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Preparation of this document

Although aquatic food is a crucial source of high-quality proteins and nutrients and plays a vital role in the global food supply system, it also yields significant by-products during processing. These by-products are often deposed of as “waste” or used as low-value animal feeds, leading to environmental problems or resource waste. In fact, the by-products of aquatic food are rich in diverse bioactive ingredients and can serve as valuable and important raw materials for high-value products in many industries. The preparation of this document was motivated by a growing need for the treatment of massive quantities of by-products of aquatic food processing worldwide. The publication guides the production of high-value items, from the selection of raw material to the application of production technology, facilitating the transformation of the by-products of aquatic food processing and promoting their high-value utilization within the aquatic food system for improved human nutrition, health, environmental sustainability and global economic well-being.

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Abstract

The processing of aquatic food generates substantial by-products, including animal heads, skins, bones, scales, visceral organs and shells, etc., which can constitute between 30 percent and 70 percent of the whole body of aquatic organisms. These by-products retain numerous bioactive molecules suitable for extraction and application in the nutraceutical, functional food, pharmaceutical, biomedical, cosmetic and material industries, and have the potential to yield high-value products. The transition from aquatic food waste to high-value products presents multiple benefits, including: (i) enhanced human nutrition and health through nutrient and bioactive component provision; (ii) mitigation of environmental pollution by reducing waste; and (iii) improved economic returns because aquatic food waste is transformed into high-value products rather than low-value animal feeds or fertilizers.

This guide presents strategic and technical insights by outlining key principles for producing high-value items, including collagen, gelatine, bioactive peptides, chitin, chitosan, chondroitin sulphate, fish leather and fish oil, from the by-products of aquatic food processing.

Contents

Figures

Abbreviations

$(NH_4)_2SO_4$	ammonium sulphate
BSE	bovine spongiform encephalopathy
$Ca(OH)_2$	calcium hydroxide
$CaCO_3$	calcium carbonate
CO_2	carbon dioxide
CS	chondroitin sulphate
DH	degree of hydrolysis
DHA	docosahexaenoic acid
ECM	extracellular matrix
EDTA	ethylenediaminetetraacetic acid
EPA	eicosapentaenoic acid
FAO	Food and Agriculture Organization of the United Nations
FMD	foot-and-mouth disease
GAG	glycosaminoglycan
GalNAc	N-acetyl-β-D-galactosamine
GlcA	β-D-glucuronic acid
H_2O_2	hydrogen peroxide
H_2SO_4	sulfuric acid
H_3PO_4	phosphoric acid
HA	hyaluronic acid
HCl	hydrochloric acid
HOAC	acetic acid
IduA	iduronic acid
$KMnO_4$	potassium permanganate
KOH	potassium hydroxide
LAB	lactic acid bacteria
LAC	lactic acid
MAE	microwave-assisted extraction
n-3 PUFAs	omega-3 polyunsaturated fatty acids
Na_2CO_3	sodium carbonate
Na2S	sodium sulphide
NaCl	sodium chloride
$NaClO_2$	sodium chlorite
NaOH	sodium hydroxide
NH_4Cl	ammonium chloride
PCBs	polychlorinated biphenyls
pH	potential of hydrogen (an indication of acidity)
pI	isoelectric point
RSM	response surface methodology
SFE	supercritical fluid extraction
SFE-CO_2	SFE with carbon dioxide
UAE	ultrasound-assisted extraction

Introduction

In 2020, 178 million tonnes of aquatic animal food with a value of approximately USD 406 billion were produced globally alongside 36 million tonnes of algae. The top species were finfish (124 million tonnes), algae (36 million tonnes), molluscs (24 million tonnes) and crustaceans (17 million tonnes) (FAO, 2022).

Aquatic food is rich in proteins and polysaccharides and is a crucial source of nutrients for over 3.1 billion people, contributing 20 percent to their average per capita animal protein consumption (FAO, 2016). However, the edible portions of aquatic food constitute only a small part of harvested resources. The by-products of aquatic animal food processing represent between 30 percent and 70 percent of whole animals and amount to at least 50 million tonnes annually. Unfortunately, their valorization is inadequate. Some are used for low-value products such as fishmeal, fertilizer, fish silage and fish sauce, or they are discarded in the sea, rivers or landfills, leading to environmental pollution (Li and Li, 2023; Nirmal *et al.*, 2020; Amiri *et al.*, 2022; Suresh *et al.*, 2018).

Aquatic food offers high protein, low fat and diverse nutrients, presenting a healthy and safe source of food that is free from terrestrial animal viruses such as bovine spongiform encephalopathy (BSE), foot-and-mouth disease (FMD) and other prion diseases. By-products from aquatic food processing are rich in bioactive molecules, providing materials for high-value products such as collagen, gelatine, bioactive peptides, chitin, chitosan and chondroitin sulphate. Disposing of these by-products or using them solely to produce low value products is a significant waste of resources.

FIGURE 1. **Fish processing by-products**

This guide explores the classification, production, utilization and challenges of creating high-value products from the by-products of aquatic food processing. It emphasizes the imperative to maximize their value, support global food systems, generate economic profit and expand the aquatic product value chain through innovative technologies (Amiri *et al.*, 2022; Šimat, 2021).

2. Production and utilization of high-value products from the by-products of aquatic food processing

2.1 COLLAGEN

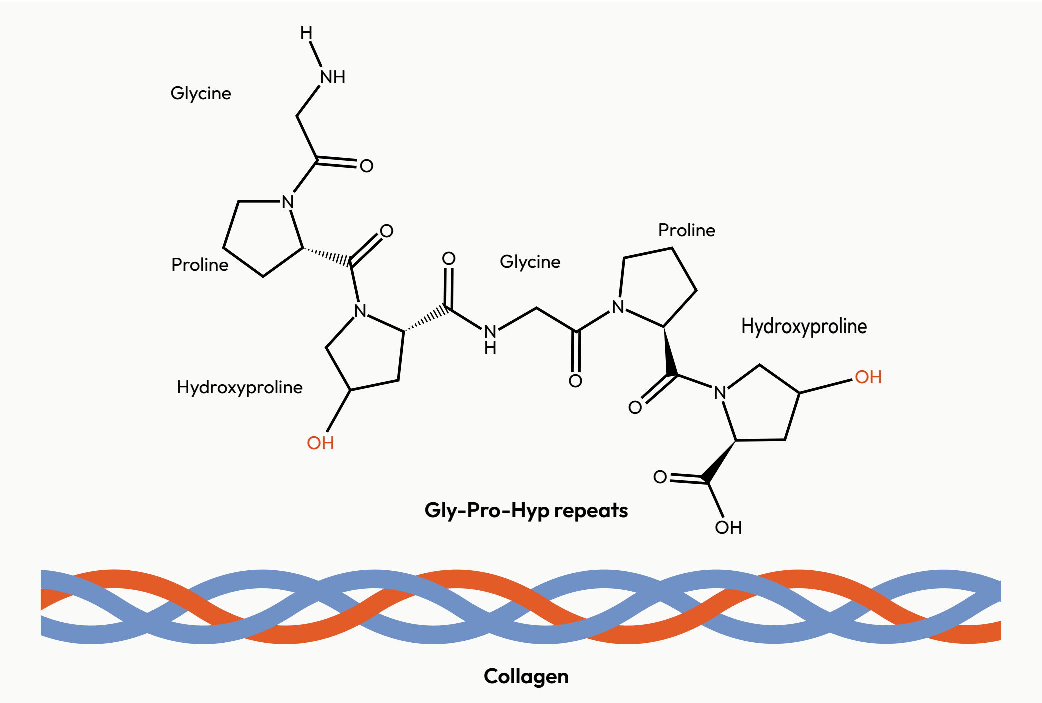
2.1.1 Product description

Collagen, with its significant nutritional, health and medicinal benefits, is widely used in the food, cosmetic, pharmaceutical, tissue engineering and biomedical industries. Exhibiting a triple helix structure with primary structure repeats of Gly-X-Y (where Gly is glycine, X is mostly proline and Y is hydroxyproline), collagen encompasses at least 29 identified types based on amino acid composition, sequence and structural properties (Liu *et al*., 2015).

Traditionally sourced from the skins and bones of terrestrial animals, collagen constitutes about 30 percent of total body proteins and serves a vital role in providing structure, strength, elasticity and support to skin, muscles, bones, tendons, ligaments and other connective tissues. However, a shift towards collagen derived from aquatic sources is gaining acceptance, particularly among religious consumers concerned about diseases such as BSE, FMD and other prion diseases in terrestrial animals (Salvatore *et al*., 2020).

The by-products of aquatic food processing, including fish skin, fins, scales, bones, and swim bladder, offer a rich source of collagen. Despite being discarded as "waste" or used for low-value fishmeal and fertilizers, these by-products present an opportunity to produce high-value collagen. Using collagen from the by-products of aquatic food processing not only adds value to these materials but also contributes to the reduction of environmental pollution.

Figure 2. Schematic diagram of the collagen structure



2.1.2 Raw material

The raw materials selected for commercial collagen production from the by-products of aquatic food processing need to be abundant in collagen and available in substantial quantities. Commonly used raw materials for collagen production include fish bones, fins, scales and swim bladders, and particularly fish skin. Type I collagen is predominant in collagens derived from fish processing by-products, although certain fish species may also contain minor amounts of type II, type V, or type XI collagen, which are different in amino acid composition and structure (Laasri *et al*., 2023).

2.1.3 How to make collagen from the by-products of aquatic food processing

Collagen sources, types, processing methods and extraction conditions are the main parameters that determine the properties of extracted collagen. Producing collagen from the by-products of aquatic food processing typically involves four key procedures: sorting and cleaning, raw material pretreatment, collagen extraction, and collagen recovery.

Sorting and cleaning

The by-products of aquatic food processing are rich in collagen and other proteins, lipids, polysaccharides and minerals. To obtain high-quality collagen from these by-products involves an initial step of minimizing other contaminations. The raw material from fish processing by-products undergoes sorting based on fish species and tissues. Non-collagen containing tissues are eliminated and washed thoroughly. Subsequently, the raw material is either cut or milled into small pieces. This enhances the efficiency of collagen extraction in the subsequent pretreatment phase.

Pretreatment of the material

The pretreatment procedure of the material for collagen production is quite different according to the types of raw material.
The pretreatment of fish skin and swim bladder:

1. Deproteinization with sodium hydroxide (NaOH);
2. Defatting with butanol or isopropanol, surface-active agent, sodium hydroxide; and
3. Washing the material for extraction.

The pretreatment of fish bone, scales, and fins which contain many mines like Ca_2+:

1. Deproteinization with sodium hydroxide;
2. Demineralization of Ca_2+ or inorganic materials with ethylenediaminetetraacetic acid (EDTA);
3. Defatting with butanol or isopropanol, surface-active agent, sodium hydroxide; and
4. Washing the material for extraction.

