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Value Addition to Leather Industry Wastes and By-Products: Hydrolyzed Collagen and Collagen Peptides

Ali Yorgancioglu, Bahri Başaran and Aykut Sancakli

Abstract

Environmental consciousness and constraints in developed societies over the past 20 years have brought about a dramatic impact on tannery operations worldwide. Leather industry has been categorized as one of the most polluting industries, and it spoils the continuity of environmental rhythm because of the generation of liquid, solid and gaseous wastes and also by-products. Solid organic wastes involving untanned (trimmings, fleshings and splits) and tanned (trimmings, splits and shavings) wastes and by-products depending on their proteinic character have an advantage of recovery and reuse potentials instead of disposal to landfills in terms of environmental sustainability. These solid wastes and by-products are not properly treated and disposed of; hence, they can cause environmental damages to soil and groundwater as well as release emissions and poisonous greenhouse gases into the atmosphere. Valorization of these tannery solid wastes and by-products with different methods and processes is highly important for the perspective of eco-benignity and with respect to converting into new value-added products. This chapter focuses on the evaluation of the tannery solid wastes and by-products by partial and total denaturation and hydrolyzation. This paper also examines in general the specifications, production techniques and applications of collagen peptides in several industries.

Keywords: leather, solid wastes and by-products, technical gelatin, collagen peptides, collagen hydrolysates

1. Introduction

Leather manufacturing, which is an allied industry and subsector for textile, is the first making practice in primitive period of humankind. Different types of animal skin products were used throughout the first ages as parchment and vellum or by making the raw material resistant to putrefaction, heat, chemicals and environmental effects with smoke, potash alum and natural tannin extracts from different plant parts. Traditionally, these products obtained by modification of by-products of meat industry have all been classified as leather, which is a serviceable product. In this respect, the leather industry could have been distinguished as an environmental industry, since it processes waste products from meat production [1].

These natural products generally consist of long thick collagen fibers, fiber bundles and thin elastin fibers of interweaving in three-dimensional ways. Other features such as hairs and hair roots and also fat cells are present in three-dimensional woven structure that predominates and gives skin-based materials providing many of their unique physical and mechanical qualities [2].

The leather-making operation assists in converting the raw hide or skin, a highly putrescible material, into leather, a stable material, which can be used in manufacturing a wide range of products. These include shoes, clothing, leather goods, furniture, upholstery for car seats and interiors, boats and aircraft, and many other goods in daily use. The whole process involves a sequence of complex chemical reactions and mechanical processes [3].

The processing of leather involves four main stages: beamhouse, tanning, post-tanning and finishing. The first phase of the hide processing is called beamhouse operations and involves multiple mechanical, chemical and biological unit operations. Its objective is to remove dirt, hair, epidermis, noncollagenous proteins and grease from raw skin, and open up the collagen fibers to favor the subsequent tanning process [4]. The process is performed in a drum by mixing the raw hides with an alkaline solution containing lime and reducing agents, usually sulfide salts, the hair being chemically removed from the surface of the hide [5]. The beamhouse operations are the most water consuming and the effluents generated present very high organic load [6].

The tanning process is one of the oldest procedures in the world, and currently, these industrial activities are based on chemical processes involving several organic and inorganic compounds [7]. This step gives the leather stabilization against the wet and dry heat, bacterial growth, mechanical stress and enzymatic attack, among others, and forms the basis of leather production. This stabilization is attributed to the formation of new chemical cross-links in the matrix proteins [8]. The tanning stages are classified as mineral, vegetable and synthetic. When the skin stabilization is achieved by a suitable inorganic salt, the process is known as mineral tanning, and the most commonly used mineral tanning salt is the basic chromium sulfate ($\text{Cr}(\text{OH})\text{SO}_4$). If the leather is tanned with chromium salt, it is called as wet-blue leather. Chromium (III) salts are the most extensively used compounds due to the quality and high stabilization ability they impart to leather [9].

The third part in leather production is post-tanning process. The tanned leather is considered a commodity, that is, it may be used to produce several articles. Each post-tanning operation is directed to the article that will be produced, such as garment, shoe upper and upholstery [10]. The aim of the post-tanning processes is to enhance the aesthetic properties of leather by coloring it and changing some physical and mechanical properties of the material by retanning, dyeing and fatliquoring stages [10].

The finishing step complements the previous stage, tanning, and provides the leather with the required physical and mechanical properties, such as color, tensile strength, impermeability, softness, flexibility and elasticity with different kinds of binder, pigment, wax and oils [11]. This operation consists of coating and changing the surface of leather. It is related to the fashion appearance, but also to conferring properties such as abrasion resistance, gloss, handle, flex, adhesion and rub fastness as well as other properties as required for the end use including extensibility, light and perspiration fastness, water vapor permeability and water resistance [10].

Leather industry has been categorized as one of the highly polluting industries because large quantities of water and different chemicals have been used during tanning process and different solid, gaseous and liquid wastes are generated that have an adverse effect on the environment [12]. These wastes have different characteristics because different chemicals are applied to the raw hides in different

ratios. Solid wastes generated in tanneries mainly include salts, raw trimmings, hair wastes, fleshings, splitting wastes, chrome shavings, buffing dusts, crust trimmings and finished trimmings. These solid wastes and by-products are not properly treated and disposed of, and they can cause environmental damages to soil and groundwater as well as release emissions of odor and poisonous greenhouse gases into the atmosphere by direct landfill or incineration, which is an unsustainable way [13].

