsulate PUFAs can either be used as a raw mixture or undergo a Maillard reaction to form a complex with enhanced properties.<sup>[12]</sup> The encapsulation process involves creating a protective barrier around the oil, often using a biopolymer, to shield it from environmental factors such as oxygen and light. Among the biopolymers used for encapsulation, gelatin is widely preferred due to its versatility and functional properties.[13]

# THE ROLE OF GELATIN IN ENCAPSULATION

Fish oils, rich in omega-3 PUFAs, offer numerous health benefits for human healthcare and nutrition. However, their use has traditionally been hindered by challenges such as a fishy odor and taste, poor water solubility, and susceptibility to oxidation. In response, fish oil encapsulation preparations have been developed to eliminate these disadvantages, leading fish oil encapsulation to become a sought-after research topic in the field of food science.[14] Its many uses in the pharmaceutical sector are as an encapsulating and suspending agent. Gelatin has good advantages in the complex coacervation method for encapsulating oil because it can interact with oppositely charged molecules, has varying pH-dependent charge density, adequate charge density in the chains, and distinct gelling behaviors.<sup>[15]</sup> The oil powders were produced through a combination technique involving the coacervation of gelatin and sodium hexametaphosphate, with the assistance of starch sodium octenyl succinate (SSOS) during freeze-drying.<sup>[16]</sup> In contrast to other gelatins, cold-water fish skin gelatin (CFG), which has an unclear isoelectric point, a smaller molecular weight, increased hydrogen bonds, and an extended gel formation time, cannot create complex coacervates essential for the production of oil powders. For oil powders derived from different gelatins, the type and concentration of gelatin did not show pronounced impacts on small-scale structures but did significantly affect physicochemical characteristics.<sup>[17]</sup> Earlier research has utilized this technique to encapsulate fish oil by using the electrostatic effect between gelatin and acacia gum, SHMP, anionic gum Arabic, and almond gum. Nevertheless, the products' encapsulation efficiencies were typically <90%. We have recently created freeze-dried fish oil powders with over 95.2% fish oil encapsulation effectiveness by the coacervation of gelatin and SHMP complex and SSOS as a drying aid.<sup>[18]</sup> Although our research indicated promising potential for utilizing fish oil preparations, the specific impact of gelatin type and concentration on fish oil powders' properties is still poorly understood. The amino acid composition of gelatin, which is similar to its parent collagen, is influenced by the species of animal and the type of tissue. Differences also influenced variations in chemical properties in molecular weight distribution due to variations in conditions of nature or extraction. Gelatins derived from cow and pig skin are frequently utilized in food industry production due to their abundant availability.<sup>[15]</sup>

# TYPES OF GELATIN USED IN ENCAPSULATED FISH OIL

The kinds of gelatin used to encapsulate fish oil include porcine skin gelatin (PSG), bovine skin gelatin (BSG), fish gelatin (FG), and CFG.<sup>[19]</sup> Gelatins, versatile materials with applications in fields such as medical tissue engineering, drug delivery, cosmetics, and food science, are derived from various animal tissues such as bones and skin. The specific source (e.g., mammalian, poultry, or fish) and extraction method significantly impact the functional characteristics of gelatins. Therefore, it is essential to investigate the effects of gelatin type on the preparation and properties of gelatinbased products.<sup>[14]</sup> The specific advantages and limitations of these gelatin types are summarized in Table 1.

#### PSG

PSG is made from the skin of pigs. Gelatin is a heterogeneous mixture of water-soluble proteins of high average molecular mass in collagen.<sup>[20]</sup> PSG produced from acidic treatment is known as type A gelatin.<sup>[21]</sup> Gelatin dissolves more readily in hot water than in cold water. It is practically insoluble in most organic solvents such as alcohol, chloroform, carbon disulfide, carbon tetrachloride, ether, benzene, acetone, and oils.<sup>[22]</sup> It serves as a protective coating that helps stabilize the fish oil by preventing oxidation, preserving its nutritional quality, and controlling the release of the oil.<sup>[23]</sup> Porcine gelatin is often chosen for fish oil encapsulation because of its excellent gelling properties, clarity, and availability, making it a cost-effective and efficient option. However, its use may be restricted due to dietary or religious concerns, prompting some manufacturers to use alternatives such as bovine or FG in certain markets.<sup>[24]</sup>