Extraction of collagen

The extraction of collagen from the by-products of aquatic food processing can be mainly divided into four types of methods: acid extraction, alkaline extraction, enzyme extraction, and other extraction techniques (Pal and Suresh, 2016). Typically, the extraction process is carried out at low temperatures (around 4 °C) to prevent collagen degradation.

1. Acid extraction. Acid is used as the common method for collagen extraction. Acetic acid (HOAC) is the most common method used to extract collagen and it produces the highest yield. However, citric acid, lactic acid (LAC) and hydrochloric acid (HCl) can also be used to extract collagen from the pretreated by-products.
2. Alkaline extraction. NaOH and sodium carbonate (Na_2CO_3) can also be used to extract collagen from the pretreated by-products. However, the quality of alkaline-extracted collagen is not as good as acid-extracted collagen or

enzyme-extracted collagen. This is because alkaline can break the triple helix structure of collagen.

3. Enzyme extraction. Enzymes are often used as an effective, and environmentally responsible method for the extraction of collagen. Various enzymes, such as pepsin, trypsin and alcalase can be used to extract collagen from the by-products of aquatic food processing. Pepsin is the most widely used enzyme for collagen extraction. Enzymes can be used alone or in combination with other enzymes and acids to maximize the collagen yield.
4. Other extraction. Supercritical fluid extraction (SFE) technology or ultrasound-assisted technology can also be used to extract collagen from the by-products of aquatic food processing. However, these methods require a high degree of investment in equipment or instrumentation.

Recovery of collagen

Posttreatment of collagen includes precipitation, dialysis and freeze-drying. The extracted collagen is precipitated by a high concentration of sodium chloride (NaCl) and centrifugation. The precipitated collagen is then dissolved in a small volume of HOAc and dialysed in distilled water. Finally, the dialysed collagen solution can be used directly or lyophilized with a freeze drier.

FIGURE 3. **Schematic diagram of collagen production using the by-products of aquatic food processing**

2.1.4 Utilization

Collagen can be widely applied in the food, cosmetic and pharmaceutical industries because of its remarkable attributes, such as a high capacity for water absorption, biocompatibility, low immunogenicity, biodegradability, high porosity, and the ability to penetrate a lipid-free interface, etc. (Pal and Suresh, 2016; Espinales *et al*., 2023).

Food industry

Collagen serves as a versatile food additive in various food processing applications because it enhances overall food quality. For instance, it acts as a binder and extender, improving the binding properties of hams during cold storage and cooking. Additionally, collagen functions as a texture improver and moisture emulsifier, minimizing pressing and thawing losses in hams and preserving the sensory properties of chicken patties. In sausage production, collagen serves as an edible casing, preventing the contact of oxygen and water vapour with sausages (Pal and Suresh, 2016).

Cosmetic industry

Collagen (type I) is one of the most important structural proteins in human skin and can provide structure, strength and elasticity to the skin. Collagen has been widely used in cosmetic products because of its moisturizing, regenerating and film-forming properties. Cosmetic products that include collagen are collagen creams, collagen moisturizers, collagen facial masks, collagen body lotions, collagen body oil, etc.

Pharmaceutical industry

Collagen has been used in the pharmaceutical industry because of its excellent biocompatibility, easy biodegradability and low immunogenicity. Collagen is used as a material to produce various clinical products for skin repair, wound healing, tissue repair, tissue regeneration and aesthetic contouring.

2.2 GELATINE

2.2.1 Product description

Gelatine, a transparent, flavourless and water-soluble protein mixture, results from the denaturation and hydrolysis of collagen. Despite sharing a primary structure with collagen and featuring repeats of the triplet Gly-X-Y, gelatine differs by being heat-denatured collagen with a disordered structure (Tang *et al*., 2022). Its unique temperature-sensitive hydrocolloid properties make gelatine widely utilized in the food, pharmaceutical, cosmetic and photographic industries.

Most commercial gelatine is currently derived from mammalian sources, such as pig skins, bovine hides and cattle bones, with only a minor percentage (~1 percent) sourced from fish (Rather *et al*., 2022). Commercial gelatine is categorized into three types: type A, produced through partial acid hydrolysis of collagen; type B, derived from alkali hydrolysis; and enzymatic gelatine, created through collagen enzymatic hydrolysis (Karim and Bhat, 2009).

Fish-derived gelatine carries no risk of zoonotic virus transmission and has no consumption limitations for religious reasons. The production of gelatine from the by-products of aquatic food processing not only maximizes the value of aquatic resources but also contributes to human health.

2.2.2 Raw material

Gelatine, being a hydrolyzed form of collagen, utilizes the same raw materials as the collagen produced from the by-products of aquatic food processing. Fish skin is a commonly used raw material in the industrial production of gelatine because it offers versatility. Fish bones, fins, scales and swim bladder can also serve as alternative raw materials for gelatine production. However, it is crucial to note that the quality of gelatine is significantly influenced by the fish species or tissues processed, as well as the processing methods selected.

2.2.3 How to make gelatine from the by-products of aquatic food processing

The production process of gelatine typically consists of four main procedures: sorting and cleaning, pretreatment of the raw material, extraction of the gelatine, and recovery of the gelatine. Usually, the production of different types of gelatine (type A or type B) depends on the treatment method of the collagen. Type A gelatine is pretreated with acid before the hydrolysis of gelatine with an isoelectric point (pI) of around 8.0 to 9.0. Type B gelatine, which is pretreated with alkaline, has a pI of around 4.0 to 5.0 (Babin and Dickinson, 2001).

Sorting and cleaning

The raw material of fish processing by-products is sorted based on different species and tissues. The contaminants are removed from the raw material and cleaned with water. The material is then cut or milled into small pieces to facilitate pretreatment and improve the efficiency of gelatine extraction.

Pretreatment of the material

The pretreatment of raw material for collagen production is a little different for different tissues. Production of collagen from fish skin and swim bladder can be conducted as follows, but the production of collagen from mineral-rich tissues such as fish bones, fins and scales require an additional decalcification with EDTA in the pre-treatment phase.

1. Acidic pretreatment. Acidic treatment is most suitable for the less covalently crosslinked collagens in the by-products of fish processing. Usually, fish skin is treated with acids such as HCl, sulfuric acid ($H2SO_4$), and phosphoric acid (H_3PO_4) to encourage adequate swelling and to disrupt the non-covalent bonds at a low temperature (4 °C to 10 °C) in a few hours (Karim and Bhat, 2009).
2. Alkaline pretreatment. Alkaline such as NaOH and calcium hydroxide [$Ca\ (OH)_2$] can also be used to treat fish processing by-products for collagen production. Alkaline can remove other non-collagenous materials and produce much purer gelatine. However, alkaline treatment for gelatine production usually takes a few days at a low temperature (4 °C to 10 °C), which takes much more time than acidic treatment (Karim and Bhat, 2009).
3. Enzyme pretreatment. In addition to the most commonly used chemical treatment of raw materials, enzyme treatment can be used as a potential alternative approach to produce gelatine from the by-products of fish processing. However, enzyme treatment has not been widely applied in the industrial production of gelatine from the by-products of fish processing for many reasons.

Extraction of gelatine

Heat denaturation and hydrolysis are the most common approaches to extract gelatine from pretreated materials. However, the water temperature for gelatine extraction from different sources varies substantially. Usually, the extraction temperature of fish gelatine is performed at temperatures ranging from 40 °C to 80 °C for 1 hour to 9 hours, with a common temperature of 45 °C overnight (Karim and Bhat, 2009).

Recovery of gelatine

Gelatine is usually used in solid or powdered form, which is easy to store and use. After the filtration and concentration of the gelatine extraction, the gelatine is recovered by drying, through processes such as freeze drying and hot air drying.

FIGURE 4. **Schematic diagram of gelatine production using by-products of aquatic food processing**

2.2.4 Utilization

Owing to its exceptional properties, such as water-binding, gel formation, film formation, foam creation and emulsification, gelatine is extensively used in the food, pharmaceutical, cosmetic and photographic industries (Alafaro *et al.*, 2015). Warm water fish gelatine is widely used across various sectors, in the same way that mammalian gelatines are. However, gelatine derived from cold water fish has more limited applications because of its diminished gelling power (Alfaro *et al.*, 2015). While fish gelatine currently contributes only 1 percent to global gelatine production, it holds immense promise as an alternative to mammalian gelatines, especially because its production would allow for the high-value utilization of fish processing by-products. The following examples illustrate the diverse applications of fish gelatine.

Food industry

Gelatine serves as a valuable additive for enhancing the consistency, elasticity and stability of various food products, including sweets, edible films, encapsulation, yogurt, acid milk gels and the clarification of fruit juices (Huang *et al.*, 2019).

Pharmaceutical industry

Gelatine finds widespread use in pharmaceutical applications, contributing to the production of capsules, ointments, cosmetics, tablet coatings and emulsions. It is also employed in the creation of microencapsulated health foods and dried products such as vitamins and pharmaceutical additives. Fish gelatine, specifically, holds potential for application in tissue engineering and wound healing (Lv *et al*., 2019).

Photographic industry

Gelatine plays a crucial role in the photographic industry, serving as a key component in the production of various photographic films. Its unique attributes, acting as a gelling agent and surfactant, enable the suspension of particles such as silver chloride or light-sensitive dyes (Alfaro *et al*., 2015).

2.3 PEPTIDES

2.3.1 Product description

A peptide refers to a short chain composed of two or more amino acids. Peptides are named according to the number of amino acid molecules involved, such as dipeptide, tripeptide, tetrapeptide, pentapeptide, hexapeptide, or polypeptide, for a few amino acid molecules. While the distinction between peptides and proteins is not precisely defined, polypeptides typically have a molecular weight of <10 000, consisting of 2 to 100 amino acids. Peptide bonds (–CO–NH–) link amino acids, where the carboxyl group (–COOH) of one amino acid forms a covalent bond with the amino group ($-NH_2$) of another, releasing an H_2O molecule. While peptides share a structural resemblance with proteins, they are smaller and simpler. Peptides exhibit exceptional nutritional value, biological activity and even medicinal properties, playing crucial roles in the body, such as in hormones, insulin and glutathione.

Peptides can be either produced directly by organisms in vivo or hydrolyzed from large-molecule weight proteins in vitro. The by-products of aquatic food processing, abundant in proteins, serve as a valuable source for producing high-value bioactive peptides, including antioxidant, antibacterial and immunomodulatory peptides. These peptides find applications in the food, functional food and cosmetic industries (Ramakrishnan *et al*., 2023).

2.3.2 Raw material

Peptides, considered a low molecular weight protein, can be generated through the hydrolysis of proteins. The by-products of aquatic food processing, particularly those derived from processing aquatic animals such as fish, shrimp, mussels and sea cucumbers, such as viscera, skin, scales and bones, abound with proteins suitable for producing peptides (Sila and Bougatef, 2016). Nevertheless, the molecular weight and bioactivity of the resulting hydrolyzed peptides are significantly impacted by the choice of raw materials and the processing methods employed.