Salt, which is used to preserve hides or skin thrown into open dumping areas or accumulated in piles outside the tanneries, is likely to create groundwater pollution when rain washes it away. Hair wastes and lime sludge discharged into the effluent can produce choking of treatment pipelines. Trimmings, fleshings and splitting wastes putrefy easily producing noxious odors [14]. Moreover, disposal of chromium-containing solid wastes into soil and water has potential effects on public health due to the possibility of oxidation of chromium (III) into hazardous chromium (VI) [15]. These tannery solid wastes have different characteristics that mainly constitute protein (collagen) as the main component [16].

Provisions for pollution control, waste minimization and disposal, the correct use of chemicals and accident prevention are essential for minimizing potential impact on air, water and soil from the processing of hides and skin.

Collagen derivatives are value-added products extracted from solid organic wastes and by-products, and they are utilized for several industrial applications such as preparation of technical-grade gelatin, protein hydrolysates, collagen peptides and subunits [17]. The processing of hides and skin also generates by-products, which find outlets in several industrial sectors such as pet and animal food production. They can be used in cosmetics, printing inks and photography, while the latter one is an ideal candidate for fertilizer or feeding additives due to their high nitrogen content [18].

The present chapter describes the leather solid wastes, general features of collagen peptides, and their preparation methods and applications in different industries.

2. Leather solid wastes

The tanning industry worldwide produces a significant amount of solid wastes and effluents, environmental concerns about discharge and escalating landfill costs are becoming increasingly serious problems for the industry, and their management alternatives regarding overall consideration have been based on multispot [19]. Huge amounts of solid wastes are generated at different stages of leather processing and there is no actual adopted utilization method available for solid wastes; hence, handling is more difficult for tanners. Leather solid wastes generated in fleshing, trimming, splitting and shaving processes and also sludges discharged from the wastewater treatment plant both contribute to increase the volume of the wastes [20].

Generally, out of 1000 kg of rawhide, nearly 800 kg of solid wastes are generated in leather-manufacturing industries, and only 200 kg of the raw material is converted into a usable product. About 600,000 tons of solid wastes annually are generated worldwide by leather industries [21]. An example of the types and quantities of solid wastes generated in leather processing based on one ton of raw hides/skin is given in **Table 1**.

The ways to disposals and valorizations for these wastes are defined by the chemical characteristics depending on the fact that the wastes are generated in either beamhouse or tanning and after tanning. This differentiation might be, namely, untanned wastes and tanned wastes accordingly.

Solid wastes generated from processing of raw hides/skin (1000 kg)	Quantity (kg)
Conservation salts	80
Hair	100
Raw trimmings	40
Lime sludges	60
Fleshings	120
Wet-blue trimmings	30
Chrome splittings	65
Chrome shavings	95
Buffing dusts	65
Crust trimmings	35
Dry sludge from common effluent treatment plants (CETPs)	125

Table 1.
Solid wastes from tannery [22].

2.1 Untanned solid wastes

Most of the solid wastes are generated in beamhouse, especially in fleshing operation. Fleshings are solid wastes generated during a mechanical process aiming at removing the flesh deposits or fats from the inner part of the skin [23]. Fleshings contain subcutaneous tissue, fat and flesh, which are composed of protein (5–7%), fat (4–18%), lime (2–6%), sulfide (2–4%), etc. [23].

Trimming is to cut out unwanted parts of processed hides/skin just after fleshing operation is completed. Trimmings are cut-outs from the operation and may be collected and shipped to glue manufactures or other by-product manufacturers or sent for disposal in a landfill [24].

Hides are generally subjected to mechanical operation called splitting to divide the hide into two or three layers horizontally. Splitting operation can also be applied at chromium tanning stage (wet-blue stage), which is called wet-blue splitting. Whether split is untanned and obtained after liming or tanned and obtained after tannage, it is a valuable part of a hide, which is a fibrous sheet, and hence it is in fact not a waste and more precisely it is a by-product.

The untanned solid wastes, mainly including leftovers from trimming of rawhide and surplus parts after liming and fleshing, are composed of large amount of collagen and grease. The chemical composition of these solid wastes varies depending on types and quality of the raw hides/skin and also process conditions. Fats and proteins are the main components of these wastes (10.5%). Moisture amounts might be up to 60%, meaning a high water content. The aforementioned solid wastes do not contain chromium compounds [25]. For sufficient usage of these protein-rich wastes, various kinds of methods and technologies have been proposed, focusing on the extraction of collagen/gelatin by using acid, alkali and enzyme hydrolysis and subsequent purification processes. Moreover, grease residue can also be used to extract oils and fats, which can be raw material for biofuel and leather fatliquor [26].

2.2 Chromium-tanned solid wastes

The chromium tanning is based on the cross-linkage of chromium ions with free carboxyl groups in the collagen. Chrome-tanned leather also called wet-blue leather

are characterized by top handling quality, high hydrothermal stability, user-specific properties and versatility [27]. At the end of the chrome-tanning process, 60–75% of the chrome offer (Cr_2O_3) remains in the collagen structure. Additionally, small amounts of other chemicals and auxiliaries such as tensides, acids and bases (in the form of soluble “reaction salts”) remain in the wet-blue leathers. The main environmental impact of tannery solid wastes is the oxidation of trivalent chromium into the hexavalent form, which is highly toxic and has carcinogenic and mutagenic effect. Leakages from chromium-containing wastes when they come to the agricultural lands cause ground water pollution and soil contamination. Water pollution affects aquatic animals, which are common sources of food, and soil contamination poses health effects through food chain and also poses a health hazard through inhalation of toxic dust, which can be inhaled by both people and livestock [28].