#### BSG

A protein-based gelling agent known as bovine gelatin. It is created by partially hydrolyzing collagen, a protein extracted from animal tissues such as bone and skin. The gelatin molecule is made up of amino acids joined together by amide linkages in a long molecular chain.<sup>[25]</sup> Gelatin is a combination of numerous large polypeptides that originate from the controlled breakdown of tough, insoluble collagen found in various parts of animals, such as bones, hides, skins, tendons, and sinews. Due to its distinctive technical and functional properties, gelatin is widely used in the catering, pharmaceutical, medical, cosmetic, and photographic sectors for thickening, clarifying, emulsifying, and gelling purposes. Gelatin from bovine skin produced from alkaline treatment is known as type B gelatin.<sup>[26]</sup> Recent reports indicate that the main sources of gelatin are pigskin (46%), bovine hides (29.4%), bovine bones (23.1%), and other sources (1.5%).<sup>[27]</sup> Its thermo-reversible nature allows it to be melted and set repeatedly, which is advantageous in encapsulation processes involving heat.<sup>[28]</sup> The effects of adding BSG hydrolysate obtained with subtilisin on water-holding capacity in a thermally processed chicken meat model were investigated.<sup>[29]</sup> In addition, BSG is generally straightforward, facilitating visual inspection of the encapsulated product, and it is costeffective due to its widespread availability.<sup>[30]</sup>

FG

It is often recognized that fish are a great source of protein for humans. The edible part of fish is mostly meat (muscle); thus, fish processing industries produce large quantities of inedible parts or wastes such as skin, fins, bones, scales, and viscera.<sup>[31]</sup> FG is a protein substance obtained from fish skin or scales high in collagen, using hot water extraction. The Gelatin molecule consists of amino acids connected by amide linkages in a lengthy molecular chain. These amino acids perform an imperative function in the building of connective tissue in humans.<sup>[32]</sup> Fish skin and bone waste is one alternative source of raw material for making gelatin.<sup>[33]</sup> Fish oil is encapsulated using various methods, including complex coacervation, spray-drying, freeze-drying, and electrospraying. Complex coacervation is a popular method that uses gelatins as wall materials.<sup>[34]</sup> FG soft capsules can be made from a mixture of FG, glycerol, and water. The composition of amino acids in FG is different from traditional gelatin and is better suited to the human body.<sup>[35]</sup>

#### CFG

Gelatin extracted from cold water fish skin, such as that of Alaska pollock, possesses low-temperature gelling characteristics, often forming a gel at temperatures below 11.9°C.<sup>[36]</sup> CFG has a lower melting point than other gelatins used in colder environments but is less stable at higher temperatures. This characteristic allows the rapid discharge of encapsulated materials, which may offer advantages in fields such as pharmaceutical formulations or food technology.<sup>[37]</sup> The utilization of CFG in microencapsulation of vitamins, colorants, and flavoring agents such as lemon, garlic, black pepper, and other flavors is made possible by the low melting temperatures.<sup>[15]</sup> There were variations in the physical and rheological properties and water vapor permeability between CFGs and mammalian gelatins.<sup>[38]</sup> Like other gelatins, CFG can encapsulate fish oil to protect it from oxidation, thereby extending shelf life and preserving the nutritional value of omega-3 fatty acids.<sup>[5]</sup>