2.3.3 How to make peptides from the by-products of aquatic food processing

There are three main procedures in the production of peptides: sorting and cleaning, hydrolysis, separation and recovery of peptides.

Sorting and cleaning

The raw material consisting of the by-products of aquatic food processing is sorted according to different species and tissues. The contaminants are removed from the raw material and cleaned with water. The raw material is then cut or milled into small pieces to facilitate the hydrolysis.

Hydrolysis of the materials

The hydrolysis of the by-products of aquatic food processing predominantly consists of four methods: enzymatic hydrolysis, autolysis, chemical hydrolysis and fermentation hydrolysis. Of these, enzymatic hydrolysis is the most common method used to produce hydrolysate (Ananey-Obiri, Matthews and Tahergorabi, 2019).

1. Enzymatic hydrolysis. Enzymatic hydrolysis is the addition of various enzymes, such as papain, pepsin, trypsin, chymotrypsin, flavourzyme, neutrase, protamex and bromelain to digest the by-products of aquatic food processing into small fragments. The degree of hydrolysis (DH) is a fundamental parameter that characterizes the production of protein hydrolysates, which are defined as the percentage of broken peptide bonds in relation to the original protein (Zamora-Sillero, Gharsallaoui and Prentice, 2018). Raw materials, enzyme types and enzymatic conditions of pH, temperature, time, concentration and solid–liquid ratio play critical roles in the DH and quality of the hydrolysates (Chalamaiah *et al*., 2012). Usually, response surface methodology is employed to optimize the complex enzymatic conditions of the by-products of aquatic food processing (Halim, Yusof and Sarbon, 2016).
2. Autolysis. Autolysis is a simple method of producing hydrolysates from the by-products of aquatic food processing. Because these by-products, especially the viscera of aquatic animals, contain various endogenous enzymes, they can be used to produce bioactive peptides. Enzymatic conditions of pH, temperature, time, enzyme quantity and solid–liquid ratio also play critical roles in the hydrolysis rate and quality of the hydrolysates (Huang, Wang and Tu, 2023). However, the main limit to the production of peptides by autolytic hydrolysis is a reduction of bioactivity and the inhomogeneous hydrolysate (Zamora-Sillero, Gharsallaoui and Prentice, 2018).
3. Chemical hydrolysis. The chemical hydrolysis of the by-products of aquatic food processing involves the break-down of the proteins into peptides and amino acids using either acid or alkaline, such as HCl, and NaOH. Although this hydrolysis method is simple and cheap, it is difficult to control the progress of hydrolysis and the quality of the hydrolysates, such as bitter taste, reduced nutrition and poor functionality (Ananey-Obiri, Matthews and Tahergorabi, 2019).
4. Fermentation hydrolysis. Fermentation hydrolysis uses microorganisms to break down proteins into peptides and amino acids. Many different microorganisms have been identified and applied to produce hydrolysates or bioactive peptides from the by-products of aquatic food processing, such as lactic acid bacteria Pediococcus acidilactici, Enterococcus faecium and Aspergillus oryzae (Marti-Quijal *et al.*, 2020; Fang *et al.*, 2017). This method also offers a unique advantage in that it can remove some hyperallergic or antinutritional components such as trypsin inhibitors, glycinin, β-conglycinin, and phytate from the material (Hou *et al*., 2017). However, the bioactivity and quality of the hydrolysates derived from the by-products of aquatic food processing are highly dependent on the microorganisms and fermentation conditions.

Separation and recovery of peptides

The hydrolysates derived from the by-products of aquatic food processing are composed of various peptides with different molecular weights, free amino acids, unhydrolyzed proteins, and small amounts of other ingredients. Typically, the bioactivity and functions of the peptides are strongly related to their molecular weights. The separation of peptides with specific molecular weight ranges consists of two main methods: gel filtration chromatography and membrane separation (Pezeshk *et al*., 2019). Separation or analysis of a small amount of hydrolysate often uses gel filtration chromatography (Idowu *et al*., 2019) while membrane separation is more suitable for large-scale hydrolysate separation, especially in industrial production (Zhou *et al*., 2023). Finally, the fractionated peptides are concentrated or lyophilized into powders for use.

FIGURE 5. **Schematic diagram of peptide production from the by-products of aquatic food processing**

2.3.4 Utilization

Peptides derived from the by-products of aquatic food processing exhibit diverse bioactivities, such as antioxidative activity, skin protective activity, antimicrobial activity, anti-obesity activity, antihypertensive activity and anticancer activity (Ngo and Kim, 2013; Halim, Yusof and Sarbon, 2016; Sila and Bougatef, 2016; Fernando, Jayawardena and Wu, 2023). Their bioactivity, functionality and nutritional value make them suitable for incorporation into nutraceuticals, functional foods and cosmetics. Although the peptides derived from the by-products of aquatic food processing are not currently used in the pharmaceutical industry, their promising medicinal activities suggest potential applications in drug development, particularly for preventative treatments targeting conditions such as cancer, arteriosclerosis and diabetes (Fernando, Jayawardena and Wu, 2023).

These peptides, such as fish and sea cucumber peptides, are rich in essential amino acids and are easily absorbed because of their low molecular weight. These qualities enhance their suitability for use in foods, nutraceuticals and functional foods. Additionally, collagen peptides can be used extensively in the cosmetic industry to produce creams, moisturizers, facial masks, body lotions, etc.

2.4 CHITIN AND CHITOSAN

2.4.1 Product description

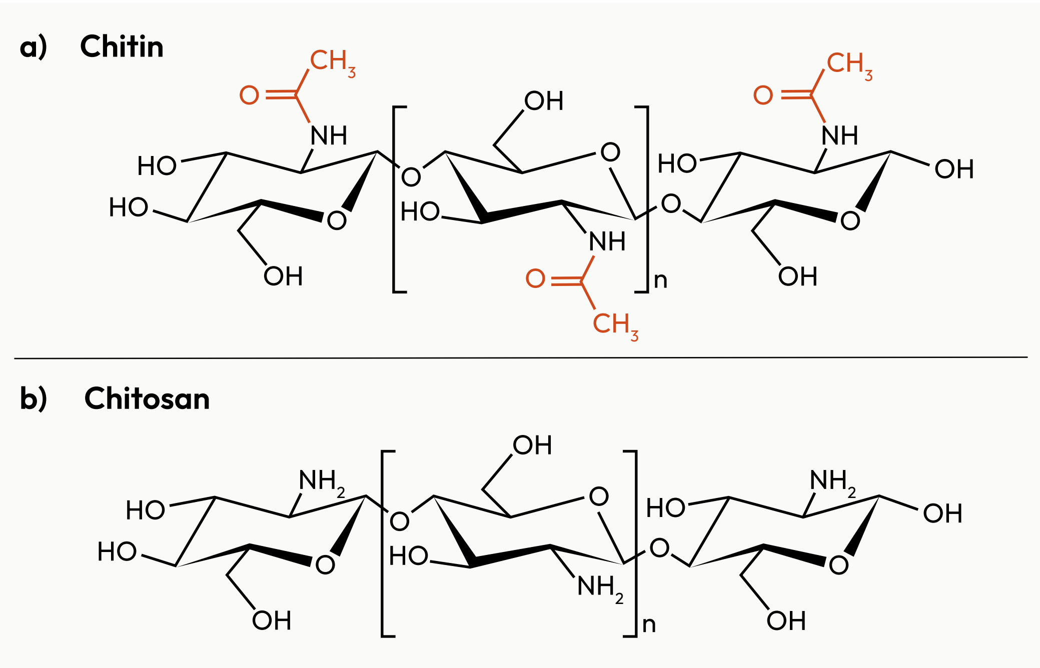
Chitin, the second most abundant natural biopolymer, is a major constituent of the exoskeletons of crustaceans, invertebrates and insects, as well as the cell walls of fungi and yeasts (Tan *et al.*, 1996; Shahidi and Abuzaytoun, 2005; Rinaudo, 2006; Iber *et al.*, 2022). This structural polymer, present in three polymorphic forms (α-chitin, β-chitin, and γ-chitin), is known for its ability to form solid structures independently or combine with substances such as calcium carbonate ($CaCO_3$) to enhance strength, as seen in the shells of crabs and shrimp.

The α-chitin form, characterized by anti-parallel strands, is the most stable and prevalent in nature. It is found in the shells of crustaceans, insect cuticles, fungal and yeast cell walls, and marine sponges (Hamed, Özogul and Regenstein, 2016). β-chitin, which features parallel chains, is less common and found in specific organisms such as squid pens, extracellular fibres of diatoms, and the spines and chaetae of annelids (Lavall *et al.*, 2007; Huang *et al.*, 2018; LeDuff and Rorrer, 2019). γ-chitin, a mixed form of α-chitin and β-chitin, is observed in squid pens, cuttlefish bones and certain beetle cocoons (Rinaudo, 2006; Hamed, Özogul and Regenstein, 2016; Arrouze *et al.*, 2021). Because of its crystalline network structure and extensive hydrogen bonding, chitin remains insoluble in common solvents, presenting challenges in various applications (Pohling *et al.*, 2022).

Chitosan, a deacetylated derivative of chitin with a deacetylation degree exceeding 50 percent, has pivotal characteristics influenced by factors such as its deacetylation degree and molecular weight. These parameters impact solubility, viscosity, ion exchange capacity and flocculation properties (Struszczyk, 2002). Chitosan exhibits solubility in aqueous acidic media, which distinguishes it from chitin. This unique quality, influenced by deacetylation degree and molecular weight, makes chitosan suitable for various applications, including protein recovery and depollution. Its solubility in aqueous acidic solutions also broadens its applications in solutions, gels, films and fibres (Rinaudo, 2006).

The distinct attributes of chitosan and its derivatives present beneficial functions and biological activities such as antioxidant, antimicrobial, antivirus, anti-inflammatory, haemostatic, hypocholesterolaemic, immunostimulating, antitumour and heavy metal adsorption effects (Xia *et al.*, 2011; Xu *et al.*, 2021a; Xu *et al.*, 2021b; Wang *et al.*, 2012; Li *et al.*, 2021; Li *et al.*, 2020; He *et al.*, 2021).

FIGURE 6. **Schematic diagram of the structure of chitin (a) and chitosan (b)**



2.4.2 Raw material

In both the laboratory and industry, chitin and chitosan are mainly produced from aquatic food waste consisting of the shells of crabs and shrimps and squid pens (Nwe, Furuike and Tamura., 2014; Hamed, Özogul and Regenstein, 2016; Yadav *et al.*, 2019). The annul global production of crustaceans grew to about 16.6 million tonnes in 2020, resulting in 7.3 million tonnes to 9.7 million tonnes of crab, shrimp and lobster waste. Production of chitin and chitosan from crustacean waste will valorize the by-products of aquatic food processing (Amiri *et al.*, 2022).