The solid wastes containing chromium namely tanned wastes are wet-blue shavings, wet-blue trimmings, buffing dusts, finished leather trimmings and wastewater treatment sludge [29]. Their chemical composition consists of fats and oils (3–6%) and mineral matters (15%). As chromium has been already used worldwide, they normally contain 3.5–4.5% of chromium as Cr_2O_3 . Sludge from effluent treatment plants contains mainly water (up to 65%), organic substances (30%) and chromium (III) (around 2.5%) [25, 30].

Chrome shaving wastes are generated during the machine process of thickness adjustment of wet-blue leathers based on the required thickness. Shavings are mainly the scraps from the flesh side of leather, which are carried out by cutting unusable parts of leather and rags created during shaving operation [31]. Utilization or safe disposal of shavings continues to pose a serious challenge in many countries and is more critical because of their compositions. While processing one ton of raw hide, approximately 95–100 kg of wet-blue shavings are produced [32, 33]. Currently, a part of the chrome shavings is used in the manufacture of different types of areas such as leather board, collagen peptides, gelatin, animal feed and fertilizers. Unused portion of shavings is dumped in open areas around tanneries posing a serious environmental hazard [34].

2.3 Environmental and health impacts of leather solid wastes

The tannery solid wastes can cause severe problems associated with its organic load, inorganic matter, chromium, suspended solids, total organic and ammoniacal nitrogen, sulfide, and chloride, among others, depending on the chemical and mechanical processes applied to the raw hides/skin. Accumulation of these wastes leads to sludge problem and choking of treatment pipes and finally results in the reduction in efficiency of the treatment plant [35].

Leather industry is facing a lot of solid waste problem and many tanneries are closed for not meeting biological oxygen demand (BOD) and total dissolved solids (TDS) norms [4]. It is very important to analyze the nature of these wastes in order to assure a safe disposal or application of them. Salt, which is used to preserve hides or skin, discharges huge amount of pollution load in terms of total dissolved solids and chlorides and creates groundwater pollution [36]. Hair waste and lime sludge if discharged along with the effluents are likely to choke the drains. Trimmings, raw fleshings, limed fleshings and splitting waste can putrefy easily by producing noxious smells. Some of the biodegradable tannery solid wastes cause volatile organic compound emissions and, moreover, are sources of pathogenic bacteria [37].

Shaving dust contains environmentally unfriendly chemical called chromium, and when it is dumped in the environment, it can easily enter into the surface and ground, and this heavy metal pollutes the surface water by erosion and the underground water by leaching and erosion, leading to serious health problems to aquatic

life in nearby rivers. As a result of this, pollution of surface and ground water results in shortage of drinking water for human beings and animals living at the downstream of the rivers [38].

According to Mu et al. [39], about 25% of tannery solid waste ends up as chromium-containing solid waste, which is more dangerous than other tannery solid waste. The waste generated from chrome-tanned leather is not biodegradable and toxic due to the chromium content [40]. Chromium-containing leather waste has been classified as one of the dangerous and hazardous waste if discharged into the environment without any pretreatment. Increased risks for a number of cancers such as lung cancer, testicular cancer, soft tissue sarcoma, pancreatic cancer and bladder cancer have been reported [41]. Chromium waste can also cause respiratory problems, a lower ability to fight disease, birth defects, infertility and tumor formation [42]. Chromium-containing solid waste percolates to the ground and causes ground water pollution and soil contamination. Water pollution affects aquatic animals that are common sources of food, and contamination of soil poses health effects through food chain and also poses a health hazard through inhalation of toxic dust, which can be inhaled by both people and livestock. It can damage the gills of fish; it can alter genetic materials and cause cancer [43]. Moreover, thermal incineration of these wastes is associated with serious air pollution problems due to emission of toxic hexavalent chromium (Cr^{+6}), halogenated organic compounds, aromatic hydrocarbons, etc. into the environment [42].

3. Collagen peptides

The word collagen is derived from the Greek word “kola,” which means gum, and “gen,” which means producing. It is a fibrous structural protein present in extracellular matrix and connective tissue of animals [44]. Collagen is the most prevalent protein comprising approximately 30% of the total protein of animal and human bodies and is found primarily in connective tissues including animal hairs, bones, cartilages, tendons and blood vessels [45, 46].

There are many types of collagens, which are from collagen I to collagen XIX. Animal skin or hide contains collagen type I, mostly with approximately 90% of its dry weight [47]. Collagen precursors are synthesized in the endoplasmic reticulum of cells and transported to the Golgi apparatus in order to secrete into the extracellular spaces, and maturation of collagen can occur [47, 48].

Collagen polypeptide chains and cross-linkages can be broken down by hydrolytic processes and decomposition yielding in different subunits and fragments [49]. In other words, gelatin is produced by partial denaturation of collagen in triple helical structure. Gelatin and collagen peptides are new forms yielded by hydrolysis of native collagen with lower molecular weight fragments than original structure and including a wide range of subcategories having differentiated functionalities [50]. Native collagen exhibits superior and distinct properties from collagen peptides such as higher enthalpy, greater network structure of fibrils, basic isoelectric point and high resistance to protease hydrolysis [51]. The native triple helices and fibril networks in the native collagen are more rigid and firmer than gelatin and collagen peptides [52].