#### Vegetable-based gelatin alternatives

Vegetable-based gelatin alternatives are popular among consumers looking for plant-based, ethical, and healthconscious substitutes.<sup>[39]</sup> One of the most widely used alternatives is agar-agar, derived from red algae. Agar-agar is perfect for sweets, jellies, and soups since it has no flavor and no smell, and it forms a hard, brittle gel.<sup>[40]</sup> Another option is carrageenan, sourced from red seaweed, serving as a thickener and stabilizer in different products, particularly dairy and plant-based foods.<sup>[41]</sup> Pectin, sourced from fruits such as apples and citrus, is commonly used in jams and jellies due to its ability to form a gel when combined with sugar and acid.<sup>[42]</sup> Kudzu root starch, derived from the kudzu plant, also serves as a thickening agent in sauces and desserts, offering digestive benefits.<sup>[43]</sup> Cornstarch, derived from corn, is a gluten-free alternative that helps to thicken soups, sauces, and custards. These options offer a variety of healthy choices for those following vegan, vegetarian, and allergy-friendly diets.[44]

### **COMPARATIVE ANALYSIS**

Recently, there has been a growing need for fish oil in the pharmaceutical sector, attributed to discovering the health advantages of omega-3 fatty acids, specifically EPA and DHA. The composition of fish oils is distinct because they contain a wide range of fatty acids, making them a valuable source of C20 and C22 omega-3 fatty acids with significant medical benefits.<sup>[45]</sup> Previously, fish oil was primarily utilized as a crucial component in aquaculture diets and also in diets for livestock such as poultry and swine. Because of the growing recognition of the possible benefits of omega-3 fatty acids for human health, there has been a rising curiosity about using fish oils for human consumption.<sup>[46]</sup> The attractive qualities of hardened fish oils in the human diet are due to their nutritional and physical properties. Over the last ten years, a variety of food items have been introduced, including fish oils. The major challenges in adding long-chain omega-3 oils to food products include the instability of EPA and DHA and the presence of a fishy odor.<sup>[5]</sup> A market for dietary supplements is well-established, and there is also a growing market for food ingredients made from fish oils and fish oil concentrates. The ever-increasing body of scientific research supporting the health benefits of fish oil has resulted in a significant increase in its utilization within the pharmaceutical industry, with expectations for a prominent role in the future.<sup>[47]</sup>

## PHYSICAL PROPERTIES

The qualities of both their physical and chemical nature determine how they act and react in different situations. These characteristics are vital in numerous manufacturing industries, such as food, chemical, pharmaceutical, and other fields.<sup>[48]</sup> Some physical properties to consider are boiling point, flash point, ignition point, melting point, solidification point, viscosity, plasticity, refractive index, specific gravity, and solubility. Inability to be saponified, capacity for emulsions, and flexibility.<sup>[49]</sup> The boiling point is the temperature of the oil sample's liquid form vapor pressure, which equals the pressure in the environment. An example becomes a gas when heated. The flash point is the temperature at which an oil sample will ignite when exposed to specific heating conditions.<sup>[50]</sup> The flame is moved across the oil's surface, but it will not keep burning because the ignition point is the temperature when oil will keep burning without extra heat. The melting point is when an oil sample goes from solid to liquid, whereas the solidification point is when the liquid phase is in equilibrium with a small solid phase portion.<sup>[51]</sup> Factors such as free fatty acid (FFA), oxidation, and heat treatment can influence the refractive index, a numerical value indicating the degree of light speed ratio in a vacuum compared to a test substance. Specific gravity is calculated by comparing the mass of a particular amount of sample material to the weight of an equal volume of water, both measured at specific temperatures, to assess relative density.<sup>[52]</sup> The term unsaponifiable denotes the part of the oil that remains unconverted into soap when treated with potassium hydroxide. This segment usually contains sterols, tocopherols, hydrocarbons, and pigments. The emulsifying capacity is the capacity in the water/oil interface allowing the formation of emulsion, whereas the plasticity is the property that has a body to preserve its shape by resisting a certain pressure.[53]