2.4.3 How to make chitin and chitosan from the by-products of fish processing

The amount of chitin derived from shell waste varies between species. In general, it amounts to 10 percent to 30 percent in the shells of shrimp and crabs, 60 percent to 75 percent in the shells of lobsters, and 6 percent to 40 percent in squid pens and cuttlefish bones. The exoskeletons also contain proteins, minerals (mainly $CaCO_3$), pigments (e.g. astaxanthin) and lipids (Hamed, Özogul and Regenstein, 2016). Thus, the production of chitin involves demineralization, deproteinization, decolouration of the shells, and additional deacetylation for chitosan production.

Sorting and cleaning

The raw materials used to produce chitin are sorted by species, followed by washing, drying and reducing into a powder. It is very important to separate the raw materials of shrimp shells and crab shells containing α-chitin from other materials containing β-chitin, such as squid pens and cuttlefish bones.

Production of chitin

1. Chemical method. The chemical method to produce chitin includes the following three main steps: demineralization of the raw material with an acidic treatment, usually using HCl, to remove mineral constituents like $CaCO_3$; deproteinization of the demineralized material with alkali (e.g. NaOH) to remove the proteins and lipids; and an optional decolouration of the material mentioned above with potassium permanganate ($KMnO_4$) or hydrogen peroxide (H_2O_2), acetone or organic solvent mixtures to obtain colourless chitin. Although the chemical method to produce chitin has many drawbacks, such as being uneconomical, environmentally unfriendly, and negatively affecting the properties of chitin, it remains the most effective and commonly used method for the industrial production of chitin (Hamed, Özogul and Regenstein, 2016).
2. Biological method. The biological method to produce chitin includes the following three main steps demineralization of the raw material with LAC-secreting microorganisms to break down $CaCO_3$; deproteinization of the material with protease-secreting microorganisms to remove the proteins; and an optional decolouration of the material mentioned above with potassium permanganate ($KMnO_4$) or hydrogen peroxide (H_2O_2), acetone or organic solvent mixtures to obtain colourless chitin. Although the biological method to produce chitin is environment friendly and safe, the efficiency and quality of chitin are closely related to the microorganisms and the culture conditions. Thus, this method is currently limited to laboratory-scale production of chitin (Hamed, Özogul and Regenstein, 2016; Kaur and Dhillon, 2015).

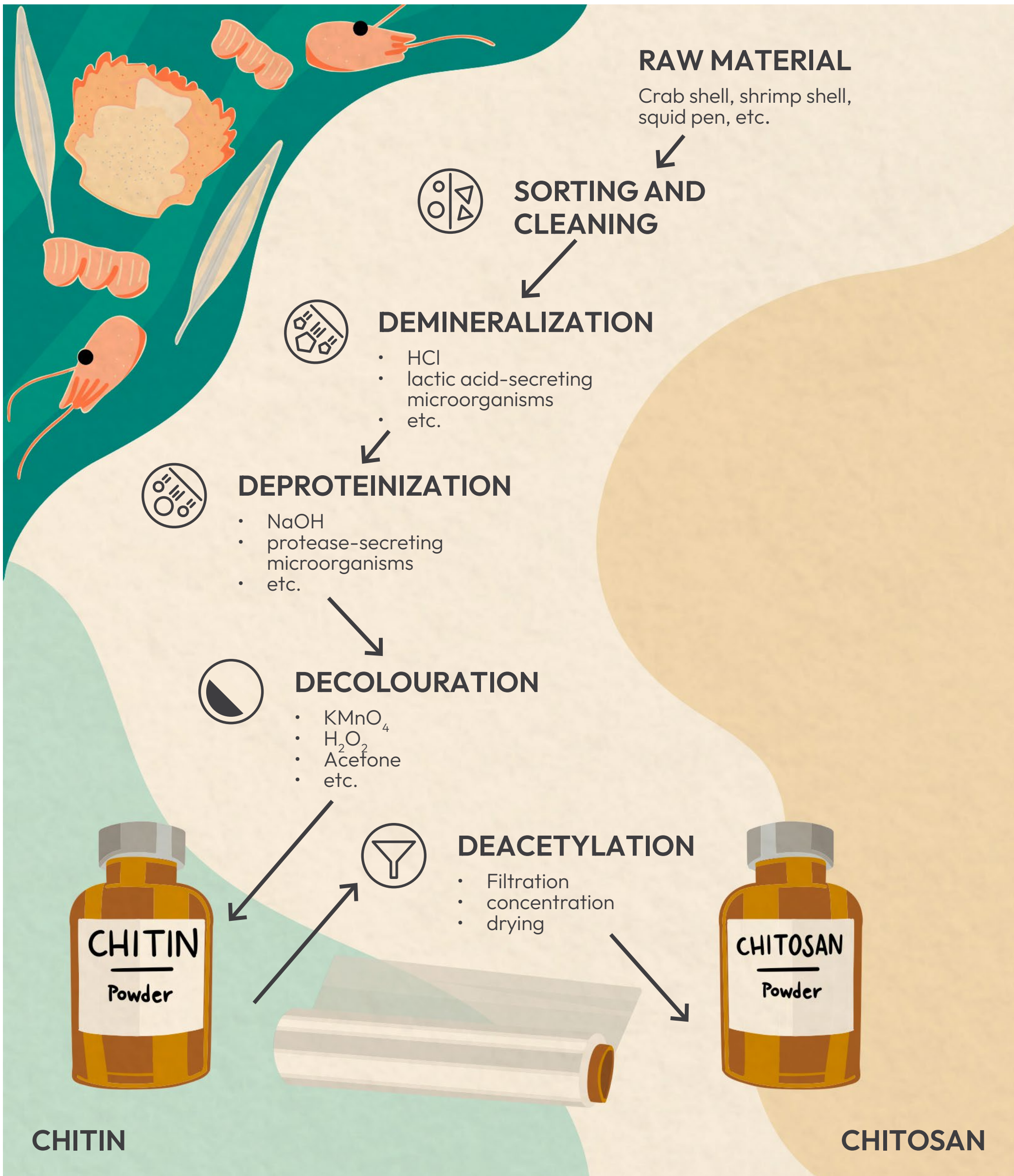
Production of chitosan

1. Alkaline method. The alkaline method to produce chitosan from chitin traditionally uses hot NaOH. The resulting chitosan, with different degrees of deacetylation, is largely dependent on the reaction temperature, time and

concentration of the alkali solution. The alkaline method to produce chitosan is often used together with the chemical method to produce chitin (Shahidi and Abuzaytoun, 2005). The alkaline production of chitosan is environmentally unfriendly and requires a lot of energy.

2. Enzymatic method. The enzymatic method to produce chitosan traditionally uses chitin deacetylase to remove the acetyl groups from the chitin. The chitin deacetylase is often found in fungi, bacteria, protozoa, algae and insects (Ma *et al.*, 2020; Huang *et al.*, 2022). This method to produce high-quality chitosan is effective but expensive.

FIGURE 7. **Schematic diagram of chitin and chitosan production using the by-products of aquatic food processing**



2.4.4 Utilization

Chitin, chitosan and their derivatives have been widely applied in many sectors such as the food, cosmetics, pharmaceutics, textiles, agriculture, water and waste treatment industries (Rinaudo, 2006; Bhatnagar and Sillanpää, 2009; Elieh-Ali-Komi and Hamblin, 2016; Shahidi, Arachchi and Jeon, 1999).

Food industry
Chitin, chitosan and their derivatives have been used in many ways in the food industry. For instance, they can be used as natural antimicrobial preservatives in food processing and storage because of their strong antimicrobial reaction to bacteria, yeast, fungi and viruses (Shahidi, Arachchi and Jeon, 1999; He *et al.*, 2021; Qin and Li, 2020). They have also been successfully used as edible food and fruit packaging film because of their good film-forming properties, biodegradable nature, antioxidant and antimicrobial activity (Elsabee and Abdou, 2013; Han and Aristippos, 2005). Chitosan salts can be used as a clarifying agent for natural fruit juice because of their strong positive charge (Chatterjee *et al.*, 2004) and chitosan has been used as a food additive, food quality enhancer and functional food because of its effects on dietary fibre and functional ingredients (Shahidi, Arachchi and Jeon, 1999).

Pharmaceuticals and medicine industry
Chitosan and its derivatives have been used in various pharmaceuticals and medicine applications. One of the main biomedical commercial applications of chitosan is in wound healing in the form of nonwovens and nanofibre materials, gels, films, composites and sponges, which can stop bleeding and accelerate wound healing and dermal regeneration. Chitosan is a biocompatible molecule with low immunogenicity and can also be used for tissue engineering and bone regeneration. Chitosan-based material can be used to deliver drugs for controlled drug release, encapsulation, enzymes, cell immobilization and as a gene carrier, and in ophthalmology, dentistry and veterinary in the form of coated colloidal systems, hydrogels and nanoparticles (Jayakumar *et al.*, 2010a; Jayakumar *et al.*, 2010b).

Textile industry
Chitosan and its derivatives have good film forming ability, improve dyeing and thickening and are biocompatible and biodegradable. They also have antimicrobial, antistatic and deodorizing properties that are non-toxic and non-allergenic. Chitosan and its derivatives are also used as antimicrobials and cosmetotextiles to produce new functional products in the textile industry (Morin-Crini *et al.*, 2019, Jagadish *et al.*, 2017).

Agriculture
In place of traditional chemical pesticides, Chitosan and its derivatives have excellent antibacterial, antifungal and antivirus properties that can be developed into new, low pollution pesticides to protect plants against pathogens (Qin *et al.*, 2012; He *et al.*, 2021; Guo *et al.*, 2007; Fan *et al.*, 2023). The properties of chitosan can also boost the innate immunity of plants to defend against pathogen infections (Chandra *et al.*, 2015). Chitosan and its derivatives can also be used as plant growth regulators to promote plant growth and enhance crop yield (Morin-Crini *et al.*, 2019). In addition, nanoparticles in chitosan and its derivatives can be used as a way to control the release rate of fertilizers or pesticides for slow, controlled and targeted delivery of agrochemicals to plants (Riseh, Vazvani and Kennedy, 2023; Fan *et al.*, 2023).