3.1 Structural and chemical characteristics of native collagen

All proteins are composed of linear chains of amino acids attached together by peptide bonds, thereby making oligomers, which are the primary structure of the protein. Being arranged into sequences of different amino acids gives way to fold

up the chains into a functional protein. Intermolecular and intramolecular bonds in the structure can enhance the folding of the peptide chains. This folding induces the weak forces such as hydrogen bonds and electrostatic, hydrophobic and van der Waals interactions [53]. Considering the reactivity of collagen molecule, the peptide bond itself is prioritized, capable of participating in hydrogen bonds with both hydrogen-bond donor and acceptor groups. For example, a carbonyl group in any protein has two lone pairs of electrons having the capability of accepting hydrogen bonds. In addition, the electronegative nitrogen induces a partial positive charge on its attached hydrogen, allowing the hydrogen to function as a hydrogen-bond donor [54]. Hydrogen bonding between peptide bonds is the basis of protein secondary structural formation, namely, helices, yielding in pleats and turns in the structure [55]. The side chains of amino acids are capable of a variety of interactions including hydrogen bonds, ionic bonding, hydrophobic interactions, van der Waals interactions and disulfide bonds. These interactions and secondary structural elements are responsible for taking a shape of the tertiary structure of proteins, their actual three-dimensional shape [56]. Due to the interactions between side chains of various amino acids, the protein molecule will bend and twist so as to gain individual stability or lower energy state.

Proteins included in the number and arrangements of subunits to give functionality are referred to as quaternary structure. The proteins comprising individual subunits may be identical, or they may be different. Like the secondary and tertiary structures, the quaternary structure of a protein is determined by its primary structure [57].

Collagens are trimeric molecules made up of three polypeptide chains, which contain the sequence repeat of (Gly-X-Y)_n, X being frequently proline and Y hydroxyproline. These repeats allow the formation of a triple helix based on three polypeptide chains bound to each other by hydrogen bonding, which is the characteristic feature of the collagen [58]. The side chains of each X and Y residue are at the surface of triple helix, giving the collagen molecule a significant capacity for lateral interactions with other molecules of extracellular matrix and resulting in the formation of various supramolecular assemblies [59, 60]. An interchain hydrogen bonding between glycine and amide group in an adjacent chain is a key factor in stabilizing the collagen triple helix [61].

Collagen protein is more hydrophilic than lyophilic moieties due to the chemical nature of numerous amino acids present in its structure [62]. It has a highly complex structure and interacts with each other at the molecular level to form broader systems with distinctive properties [63]. The chemical structure of collagen type I is shown in **Figure 1**.

Basic properties create characteristic structure of collagen fibril helicoidal structure is; fibril diameters ranging from 10 to 500 nm [64], average molecular weight of 285,000 Da [65] and glycine ratio of 1/3 in the polypeptide chain consisting of 1400 amino acids [66].

Having been readily recognized in tissues with commonly white and opaque colors, collagen fibers are considered as viscoelastic materials having high tensile strength and low extensibility. The tensile strength of collagen depends on the formation of covalent intermolecular cross-links between the individual protein subunits [67].

The collagen family is highly complex and shows a remarkable diversity in molecular and supramolecular organization, tissue distribution and function. Collagen types are classified in several subfamilies according to sequence homologies, similarities in their structural organization and supramolecular assembly. The availability of 27 collagen types was reported and they are classified by their size, function and amino acid distribution that differ considerably in their

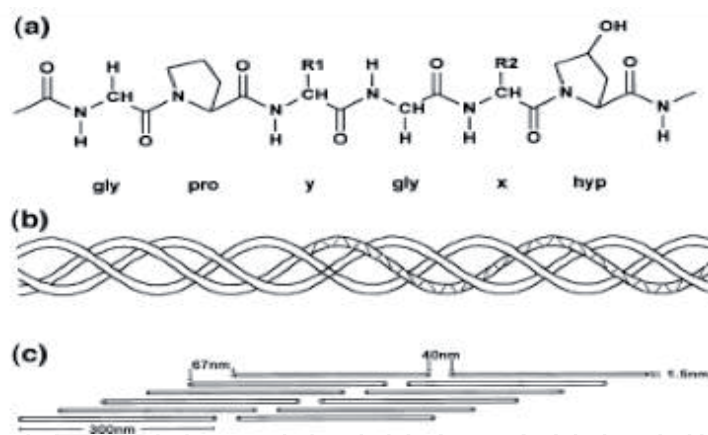


Figure 1.

Chemical structure of collagen type I. (a) Primary amino acid sequence, (b) secondary left-handed helix and tertiary right-handed triple-helix structure, and (c) staggered quaternary structure [63].

molecular structure [68]. The individual members are numbered with roman numerals. The family is subdivided into different classes: the fibrillar collagens (types I, II, III, V, XI, XXIV and XXVII), basement membrane collagens (type IV), fibril-associated collagens with interrupted triple helices (FACIT collagens, types IX, XII, XIV, XVI, XIX, XX and XXI), short chain collagens (types VIII and X), anchoring fibril collagen (type VII), multiplexins (types XV and XVIII), membrane-associated collagens with interrupted triple helices (MACIT collagens, types XIII, XVII, XXIII and XXV) and collagen type VI. The types indicated by an asterisk are heterotrimers, consisting of two or three different polypeptide chains. Type IV collagens contain six different polypeptide chains that form at least three distinct molecules and type V collagens contain three polypeptide chains in probably three molecules [68].