## **CHEMICAL PROPERTIES**

Assessment of moisture, contaminants, acidity, FFAs, saponification, iodine, thiobarbituric acid, peroxide, anisidine, total oxidation, color, mineral and heavy metal content, and other characteristics are all part of chemical characterization. AcV represents the amount of potassium hydroxide needed to neutralize organic acids in one gram of fat, indicating the level of FFAs present.<sup>[54]</sup> An elevated level of FFAs in an oil or fat sample suggests. The decomposition of triglycerides through hydrolysis. SaV is the amount of potassium hydroxide needed to saponify one gram of fat in specific circumstances. It represents the mean molecular weight (or size of the chain) of the fatty acids found in the sample as triglycerides.<sup>[55]</sup> The greater the SaV, the smaller the average fatty acid length, and the lower the mean triglyceride molecular weight, and vice versa. IiV quantifies the degree of unsaturation in oil and fat by indicating the amount of iodine taken in by 100 g of the substance. The greater the IiV, the higher the amount of unsaturation found in the fat.<sup>[56]</sup> TBARS are generated due to lipid peroxidation the breakdown of fats and can be identified through the TBARS test with thiobarbituric acid as a reagent. PeVmeasures the reactive oxygen levels indicated in the meq of the amount of free iodine per kilogram of fat. It assesses how much a sample of oil has experienced initial oxidation;

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Table 1: Comparison of different types of gelatins used in fish oil encapsulation				
Types of gelatins	Source	Properties	Advantages	Limitation
1. Porcine skin gelatin (PSG)	Pig skin	Type A gelatin (acidic treatment). Good gelling properties. Soluble in hot water.	Cost-effective. Excellent gel formation and clarity.	Restricted in some markets due to dietary and religious concerns.
2. Bovine skin gelatin (BSG)	Cow skin	Type B gelatin (alkaline treatment). Thermo-reversible. Strong water-holding capacity.	Widely available. Cost-effective. Clear and stable.	Similar to PSG in terms of limitations (dietary restrictions).
3. Fish gelatin	Fish skin and bones	Different amino acid composition from mammalian gelatin. Good solubility at lower temperatures.	Suitable for those with dietary restrictions. More compatible with human biology.	Weaker gel strength compared to mammalian gelatins. Higher cost.
4. Cold-water fish Gelatin (CFG)	Cold-water fish skin (e.g., Alaska pollock).	Low-temperature gelling, gel formation below 11.9°C, lower melting point, less stable at high temps.	It protects encapsulated materials from oxidation, extends the shelf life of omega-3 fatty acids, and facilitates the rapid discharge of materials.	Less stable at higher temperatures and may not be suitable for hot environments.
5. Vegetable-Based Gelatin Alternatives	Plant-based sources (e.g., red algae, fruits, roots).	Agar-agar (hard, brittle gel), carrageenan (thickener, stabilizer), pectin (gel with sugar/acid), kudzu root starch (thickener), cornstarch (gluten-free thickener).	Plant-based, ethical, vegan-friendly, suitable for health-conscious and allergy-friendly diets.	Compared to animal-based gelatins, different gelling properties may not replicate all functional aspects of traditional gelatin.

the level of secondary oxidation can be calculated through the panisidine test. Because it can identify unsaturated aldehydes, which are more likely to yield unsatisfactory findings, it is instrumental in food safety monitoring.<sup>[57]</sup>

# **CONCLUSION**

Fish oil containing high levels of omega-3 PUFAs offers numerous health benefits, particularly in reducing inflammation and managing cardiovascular risks. However, its high susceptibility to oxidation presents challenges related to reduced efficacy, unpleasant odors, and decreased shelf life. Encapsulation has emerged as a promising solution to protect fish oil from oxidation and improve its stability and bioavailability. Gelatin, a versatile biopolymer derived from various animal sources, plays a significant role in encapsulation. Different kinds of gelatin-PSG, BSG, and FG—are commonly used to encapsulate fish oil, each offering unique properties. PSG is favored for its excellent gelling properties and cost-effectiveness, whereas BSG offers thermoreversible advantages and is widely available. FG, derived from fish skin and bones, is a suitable alternative, especially for dietary and religious restrictions. It is considered more compatible with the human body due to its unique amino acid

composition. The choice of gelatin for encapsulating fish oil depends on factors such as EE, sensory quality, and oxidation stability. While porcine and bovine gelatins are more widely used due to their availability and functional properties, FG is becoming a viable alternative, particularly in markets with specific dietary requirements. The ongoing development of encapsulation techniques continues to improve the shelf life and sensory attributes of fish oil products, ensuring the preservation of its health benefits for consumers.

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