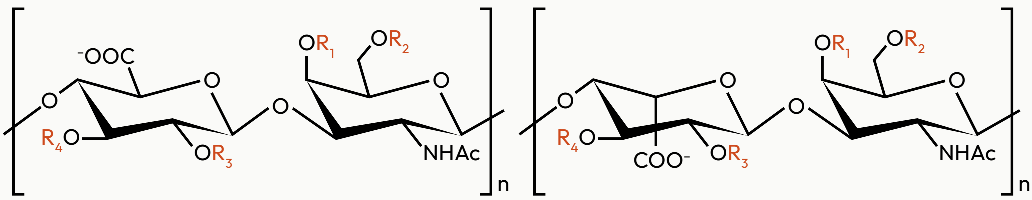
Environmental treatment industry
Chitosan and its derivatives absorb metal ions, dyes and chemicals such as pesticides, phenol derivatives, polychlorinated biphenyl and radioisotopes because of their outstanding capacity to bind to pollutants, versatility and biodegradability in the form of gels, beads, sponges, films and fibres (Morin-Crini *et al.*, 2019). They can also be used as an antifouling agent or recovery material for precious metals (Wang *et al.*, 2012).

2.5 CHONDROITIN SULPHATE

2.5.1 Product description

Chondroitin sulphate (CS), the most abundant glycosaminoglycan (GAG) in the human body, is an important structural component of the extracellular matrix in cartilaginous tissues because it binds core protein to form aggrecan, the most important proteoglycan in cartilage. CS is a linear polysaccharide with a structure of 20 to100 repeated disaccharides containing N-acetyl-β-D-galactosamine (GalNAc) and β-D-glucuronic acid (GlcA). It can be classified into different types based on the position of a sulphate group. Some CS was proved to exist in a different repeated disaccharide containing GalNAc and iduronic acid (IduA) and then renamed, e.g. CS-B, also named dermatan sulphate (Abdallah *et al.*, 2020; Pomin *et al.*, 2019).

FIGURE 8. **Schematic diagram of the structure and classification of chondroitin sulphates**



CS-O: $R_1=R_2=R_3=R_4=H$
CS-A: $R_1=SO_3^-$, $R_2=R_3=R_4=H$
CS-C: $R_2=SO_3^-$, $R_1=R_3=R_4=H$
CS-D: $R_2=R_3=SO_3^-$, $R_1=R_4=H$
CS-E: $R_1=R_2=SO_3^-$, $R_3=R_4=H$
CS-K: $R_1=R_4=SO_3^-$, $R_2=R_3=H$
CS-L: $R_2=R_4=SO_3^-$, $R_1=R_3=H$
CS-M: $R_1=R_2=R_4=SO_3^-$, $R_3=H$

DS-iO: $R_1=R_2=R_3=R_4=H$
DS-iA: $R_1=SO_3^-$, $R_2=R_3=R_4=H$
DS-iB: $R_1=R_3=SO_3^-$, $R_2=R_4=H$
DS-iC: $R_2=SO_3^-$, $R_1=R_3=R_4=H$
DS-iD: $R_2=R_3=SO_3^-$, $R_1=R_4=H$
DS-iE: $R_1=R_2=SO_3^-$, $R_3=R_4=H$
DS-iK: $R_1=R_4=SO_3^-$, $R_2=R_3=H$

The structural variations of CS lead to different pharmacological effects (Sugahara and Yamada, 2000; Inokuma *et al.*, 2023). CS has shown remarkable antioxidant, anticoagulant, articular cartilage repair, corneal lesion healing, antidiabetic and antiproliferative effects, preventing the hardening of arteries and relieving arthritis (Yang *et al.*, 2020) and has been widely used in medical applications, biomaterial applications, functional food, cosmetics and other fields.

2.5.2 Raw material

Animal cartilage from marine organisms (shark, skate, squid and sturgeon) and terrestrial animals (bovine, porcine and avian) are used as the main materials for CS production (Urbi *et al.*, 2022; Sim *et al.*, 2007; Wang *et al.*, 2020). Animal cartilage has different structures and characteristics, depending on the extraction method and the animal species (Trivedi *et al.*, 2016). The by-products of aquatic food processing, including cartilage, bone, head, eyes, fins and skin contain many chondroitin sulphates, in particular the cartilage of sharks, catsharks, skates, squid, octopus, blue sharks and the bones of monkfish, spiny dogfish, cod, tuna, sturgeon, and salmon. Of these, the cartilage and fins of sharks have been most commonly used to produce CS.

2.5.3 How to make chondroitin sulphate from the by-products of fish processing

The production of chondroitin sulphate from the by-products of aquatic food processing includes the following three steps: sorting and preparation of raw materials; extraction and recovery of CS; and isolation and purification of CS.

Sorting and preparation of raw materials

The raw materials used to produce CS are sorted by species because different raw materials may contain different types of CS and result in a mixture of different CS types. Subsequently, the raw materials are washed and milled before the extraction of CS.

Extraction of chondroitin sulphate

Extraction of total GAGs is the first step for the preparation of chondroitin sulphate. Currently, there are two main methods for the extraction of GAGs from the by-products of aquatic food processing: the alkali method and the enzyme method.

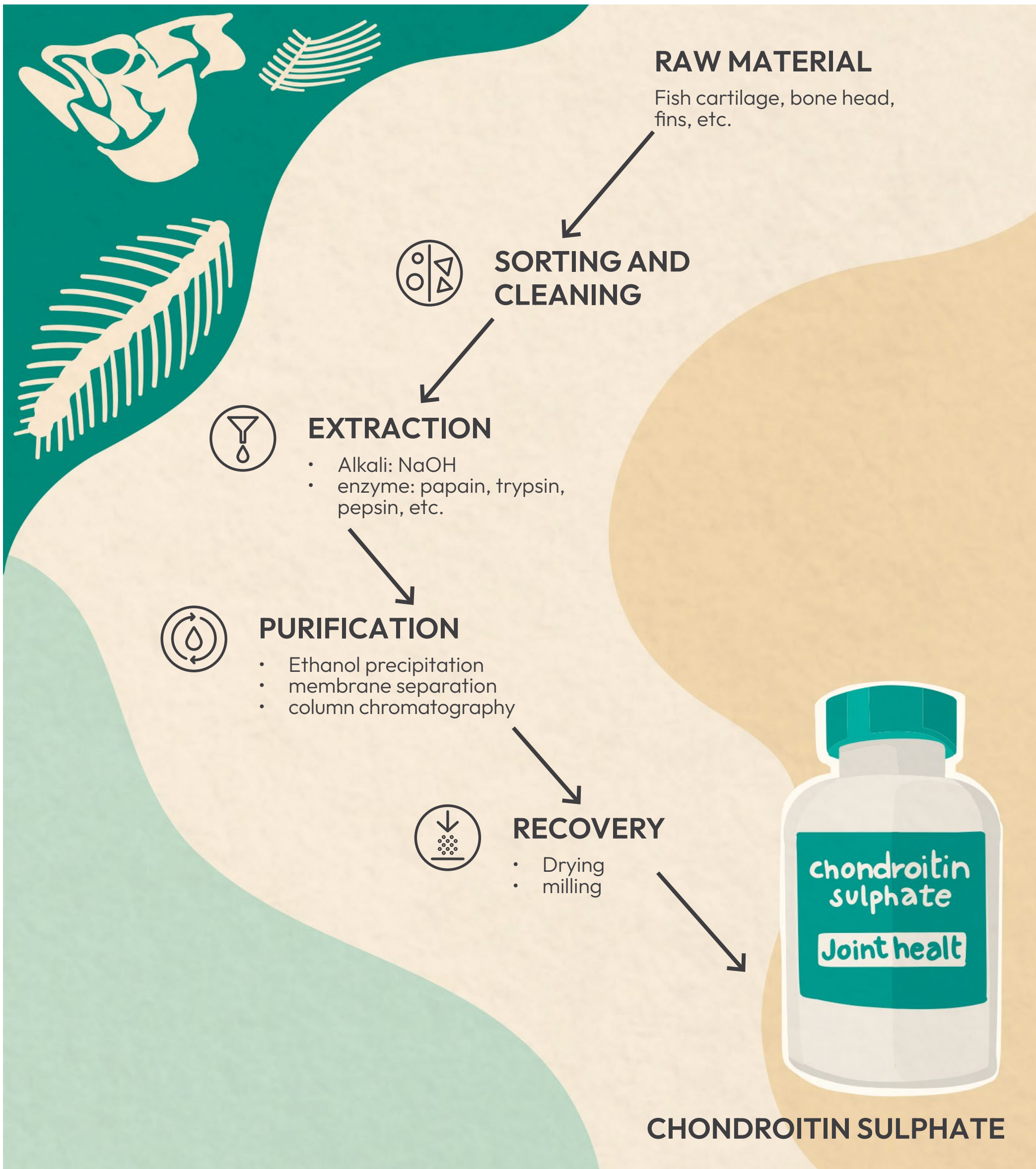
1. Alkali method. Generally, NaOH is used to extract the crude GAGs from the by-products of aquatic food processing. The yield and quality of the CS are largely dependent on the concentration of NaOH, reaction temperature and extraction time. In addition, ultrasonic methods can be used to accelerate the extraction of CS. Although the alkali method is a simple and efficient way to extract CS, the high concentration of alkali can cause degradation and reduced activity of the CS.
2. Enzyme method. The enzyme method is a common technique used to extract crude GAGs from cartilage by enzyme digestion of the core protein. In general, the raw materials are treated with enzymes such as papain, trypsin, pepsin, pronaze, alcalaze and flavourzyme. Raw materials, the types and concentration of enzymes and reaction conditions of the pH value, temperature, time and solid–liquid ratio play a critical role in the extraction of crude GAGs. A combination of two or more enzymes may also have a higher yield with a reduction in treatment time because of the synergistic effects of different enzymes (Xie, Ye and Luo, 2014). Therefore, using a combination of different enzymes is recommended for the extraction of crude GAGs with higher purity.

Purification and recovery of chondroitin sulphate

There are three main methods to recover and purify the CS from the hydrolysate containing all the GAGs, proteins and peptides.

1. Ethanol precipitation. The hydrolysate is precipitated by different concentrations of ethanol to isolate and recover the CS. Although purer CS can be obtained after repeating ethanol precipitations, a significant amount of ethanol will be used in large-scale production of CS (Gavva *et al.*, 2020).,
2. Membrane separation. Ultrafiltration membranes with different pore sizes can be used as an alternative to separate the CS from peptides and other components based on differences in molecular weight.
3. Column chromatography. Gel filtration chromatography and anion exchange chromatography can be used to separate the CS from other molecules according to the differences in their molecular weight and surface charges. Finally, the recovered CS is dried and milled into powder for further use (Shen *et al.*, 2023).

FIGURE 9. **Schematic diagram of chondroitin sulphates production using the by-products of aquatic food processing**



2.5.4 Utilization

CS has been used extensively in medicine, health food, cosmetics and other fields over the years. Its remarkable properties, including excellent articular cartilage repair, corneal lesion healing, anticoagulant, antidiabetic, and antiproliferative effects, make it particularly valuable. CS has demonstrated efficacy in preventing arterial hardening, relieving arthritis and promoting tissue regeneration, making it a versatile compound with numerous therapeutic applications (Yang *et al.*, 2020; Muzzarelli *et al.*, 2012).