Each collagen type has its own specific amino acid composition and performs a distinctive role in tissues. Types I, II and III are of the most abundant collagens, which are responsible for tissue strength, elasticity and water retention capacity [69]. Type I collagen is the main structural component of extracellular matrix. It consists of one $\alpha 2$ chain and two $\alpha 1$ chains, which are encoded on chromosome 7 and 17 in humans [70]. Generally, type 1 collagen is the most commonly used in industrial scale especially in tissue repair and replacement, and they are intensive in skin, tendon, bone, cornea, dentin, fibrocartilage, large vessels, intestine, uterus, dermis, cornea and connective tissue [71]. It has outstanding mechanical properties and is present in virtually every extracellular tissue with mechanical function. In tendons and ligaments, collagen transmits the force from muscles to bones and stores elastic energy. Smooth walking would not be possible without these properties. Collagen also represents most of the organic matrix of bones and tooth dentin and confers them their fracture resistance. It is a major constituent of skin and blood vessels and is even present in muscles, which could not function without a collagen-rich matrix around the contractile cells. A slightly different type of collagen type II is a critical component of a tissue as soft as articular cartilage. The function of collagen is not only mechanical. In the cornea of the eye, for example, the ordering of collagen fibrils confers transparency in addition to mechanical stability [69]. Type II collagen is prevalent in hyaline cartilage, vitreous, nucleus pulposus, notochord and intervertebral disc. It provides biomarkers for osteoarthritis. Type III collagen is present in fetal dermis and epidermis, veins, uterus, synovium, connective tissue around muscles and also in small quantities in areas where type I collagen is present. Type III collagen is functional of fibrillogenesis of collagen I and for normal cardiovascular development [72].

3.2 Industrial sources of collagen peptides

The major sources of collagen for fabrication are bovine and porcine species, where collagen was extracted from the hides and skin and also bones of pigs and cows. Bovine hides, a by-product of meat production, are one of the major industrial sources of collagen [49]. The bovine hide is composed of approximately 30% protein, and the inner corium layer of the hide is rich in collagen. This collagen has a high denaturation temperature in comparison to collagen from other sources. Bovine hide is practiced upon in different development stages such as bovine dermis used for tendon regeneration, and skin and wound healing (in the form of collagen matrix); neonatal bovine dermis is used for hernia repair, plastic and reconstructive surgery [73].

Starting from the 1930s, the most significant raw material for large-scale industrial gelatin production is porcine skin [74]. The skin and bones of pigs are utilized as a collagen sources due to some advantages. Since porcine collagen is almost similar to human collagen, it does not cause much allergic response when used in health applications. But just like the bovine source, the zoonotic diseases poses a risk of contamination and pigs are proscribed due to religious reasons [60]. Halal certification of collagen derivatives is considered to be of main importance because of beliefs and it depends on the origin of raw materials used in its manufacture and traceability from the sources until product chain. Muslims and Jew people demand Halal-certified products for their needs, which is not prohibited and obtained by entirely traceable product chains. Nonspecific collagen is highly suspected of containing porcine elements and very strongly discouraged for use by the Muslims [75]. Nonetheless, adult porcine dermis and small intestinal mucosa are used for tendon regeneration, hernia repair, skin and wound healing, and plastic and reconstructive surgery [76].

There are some other sources of gelatin, somehow industrially applicable or not. Throughout the decade, huge numbers of fish species were investigated as alternatives to the source of collagen. Bones, skin, fins and scales of fresh or salt water fishes are mainly used for collagen procurement and gelatin extraction having different chemical composition. This in turn helps to reduce environmental pollution as considerable amount of wastes occurs during fish processing [77]. Collagen studies from marine origin are carried by on marine vertebrates and invertebrates [78, 79]. Marine sources are from some marine species such as fishes, starfish, jellyfish, sponges, sea urchin, octopus, squid, cuttlefish, sea anemone and prawn [80–82]. Some of the raw material sources of collagen peptides are given in **Figure 2**.

Collagen peptides can also be produced for research purposes in small quantities from other animal body parts such as eggshells, rat-tail tendons, frog skin, kangaroo tails, chicken and duck feet, sheepskin, poultry animal skin, feet, bones and many more [46, 83, 84].

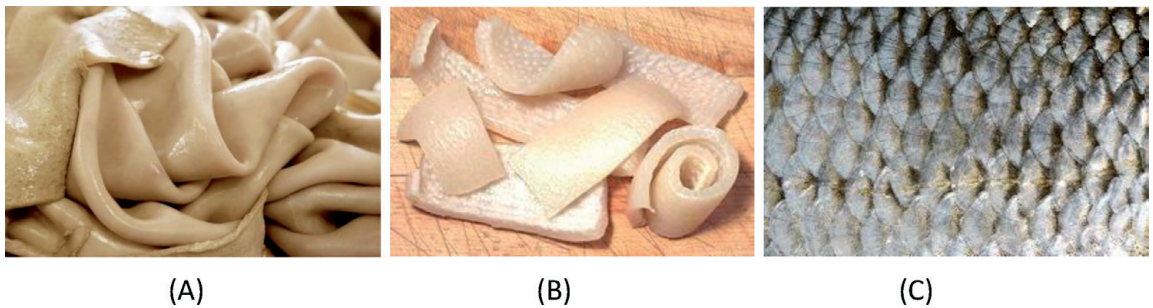


Figure 2.
Main raw material sources of collagen peptides. (A) Bovine split, (B) pig skin, and (C) fish skin.