Medical industry

CS has been used in medical applications in tablets, pills, capsules, powders and liquids, as well as pharmaceutical grade injections because of its excellent bioactivity and non-immunogenic properties (Volpi, 2009; Shen *et al.*, 2023). One of the widest applications of CS in Europe and other regions is as a safe drug for the treatment of symptomatic osteoarthritis of the fingers, knees, hip joints, lower back and facial joints. The drug provides resistance to compression, maintains structural integrity and homeostasis, slowing breakdown and reducing pain in sore muscles. (Adebowale *et al.*, 2000; Bishnoi *et al.*, 2016).

Cosmetic industry

CS is an important structural component of the extracellular matrix (ECM) and is related to cell proliferation, differentiation and migration. CS has been shown to improve skin maintenance and regeneration by inducing angiogenesis and collagen deposition (Min *et al.*, 2020). It also displays good antioxidant activity, anti-aging ability and high water-binding capacity. CS has been used as an additive in various cosmetic products, such as creams, facial masks and eye drops.

Biomaterial industry

CS has been used as a functional component in various biomaterials, including hydrogels, scaffolds and delivery systems for tissue engineering applications because of its good biocompatibility, non-toxicity, biodegradability and anionic properties. Hydrogels are insoluble in water and three-dimensional cross-linked networks that can hold large amounts of water. CS can be used alone or with other polymers such as hyaluronic acid (HA) or chitosan to prepare biocompatible hydrogel, biodegradable and hemocompatible cryogens, as well as tissue engineering, bioreactors, cell separation or scaffolding materials (Demirci *et al.*, 2021). CS has been used to prepare drug delivery systems to the human body in nanoparticles, complex film and microcapsules. These drug delivery systems embed and deliver drugs to a destination and then release them under controlled conditions to increase efficacy and reduce adverse reactions to the toxic side effect of drugs (Yang *et al.*, 2020).

2.6 OTHERS

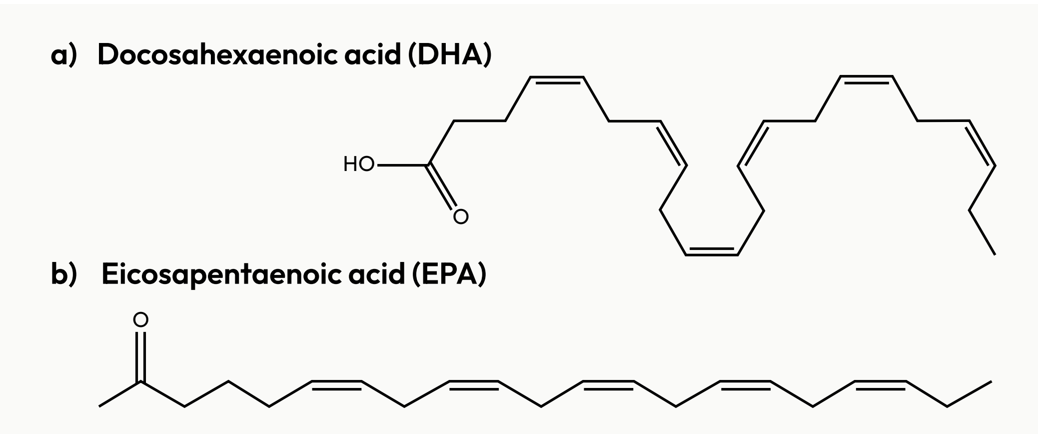
In addition to the high-value products already mentioned, by-products of aquatic food processing can serve as raw materials to produce traditional high-value products, such as fish oil and fish leather.

2.6.1 Fish oil

Product description

Fish oil is derived from fish and is rich in omega-3 polyunsaturated fatty acids (n-3 PUFAs) also called eicosapentaenoic acid (EPA), docosahexaenoic acid (DHA), vitamin A, vitamin D, natural pigments and other nutrients. EPA and DHA are two of the most important n-3 PUFAs beneficial to human health. They may decrease the onset of heart disease and reduce mortality among patients with coronary heart disease by stabilizing the heart's rhythm and reducing blood clotting. They can also have anti-inflammatory effects on the arachidonic acid in the cyclooxygenase and lipoxygenase pathway (Ellulu *et al.*, 2015). Fish oils help to decrease blood viscosity, blood pressure and increase red blood cell deformability (Morris, Sacks and Rosner, 1993; Cartwright IJ, *et al.*, 1985).

FIGURE 10. **Schematic diagram of the structure of docosahexaenoic acid (a) and eicosapentaenoic acid (b)**



Fish oil has been consumed as a health food for many years because of the many benefits it has for cardiovascular, neurology, joint, hypertension, hyperlipidaemia, chronic inflammation, autoimmune disease, rheumatoid arthritis, depression and diabetes mellitus (Greene *et al.*, 2013; Campbell *et al.*, 2013; Weitz *et al.*, 2010; Sales, Oliviero and Spinella, 2008; Vergili-Nelsen, 2003; Yi *et al.*, 2023).

Raw material

The raw materials used to produce fish oil are from fish or the by-products of various species of fish, such as mackerel, herring, tuna, flounder, salmon, sardine, anchovies, large yellow croaker, etc. However, fatty acids vary over different fish species with marine fish containing more EPA and DHA than freshwater fish. Herring, sardine and anchovies have high concentrations of EPA and DHA, salmon have medium concentrations, and sole, halibut, cod and shellfish have low concentrations (Pateiro *et al.*, 2021). Fish oil is a main by-product of fishmeal processing, but the quality of this crude fish oil is very low and requires further refinement to produce high-quality fish oil for human consumption (Norziah, Nuraini and Lee, 2009).

How to make fish oil from the by-products of fish processing

Production of fish oil from the by-products of fish processing involves two main procedures: extraction and refinement.

Fish oil extraction

There are several methods to extract fish oil. These can be mainly classified into three methods: physical method, chemical method and green method (Rubio-Rodríguez *et al.*, 2010).

1. Physical method. The physical extraction of fish oil is also considered to be wet extraction and includes homogenization, heating, pressing and filtration or centrifugation processes (Jayasinghe and Hawboldt, 2012). The physical method is the most common method used for fish oil production and is ideal for fish by-products with a high oil content such as herring, tuna, sardine and salmon. However, an oil–water emulsion is easily formed in the physical extraction, which is too stable to be separated by filtration or centrifugation (Rubio-Rodríguez *et al.*, 2010). In addition, long-chain polyunsaturated fatty acids with double bonds are not stable under high temperatures and repeated heating could reduce the quality of the fish oil (Yi *et al.*, 2023).
2. Chemical method. The chemical method uses alkaline or organic solvents, such as NaOH, potassium hydroxide (KOH), hexane, acetone, methanol and chloroform to extract fish oil from the by-products of fish processing. Alkaline can break the proteins and separate oil from the raw materials. Fish oil is hydrophobic and easily dissolved in organic solvent. The chemical method can also extract other non-lipid substances and thus reduce the quality of the fish oil. Furthermore, the presence of toxic residue from organic solvents poses a challenge when using the chemical method. Organic solvents are no longer widely used in the food industry and are more suitable for analysis in the laboratory than in industrial production (Yi *et al.*, 2023).
3. Green method. Enzymatic extraction, microwave-assisted extraction (MAE), ultrasound-assisted extraction (UAE) and SFE are the main green methods used to extract fish oil from the by-products of fish processing efficiently and in an environmentally friendly way. The enzyme method commonly uses exogenous protein hydrolases such as alcalaze, neutraze and protamex to extract oil from the by-products of fish in a mild, green and safe condition without using organic solvents or high temperatures (Aitta *et al.*, 2021; Marsol-Vall *et al.*, 2022). Enzymatic hydrolysis is an ideal but expensive method to extract fish oil from the by-products of fish processing.

MAE or UAE is an extraction technique that combines microwave energy or sonic cavitation with chemical solvent extraction by rupturing the fish tissue to release the oil to the solvent faster and more effectively. The microwave conditions, including irradiation time, microwave temperature and microwave power could significantly affect the production of fish oil from different raw materials. High temperature-induced oxidative damage to the EPA in fish oil is a significant challenge for the MAE method (Yi *et al.*, 2023). MAE and UAE have been successfully used to extract fish oil on a lab scale and are expected to be applied in industry in the future.

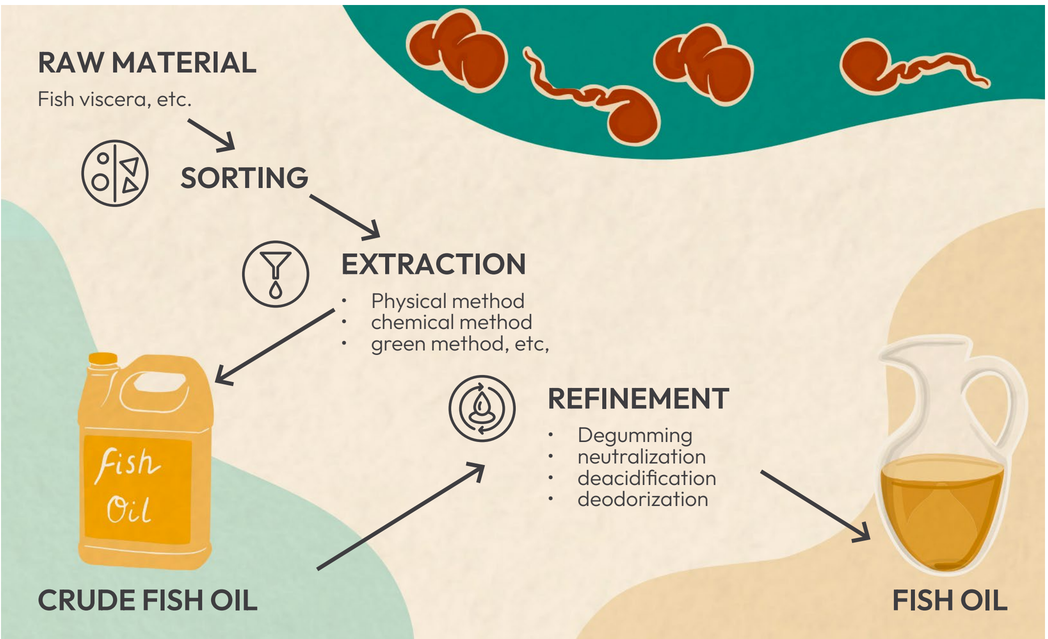
The SFE with carbon dioxide (SFE-CO_2) extraction technique is an effective method to extract fish oil under moderate conditions from the by-products of fish processing. SFE-CO_2 can reduce fish oil oxidation, especially when the fish oil is rich in DHA and EPA and contains contaminants such as arsenic. Furthermore, SFE can be used as a coupled extraction–fractionation process to remove free fatty acids and improve fish oil quality. Although SFE requires higher inversion costs, it still has some advantages over other extraction methods such as the physical method or enzymatic method (Rubio-Rodríguez *et al.*, 2012).