4. Collagen peptide processing

The collagenic substances, which are involved in multiple collagen units (not subunits) in the quaternary structure and arguably misdefine the tertiary structure also, are normally processable structures. As known very well, leather manufacturing can convert low-value raw materials, which unless untanned and disposed of have detrimental effect to the environment, into valuable final products, and collagen is one of the most substantial structural protein, economically and biologically renewable material for processing. The wastes and by-products of leather processing are discarded parts and effluents from many steps, which are still valuable due to their composition. Lime splits and scraps, as ideal substances, go for gelatin and collagen peptide production.

In the production of industrial-scale collagen peptides, different animal's skin and bones that are easily available and contains collagen protein in high proportion are being used. Collagen peptide preparation steps are dependent on final products' properties. For the first step in general practice, acid and alkali extraction methods are used to remove noncollagenous components [85].

Enzymatic and chemical hydrolysis can be used in the extraction of collagen. Being affordable, chemical hydrolysis is the most commonly used method in industrial practices. Enzymatic hydrolysis is fast and produce waste in minimal amounts, but they are more expensive to carry out [86].

Collagen peptides can be produced by sensitive enzymatic reactions according to the desired molecular weights from collagen-rich raw materials by using protease enzymes. Depending on enzyme types and hydrolyzation conditions, final products can further differ with regard to molecular weight distribution [87]. The production processes could be optimized to obtain different peptides with different functionalities. In the structural level, the cleavage of triple helix is emerged and the collagen molecule is partially broken up. Long chains are hydrolyzed to form shorter chains and further hydrolysis leads to short peptides, some of which are bioactive with body-stimulating functions [88].

Chemical methods of collagen hydrolysis are carried out by means of strong acidic and highly alkaline conditions. Acid and alkaline hydrolysis methods are cost-effective and operation is simple. They have short hydrolysis time and are applicable to industrial processes [89]. However, the uses of strong acids or strong alkaline chemicals make the hydrolysis process environmentally unacceptable [90]. During the acidic treatment, the raw material is exposed to acid for a certain period of time. As this process occurs at a controlled temperature, the structure of the skin swells to twice or thrice more than its initial volume. Both organic acids such as acetic and citric acids and inorganic acids such as hydrochloric acid can be used during acid treatment; however, organic acids are more efficient for the purpose. Acidic treatment results unraveled the structural unity and the cleavage of the noncovalent inter- and intramolecular bonds. Materials with less intertwined collagen fibers such as fish and porcine skin are the preferred choice for the acidic process [85]. For the alkaline process, the raw materials are treated in basic solutions for a duration of a few days to weeks. The most commonly used process is through aqueous sodium hydroxide and calcium hydroxide solutions. However, other basic solutions can also be used in this alkaline process. This process entails the treatment of hard or thick substance that needs very aggressive penetration by the basic solutions [91].

To meet the technical needs of the different sectors, purification stage ensures the removal of ionic and nonionic impurities resulting from the processing of raw materials. Different filtration and purification systems can be used at this stage depending on the final product needs [92]. The purified and demineralized gelatin solution consists of over 95% water. This water has to be almost completely

removed. Only dried gelatin with its normal residual water content of 10–12% has an unlimited shelf life from the microbiological point of view. In addition, dilute gelatin solutions can neither be stored nor transported easily. In the next production step, the highly concentrated and filtered gelatin solutions are sterilized. For this step, both indirect sterilization via plate heat exchangers and direct steam sterilization are used. Both methods are microbiologically safe to a very high degree [93]. After sterilization, the prepared material needs to be dried to final form. There are different drying methods used in the production of collagen peptides. Spray drying is the most commonly used method and widely used in the production of small molecular weight peptides [94].

5. Industrial applications

5.1 Food

Collagen peptides have shown to be an important ingredient in the food and beverage industries worldwide [95]. It has been used for a long time in foods globally, such as in the United States, China, Japan and many countries in Europe. Approved as Generally Recognized As Safe (GRAS), the safety of collagen peptides has been affirmed by the Food and Drug Administration (FDA) and Center for Food Safety and Applied Nutrition (CFSAN) [96]. It has been applied as protein dietary supplements, carriers in the meat processing, edible film and coatings of products and food additive to improve product's functionality [97]. In addition, collagen may boost the health and nutritional value of the products relying on its inimitable properties on human bodies [75].

The source of the raw material and the degree of processing determine the properties of the collagen peptides like gelatin, which have several different applications in the food industry [98]. The major quality parameters are their higher gel strength and suitable melting and gelling temperatures for the food industry that uses them as an additive. Due to the fact that porcine and bovine gelatins are less preferred due to religious preferences, safety concerns and economic considerations, using fish skin or bones to obtain gelatin has become popular in recent years [99]. Thanks to its many unique properties, the numerous applications of gelatin include its usage as a thickener, stabilizer, setting agent, clarifying agent, water-retaining agent and adhesive in a wide range of foods, pharmaceuticals and household products. In the food industry, gelatin can be utilized in a wide range of confectioneries, beverages, snacks, desserts and meat products [100]. Gelatin is used as an additive to improve elasticity, consistency and stability of foods like desserts, candies, bakery products, jellied meats, ice cream and dairy products. Gelatin is also used as stabilizer to modify the structure of the food products. It is added to yogurt to reduce syneresis and increase firmness [100]. In addition, type A gelatin that is isolated with acid treatment with gel strength as 70–90 g, which is relatively low, is used to fine wines and juices. Type B gelatin is processed with an alkali treatment with gel strength as 125–250 g and is used in confectionery products [101]. Collagen peptides have also been reported to have antioxidant and antimicrobial activity [102]. However, the relationship between peptide characteristics and antimicrobial activity has not been clearly demonstrated.