Fish oil refinement

The fish oil extracted via the method mentioned above is usually unrefined oil or crude oil that contains several free fatty acids, glycerides, phospholipids, sterols, proteins, pigments, tocopherols, heavy metals, dioxins or polychlorinated biphenyls (PCBs) and requires further processing to remove non-triglyceride, colourants, bad smelling and toxic compounds before becoming an edible fish oil (Rubio-Rodríguez *et al.*, 2010). Generally, the refining of crude fish oil includes several steps: degumming, neutralization, deacidification, bleaching and deodorization.

The chemical method is the conventional refining treatment used to remove other undesirable compounds from the crude fish oil. However, this method needs to use alkalis, resulting in the loss of some neutral oil and environmental problems. Physical refining processes using high temperatures to remove free fatty acids and volatile and bad smelling compounds damage the PUFAs and form many undesirable compounds. Activated carbon, silica gel and vacuum steam distillation can be used to absorb the dioxins, PCBs, pigments and bad smelling compounds from fish oil. In addition, SFE, membrane and enzymatic treatment, rather than chemical or high temperature treatment, are used as alternative methods to refine fish oil (Rubio-Rodríguez *et al.*, 2012; Kuvendziev *et al.*, 2018).

FIGURE 11. **Schematic diagram of fish oil production using the by-products of aquatic food processing**



Utilization

Fish oil products are typically categorized into two types based on the degree of processing: crude fish oil and refined fish oil. Crude fish oil is directly extracted from the by-products of fish processing without further refinement. This lower value fish oil contains various elements such as PUFAs, free fatty acids, glycerides, phospholipids, sterols, proteins and pigments or other potentially toxic components. Its primary application is in animal feed to enhance survival rates and body weight (Fedorovykh *et al.*, 2015). However, crude fish oil may have potential as sustainable biofuel to lessen demand for fossil fuels. However, research in this field is in its early stages (Yahyaee *et al.*, 2013; Adeoti and Hawboldt, 2014).

Refined fish oil enriched with essential nutrients such as DHA, EPA and other nutrients represents a high-value product sourced from the by-products of fish processing. Refined fish oil is currently extensively used in dietary and industrial applications. DHA and EPA are crucial long-chain n-3 polyunsaturated fatty acids that the body cannot naturally synthesize. They play a vital role in disease prevention and health promotion, serving as supplements beneficial to cardiovascular health, brain health, anti-inflammation, immune system support, neurological well-being, joint health, ocular health, skin health and weight management (Siscovick *et al.*, 2017; Zock *et al.*, 2016; Pateiro *et al.*, 2021).

2.6.2 Fish leather

Product description

Fish leather crafted from fish skin is distinct from traditional leathers derived from domestic animals such as cows, pigs and sheep because of its unique natural marks, patterns and structure. Rooted in centuries-old traditions practiced near rivers, streams and coasts worldwide, fish leather has emerged as a sustainable material for parkas, boots, mittens and hats, valued for its natural grain and excellent breathability (Rahme, 2021). Despite its relatively thin profile, fish leather possesses notable strength because of the dense and interwoven collagen fibres in fish skin. As the global demand for traditional leather continues to rise, fish leather provides a compelling alternative to leather derived from land animals (Omoloso *et al.*, 2020).

The use of fish skin as the primary material for fish leather presents a sustainable approach to repurposing the by-products of the fish processing industry. Transforming these by-products into robust, value-added fish leather helps to minimize bioresource waste, address environmental concerns and offers an environmentally friendly alternative to conventional leather (Palomino and Boon, 2019).

Raw material

Many kinds of fish skins from both marine fish and freshwater fish, such as shark, salmon, tilapia, stingray, carp, cod, sea wolf, bass and sturgeon, can be used as raw materials to make fish leather. However, different fish skins produce different types of leather with various textures, strength and scale patterns. Generally, because of the appearance of fish scales, larger sized fish skins with scales have more leather making value (Kanagaraj *et al.*, 2020). For example, shark skin can be transformed into a smooth leather surface after the scales are removed, while scaled fish skin can be processed into a unique leather with a "scale nest" pattern, making it much more valuable. Salmon leather, for instance, is a sustainable and innovative material made from scaled salmon skin that has special physical and aesthetic properties (Ehrlich, 2015).

How to make fish leather from the by-products of fish processing

The production methods for different fish leathers vary and generally involve the following steps: collection and sorting of fish skin, cleaning of fish skin, decolouration, liming and deliming, pickling, tanning, dyeing and finishing.

1. Collection and sorting of fish skin. The characteristics of different fish skins vary significantly, thus necessitating different processing methods for obtaining fish leather from each type. The first step is to collect and sort different fish skins.

2. Cleaning of fish skin. Fish skins sourced from fish processing are treated as by-products or waste and are sometimes mixed with residual meat and scales. It is very important to ensure that the fish skin is clean without any scales and meat or other unwanted materials. Any remaining meat and scales attached to the fish skin is completely and carefully removed without damaging the skin. In addition, it is essential to remove excess fat from fatty fish skins using detergent if necessary (Yong-an and Jing-na, 2011).
3. Decolouration. Some fish skins exhibit dark brown pigmentation throughout their dorsal area. To obtain uniformly coloured fish skin, it is necessary to remove those pigments from the fish skin with sodium sulphide (Na_2S), sodium chlorite ($NaClO_2$) or other chemicals (Duraisamy, Shamena and Bereket, 2016).
4. Liming and deliming. The decoloured fish skin is treated with a lime solution to open the fibres. The lime is and then removed with ammonium chloride (NH4Cl) or ammonium sulphate [$(NH_4)_2SO_4$] and washed with water (Duraisamy, Shamena and Bereket, 2016).
5. Pickling. The fish skin is treated with a pickling process using HCl, H_2SO_4 or LAC to neutralize the residual lime. Enzymes may also be used to make the fish skin soft, supple and pliable.
6. Tanning. Tanning is a crucial step in the process to transform fish skin into fish leather. This involves the use of specific tanning materials to crosslink the reactive sites of collagen and ensure the fibres are separated. There are several methods for tanning, such as chrome tanning, oil tanning, smoke tanning and plant tanning (Duraisamy, Shamena and Bereket, 2016; Cavali *et al.*, 2022; Rahme, 2021).

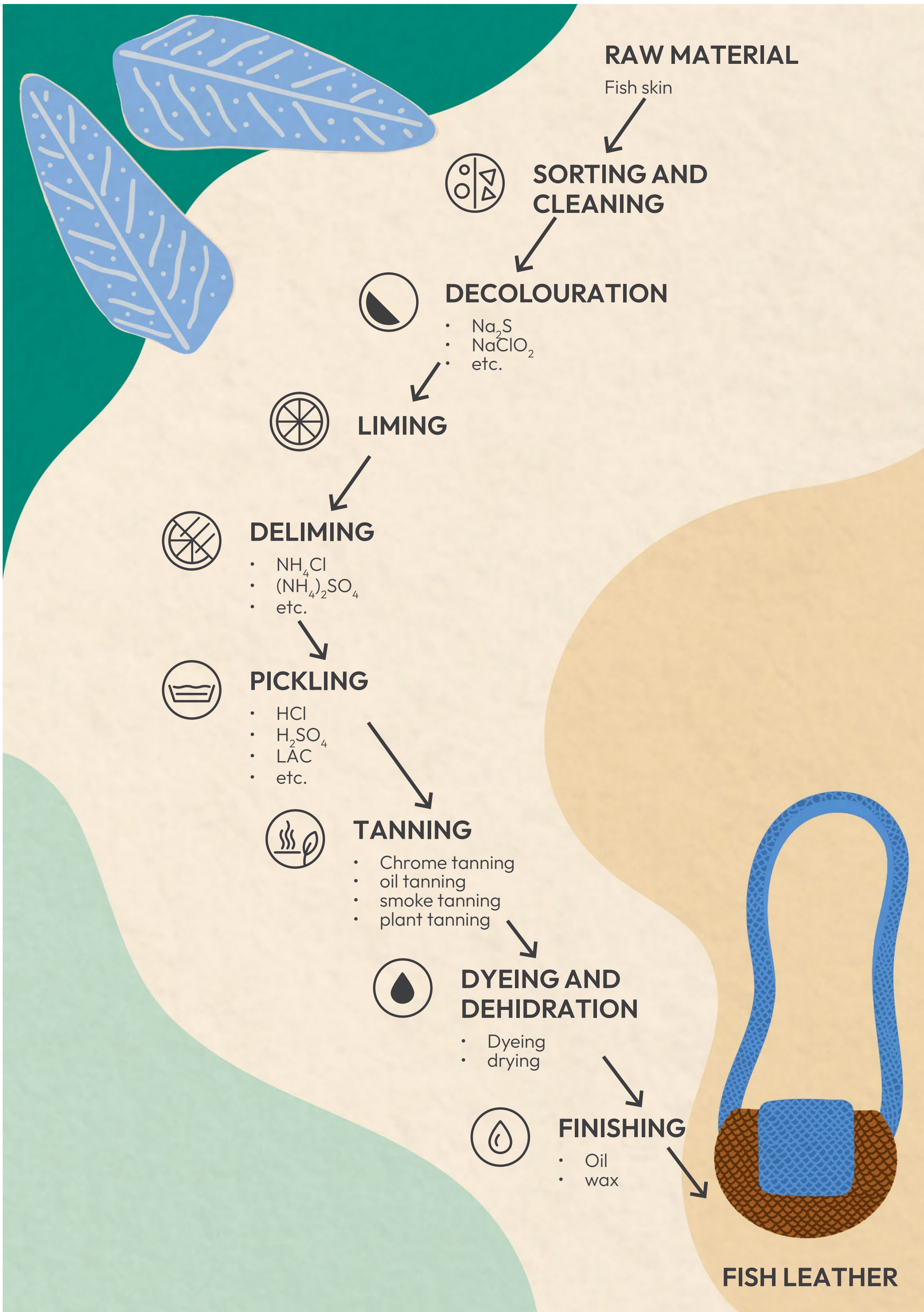
   The chrome tanning process, using chromic sulphate, is highly efficient and extensively used in the leather industry, constituting up to 90 percent of global leather production. However, chrome-based leather tanning raises many questions about health and environmental safety (Duraisamy, Shamena and Bereket, 2016; Rahme, 2021).

   Fish oil contains some unsaturated fats and can oxidize components that form a chemical bond with the fish skin. Fish oil can be used for tanning and produces yellowish, porous and soft leather (Rahme, 2021; Saranya *et al.*, 2020).

   Smoke tanning is a traditional method used in China and Japan. The smoke from burning wood contains various aldehydes featuring carbonyl groups capable of reacting with collagen's amino acids, resulting in the formation of a stable bond. This process gives the skin an attractive golden-brown colour and enhances its water resistance.

   Plant tanning uses various tannic acid-rich vegetables, fruits, woods, roots and plants to bind with the fish skin. This method is a traditional and environmentally responsible method of tanning (Rahme, 2021).
7. Dyeing and finishing. After tanning, fish leather can be dyed to obtain the desired colour. The use of natural dyes is encouraged to maintain the environmentally responsible properties of fish leather. Subsequently, the fish skin is dehydrated by using air drying or the use of a dehydrator. The fish leather can then be treated with oil or wax to improve its durability and appearance (Shirmohammadli, Efhamisisi and Pizzi, 2018).

FIGURE 12. Schematic diagram of fish leather production using the by-products of aquatic food processing

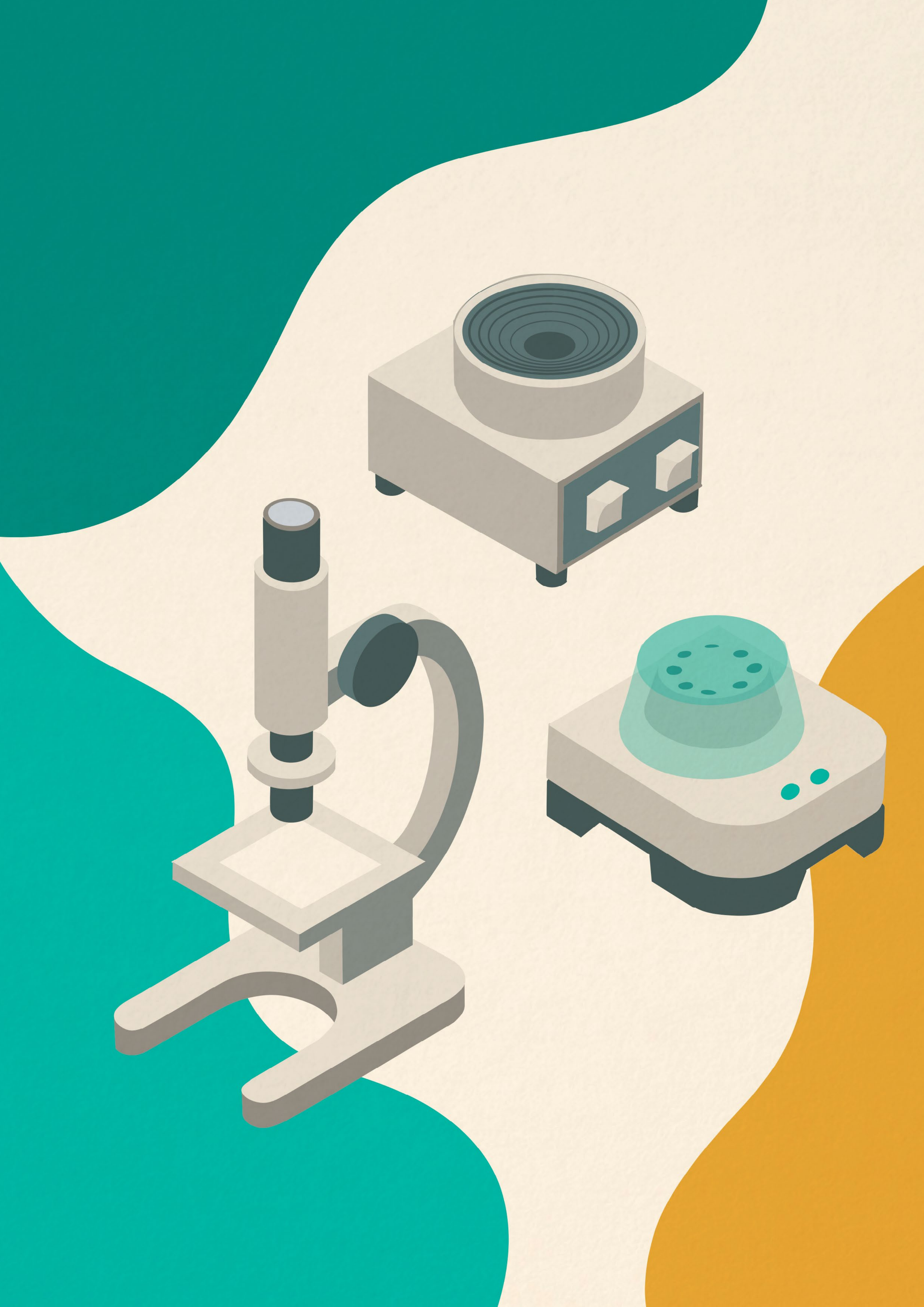


Utilization

For centuries, fish skin has served as a material for crafting leather used in clothing, shoes and homes across human cultures, including ancient Egyptian, Japanese Ainu, Inuit, southern maritime Nanai and Chinese Hezhen societies (Ehrlich, 2015; Rahme, 2021). In ancient Egypt, Nile perch and sturgeon skin were fashioned into fish leather for clothing, accessories and jewellery. Arctic cultures such as the Inuit, used salmon fish skin for crafting clothing, shoes and containers to withstand harsh, cold temperatures.

Compared to other leathers of similar thickness, fish leather exhibits superior strength because of its crosswise fibre structure, in contrast to the parallel structure of cowhide. Beyond its practicality, fish leather's natural grain, breathability, lightweight nature and flexibility have sustained its popularity from ancient times to the present day (Rahme, 2021).

In contemporary times, fish leather finds application in the fashion industry for the creation of parkas, clothes, shoes, boots, mittens, handbags, wallets, belts, hats and various accessories (Alla *et al.*, 2017). The scale patterns on fish leather offer distinctive textures, making it suitable for applications in interior design, furniture production, wall coverings and flooring, adding a touch of luxury and exotic charm. Moreover, the use of fish skin from the abundant by-products of fish processing not only represents a sustainable and innovative approach to crafting high-value products, but also contributes to waste reduction and environmental conservation. As a result, fish leather is emerging as a popular and eco-friendly alternative to traditional leather across diverse industries.



3. Future challenges and recommendations

The by-products of aquatic food processing provide an extensive source of organic raw materials, abundant in nutritive and bioactive components. While certain valuable elements can be successfully extracted from these by-products, purified and applied, several challenges persist from laboratory-scale endeavours to industrial production.

3.1 USE OF HIGH-VALUE PRODUCTS FROM THE BY-PRODUCTS OF AQUATIC FOOD PROCESSING

Currently, the use of high-value products created from the by-products of aquatic food processing remains relatively low. Only a small portion of these by-products has been repurposed into high-value items, with the majority still being used for low-value animal feed and fertilizer or discarded, leading to environmental concerns. Several factors contribute to this situation.

First, inadequate categorization and collection processes result in the mixing of various by-products of aquatic food processing, such as fish heads, bones, skin and scales, during production. This mixing complicates the isolation of high-value components individually, often relegating them to lower-value uses. Additionally, high-value by-products predominantly come from industrial sources, while those from households and restaurants are often treated as direct food waste. Encouraging the classification and collection of diverse by-products during aquatic food processing, regardless of the source, and implementing standardized freshness and quality criteria can promote the high-value use of these by-products.

3.2 TRADITIONAL AND CHEMICAL METHODS VERSUS GREEN INNOVATION TECHNOLOGIES

At present, the technologies used to produce high-value products from the by-products of aquatic food processing primarily rely on traditional and chemical methods, characterized by high energy consumption and substantial environmental pollution. There is a pressing need to develop and implement green innovation technologies for industrial production.

For example, chitin, chitosan and their derivatives offer diverse bioactivities and have yielded various high-value products in fields such as food, cosmetics, pharmaceutics, textiles, agriculture, water and waste treatment. However, the prevalent industrial strategy for extracting chitin, chitosan and their derivatives involves chemical methods, including the use of strong acids and alkalis for shrimp and crab shell treatment, that pose significant environmental threats (Hamed, Özogul and Regenstein, 2016).

Despite the existence of more environmentally responsible biological and enzymatic methods to produce chitin and chitosan, their higher cost has confined them to laboratory-scale operations (Yadav *et al.*, 2019). There is a critical need to develop and implement novel technologies that offer high performance, environmental responsibility and cost-effectiveness in the production of high-value products.

3.3 VARIATIONS IN RAW MATERIALS AND PRODUCTION METHODS

The uniformity of high-value products is often compromised because of variations in raw materials and production methods. Factors such as reaction pH, time and temperature, along with the concentration of acid, base and the types of enzymes used have a significant influence on the properties of the final products. These properties may include the average molecular weight and biological activity of peptides, the deacetylation degree of chitosan, the viscosity of gelatine and the purity of chondroitin sulphate and fish oil.

Standardizing production methods for different products on an industrial scale poses a considerable challenge. Achieving consistency in the quality of high-value products requires meticulous attention to these variables and the development of robust, standardized production processes.

3.4 TREATMENT AND MANAGEMENT OF THE BY-PRODUCTS OF AQUATIC FOOD PROCESSING

The effective treatment and management of extensive by-products of aquatic food processing represent a common challenge for coastal areas globally. Achieving high-value usage of these by-products is hindered by myriad challenges arising from disparities between countries, regions and continents, as well as variations in traditions, cultures, dietary habits and lifestyles. Economic imbalances further contribute to divergent approaches in handling and exploiting the by-products of aquatic food processing. The realization of high-value usage demands substantial investment in modern equipment, factories and technology. Consequently, some developing countries choose to either directly discard these processing by-products or convert them into animal feed, fishmeal or fish silage because of their simplicity and the minimal investment required.

Managing and treating the by-products of aquatic food processing presents a significant challenge that requires sustainable and green approaches for their high-value use. To address this challenge effectively, a range of actions and management strategies should be implemented. These include creating awareness about the high-value potential of these by-products, standardizing and classifying them, promoting established technologies, conducting research and development on innovative technologies, and increasing government guidance and investment. These collective efforts will contribute to both the high-value use of the by-products of aquatic food processing and the sustainable development of aquatic food resources.

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The processing of aquatic food generates substantial by-products, including animal heads, skins, bones, scales, visceral organs and shells, etc., which can constitute between 30 percent and 70 percent of the whole body of aquatic organisms. These by-products retain numerous bioactive molecules suitable for extraction and application in the nutraceutical, functional food, pharmaceutical, biomedical, cosmetic and material industries, and have the potential to yield high-value products. The transition from aquatic food waste to high-value products presents multiple benefits, including: (i) enhanced human nutrition and health through nutrient and bioactive component provision; (ii) mitigation of environmental pollution by reducing waste; and (iii) improved economic returns because aquatic food waste is transformed into high-value products rather than low-value animal feeds or fertilizers.
This guide presents strategic and technical insights by outlining key principles for producing high-value items, including collagen, gelatine, bioactive peptides, chitin, chitosan, chondroitin sulphate, fish leather and fish oil, from the by-products of aquatic food processing.

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