5.2 Cosmetic

Collagen can be used in cosmetics due to its biodegradability, availability and biocompatibility properties for different purposes such as in dermal fillers, skin substitutes or scaffolding, wound repairs and facial products [103].

The formation of unwanted wrinkles in the body with aging is related to the damage of the fibers in the skin. In the researches about aging, it has been determined that collagen hydrolysates contribute greatly to the repair of these fibers [104]. The introduction of collagen hydrolysate into the body ensures the stimulation of collagen formation that enables the recovery and improved tissue appearance [105]. Hence, the cosmetic industry reclaims some functionalities of its products by incorporating this biomolecule.

Collagen peptide has been known to be used in cosmetic formulations for reasons such as protecting the structure and the function of the skin, enhancing its appearance and preventing premature aging [106].

Collagen peptide is prepared in the form of liquid ampoules, powder mixes or tablets in the food and cosmetic industries. It has a regenerative effect on skin wrinkles and other signs of skin aging: collagen helps the skin remain soft and pliant and improves the hydration of the epidermis [107]. Many studies have shown that collagen sleek thin lines and can prevent the development of deeper wrinkles and grooves. Collagen is not only effective for the skin on the face but also stimulates the fiber structure of the body to repair and reduce cellulite tissue [108].

Collagen hydrolysates have also shown bioactivities such as antioxidant properties, antihypertensive activity, lipid-lowering activity, as well as reparative properties in damaged skin [109]. Moreover, it has been also observed that collagen provides the building block for elastin and collagen formation and acts as ligands in fibroblast cells to stimulate hyaluronic acid [110].

5.3 Health

Collagen is the most abundant and ubiquitous protein in the body regarded as one of the most useful biomaterials. The excellent biocompatibility and safety due to its biological characteristics made collagen the primary resource in medical applications. It has various applications in some departments such as cardiology (heart valve), dermatology (for skin replacement, augmentation of soft tissue, skin tissue engineering and artificial skin dermis), surgery (as hemostatic agent, wound repair and dressing, nerve repair and blood vessel prostheses), orthopedy (tendon, bone and ligament repair and cartilage reconstruction), ophthalmology (corneal grafts and contact lenses), urology (hemodialysis and sphincter repair) and vascular surgery (vascular graft and vessel replacement) [111].

Collagen type I is considered to be the most valuable material for tissue engineering due to its high biocompatibility and immunogenicity. It is used as the basic matrix for cell culture [73, 112]. Biomaterials based on collagen are widely used in tissue engineering such as injectable matrices and scaffolds intended for bone regeneration [73, 113]. Moreover, collagen-based eye implants are preferred for the treatment of ophthalmic disorders. Such type of collagen-based implant preparation has shown considerable applicability because it provides stable and reasonable control over the postoperative complications such as intraocular pressure [114]. Collagen-based matrices find their use as corneal transplant and as temporary patches to repair perforations in case of emergencies [115].

Collagen is used in pharmaceutical industries for different functionalities as hard and soft dry capsules, microparticles, injectable dispersions, shields in ophthalmology sponges and drug delivery system. Its application in the pharmaceutical as well as biomedical field is due to its characteristics such as weak antigenicity, immunogenicity, biodegradability and biocompatibility [116].

As a collagen peptide, gelatin is the most important material for the production of hard and soft capsules as well as film-coated and effervescent tablets. Manufacturers take into account its adhesive, gelling and film-building properties.

Orally administered medicines and dietary supplements in particular are protected by gelatin-containing capsules or tablets from light, moisture and oxygen and given a long shelf life [107, 117, 118]. Gelatin is also used as a raw material in many field of health industry as is the case with the manufacture of blood substitute [119]. These products prevent hypovolemic shock by stopping bleeding in the wound-occurred area. As local hemostatic agents, collagen sponges and films have long been used in the surgical field (e.g., in oral cavity and ophthalmological surgery, urology or gynecology) and for the treatment of wounds in dental surgery. The structural composition of the collagen material enables the absorption of large amounts of blood and makes it possible for new tissue to grow into the sponges. Since it only takes a few days for the body to completely resorb the sponges or films, they can be left in the wound without any negative effects [107, 120, 121].

5.4 Sportive nutrition

Collagen peptides are ideal supply due to their numerous beneficial health effects for modern sportsperson nutrition as high-energy supplement to maximize muscle protein anabolism [122]. They are neutral in flavor, which means that they do not leave a bitter aftertaste that has to be masked in the final product, that is, through sugar or artificial sweeteners, as is often the case with soy, whey or other protein [107, 123]. Collagen peptides have been scientifically tested and have no undesirable side effects, and there is no evidence to elicit allergic reactions. It emulsifies foams and improves the shelf life of products [107, 124].

The more protein a body expends through physical exertion, the greater its needs for an external source, for example in the form of special dietary supplements such as protein shakes, energy bars, protein snacks or sports drinks. Several studies in the past few decades have reported that protein hydrolysates from various food sources, in addition to their nutritional properties, exhibited various biological functions including hypotensive activity, anticoagulant, cholesterol-lowering ability and hypoglycemic effect [125]. Consumption of hydrolyzed collagen increases collagen synthesis and decreases knee pain while standing and walking [126]. Shaw et al. [127] tested the role of gelatin consumption in collagen synthesis. In the study, double-blinded, placebo-controlled and crossover-designed research, subjected to whom consumed 15 g of gelatin showed double-fold collagen synthesis, measured through serum propeptide levels. From the results, it was observed that consuming hydrolyzed collagen might increase collagen synthesis and potentially decrease injury rate in athletes. Studies have also shown that products fortified with collagen peptide can promote joint health, bone synthesis and antisport fatigue ability [128].

5.5 Agriculture and animal feed

5.5.1 Fertilizer

Leather processing wastes like shavings that cause environmental pollution are opulent sources of novel and valuable biomolecule “collagen” [129]. Industry has been generally oriented on the recovery of collagen from leather waste, but the remaining waste also can be used for agricultural purposes. Collagen-based fertilizer products highly are demanded in agriculture industry because of being high amino acid and organic carbon source and nitrogen content [130].

The collagen hydrolysates obtained from leather wastes are being utilized as bio-fertilizer. Several plants can also take up and absorb amino acids as an example of biostimulants; these amino acids are sometimes better nitrogen sources than ammonia or nitrates [131]. Collagen peptides are recovered and channeled as an organic

nitrogenous fertilizer to increase the yield of the crop [132]. Both plants and animal organisms can more easily absorb microelements like iron, copper, zinc, calcium, magnesium and manganese chelated with hydrolyzed collagen. The use of collagen hydrolysates in combination with potassium polyphosphates increases agricultural production by increasing the absorption of phosphorus and potassium [133].

Collagen hydrolysates obtained by chemical and chemical-enzymatic processes under moderate reaction conditions were used in a study for preparation of foliar fertilizers [134]. Hydrolysates of chromium-tanned leather shavings were used in a study as nitrogen source for growth of common bean plants and banana cultivation [135]. De Oliveira et al. [136] have studied the use of leather wastes after extraction as a nitrogen source to elephant grass. The chrome shaving wastes can also be hydrolyzed in an autoclave (150°C). The obtained product contains moisture content (7–10%), total nitrogen (10–11%), organic carbon (40%) and chromium (III) (2.5–3%). By blending with other additive components, the product can be sold as a fertilizer [133].

Both gelatin and collagen hydrolysates have positive effect on the growth of plants when applied as fertilizer. The crop yield is comparable with those obtained by using inorganic fertilizers but with a significantly high value in view of the low nitrate content, which is 20 times less. Besides, organic fertilizer improves the soil quality unlike the inorganic ones [137].

5.5.2 Animal feed

Collagen peptide due to its organic compounds such as fats, proteins and minerals plays an important role in the preparation of highly valuable animal feed [84]. Fat, protein and mineral products are in especially high demand in the animal feed industry because pure fats are excellent sources of energy and collagen is of importance for the healthy growth of animals [138].

As a collagen peptide, which is recovered from leather solid wastes, gelatin is primarily added to animal feed based on its hydrophilic properties. Its jelly-like consistency holds feed together, making it transportable and extends its shelf life [107, 139]. When animal feed is enriched with vitamins, the gelatin coatings also protect these from light and oxygen. A positive side effect of adding gelatin to feed ensures that the fur of animal remains wonderfully glossy [107, 140].

There are also many fields for collagen peptides and gelatin usage. They can be used for photography and X-ray films and inkjet applications [141], industrial paper production [93], leather board [142], glue manufacture [143], feedstock for bio-diesel production [144], leather tanning and retanning agent [145] and many more specific applications, etc.

6. Conclusion

The tanning industry is one of the oldest industries in the world and recently its pollution load onto environment has become seriously threatening for transferring the potential to next generations. It produces a significant amount of solid wastes and effluents. It is a well-known fact that removing undesired substances out of the structure in leather processing produces effluents; that is highlighted agenda which needs to be overcome and as per the composition those are able to handle for recover and reuse through the current technology.

Revaluation of leather solid wastes is one of the promising waste management strategies that provides raw materials to another industry such as food, agriculture, cosmetic, health, etc. This method may offer a solution for utilization of huge volume of leather solid wastes, which are often dumped in open landfills. Commercial

benefits of the system should be linked with both the value of the products and the disposal cost of solid wastes. In the economical point of view, feasibility should be based on converting them into value-added products instead of making a deposit for disposal.

Collagen peptides obtained from hides and their by-products have been practiced as healthful stuffs in many areas of our modern life. As the awareness of their technological value increases as time passes, this value-added material is considered to have higher interest with the usage in various fields. Analogous to collagen peptides, collagen hydrolysates and gelatins, emerged from this precious protein are involved in either partly or total denaturation. It is the process defined by disintegration of intra- and intermolecular bonds that keeps together the chains composed of amino acids in the conformation; thereby, a typical protein is formed. The discovery of benefits of collagen derivatives for health and their usage as additives has a long history and is dated back to some 8000 years ago. Today, its usage enlarges over many industries and applications includes in food, health, chemical, body care and agricultural etc. According to the molecular weight and properties, the usage of collagen derivatives increases as a gel or colloidal solution, their benefits are multiplied and this bio-based material supplies many valorization possibilities. As per the source and properties, they are bioavailable products that are digested and absorbed by human and animal body quickly and even by plants and are also easy to use in any industrial applications and processes.

In spite of the tremendous development in technology and sciences, there are still challenges ahead to better understand the collagen types and sources, structure and properties, gelatin processes and product characteristics. It seems that in the future the researches on bio-based materials as well as the efforts for their commercialization will continue intensively in a wider range of products.

Conflict of interest

The authors declare that they have no conflict of interest.

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