

## A REVIEW OF GELATINE: MULTIFUNCTIONAL ADDITIVES IN THE FOOD INDUSTRY

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### ABSTRACT

Gelatine is a significant hydrocolloid used widely across various industries, including food production, pharmaceuticals, and photography. It is classified as a water-soluble material obtained through the partial hydrolysis of collagen sourced from animal skin, bones, and other connective tissues. The primary raw materials are derived from pigs and bovines, raising concerns related to religious beliefs and health considerations. As a result, alternative sources of gelatine – such as those derived from fish, insects and poultry (e.g., chicken) – have been proposed to replace pig-derived gelatine. Gelatine possesses several useful properties based on its structure, including gel formation, protective colloid functions, stabilising and emulsifying capabilities, as well as adhesive and cohesive actions. With gelatine production expected to increase in the coming years, researchers are actively exploring ways to modify its properties and combine it with other substances to expand its applications. This review aims to deliver an in-depth analysis of gelatine, focusing on its basic structure, source of raw materials, production methods, and current uses in the food industry.

**Keywords:** food additives, gelatine, gelatine properties, hydrocolloid

### INTRODUCTION

Gelatine is a natural, soluble fibrous protein derived through the partial hydrolysis of collagen from various animal by-products. Collagen constitutes approximately 20–30% of the total protein content in animals and is found in bones, tendons, cartilage, connective tissue, and the cornea of the eyes (Vergauwen et al., 2017; Baratta et al., 2022; Tarnutzer et al., 2023). Key raw materials used in gelatine production include pig-skins, pig bones, bovine hides, and fish skins.

Gelatine has a wide range of applications across multiple industries, including food and beverage services,

pharmaceuticals, and photography. In the food and beverage industry, it serves as a stabilising, emulsifying, foaming, or gelling agent, commonly used in products such as ice cream. In pharmaceuticals, gelatine is utilised for producing soft and hard gelatine capsules. Additionally, in photography, it has been historically employed to coat light-sensitive chemicals onto film supports (Caxito et al., 2018). Gelatine is also combined with other materials, such as alginate and chitosan, to enhance its functionality in diverse applications (Kreller et al., 2021; Qiu et al., 2022).

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Despite its numerous advantages, the use of gelatine is associated with certain drawbacks, primarily due to health concerns and religious beliefs. For instance, in predominantly Islamic countries, gelatine derived from pigs or animals not slaughtered in accordance with Islamic procedures is prohibited (Mian and Muhammad, 2003). Furthermore, bovine-based gelatine has raised concerns due to its potential link with bovine spongiform encephalopathy (BSE), commonly known as “mad cow disease” (Kumagai et al., 2019).

However, the United States Food and Drug Administration (FDA) permits the production of gelatine from bovine hides under specific conditions. Processors must ensure that hides have not been in contact with the brain, spinal cord, or ocular tissues of cattle from countries with a higher than negligible risk of BSE. Additionally, hides from cattle exhibiting signs of neurological disease must not be used (European Union, 2000).

To address these issues, alternative sources for gelatine production, such as fish, poultry and insects have been proposed (Boran and Regenstein, 2009; Sarbon et al., 2013; Mariod and Fadul, 2014).

This review aims to provide comprehensive information on gelatine’s structure, properties, raw material sources, and production processes. Additionally, its applications in food production are discussed to highlight its benefits across various fields.

## STRUCTURE AND FUNCTION OF COLLAGEN

Collagen’s partial hydrolysis produces the polypeptide chain known as gelatine. Therefore, understanding the structure and function of collagen is essential for understanding gelatine. Collagen appears opaque and white, with non-branching fibrils embedded in a matrix of mucopolysaccharide and other proteins (Balian and Bowes, 1977). The type of tissue and the age of the animal from which the collagen is sourced can influence the number of fibrils present. Collagen from young animals has fewer stable cross-links, making it easily solubilised in water (Muyonga et al., 2004; Suurs and Barbut, 2020; Sorushanova et al., 2021). As the animal matures, the solubility decreases as the labile collagen structure is continuously converted into a more stable, cross-linked structure, which makes it insoluble in water (Schrieber and Gareis, 2007a). This process also

reduces collagen’s ability to bind to water molecules, as its primary function is to provide structural support for extracellular spaces between the connective tissues (Wu et al., 2021).

Collagen has a unique amino acid arrangement, marked by two modified imino acids: proline and hydroxyproline. Collagen contains only a small percentage (2%) of hydroxyproline, which is also found in elastin (Poppe, 1997). Additionally, collagen lacks amino acids such as tryptophan and has a minimal amount of cysteine and methionine (Paul et al., 2019). The molecular structure of both collagen and gelatine is primarily composed of multiple repetitions of the “Glycine-X-Y” amino acid sequence, where “X” is usually proline and “Y” is almost always hydroxyproline (Smith and Rennie, 2007; Boran and Regenstein, 2010; Goldberga et al., 2018). Glycine, proline and hydroxyproline account for 56% of the composition in gelatine with the remaining 44% consisting roughly of alanine, arginine, aspartic acid and glutamic acid. An exception occurs at the N-terminal residue of gelatine, where alanine predominates in acid-processed gelatine, while glycine is more common in alkali-processed gelatine (Karim and Bhat, 2009; Julie Chandra et al., 2022). Carbohydrate units, such as the monosaccharide galactose or disaccharide glucosylgalactose, may also be present in collagen and linked to hydroxylysine residue (Schrieber and Gareis, 2007a).

The primary structure of collagen is described as a triple helix, consisting of three left-handed  $\alpha$ -chains: two  $\alpha 1$ -chains and one  $\alpha 2$ -chain. Each turn in the chains contains three amino acid residues (Nelson and Cox, 2004; Boran and Regenstein, 2010). Each  $\alpha$ -chain comprises 1014 amino acids, with a total molecular weight of 100 kDa (León-López et al., 2019). The left-handed helix  $\alpha$ -chain is formed due to the presence of proline and hydroxyproline, which help the chains turn and stabilise the secondary structure of the single helix (Haug and Draget, 2009). Additionally, glycine is also required in every third residue of each chain, as it is responsible for the helical structure (Brigham, 2018).

The  $\alpha$ -chains are then super-twisted around each other to form a right-handed superhelix, which appears as a rigid, rope-like structure (Nelson and Cox, 2004; Vergauwen et al., 2017). This right-handed superhelix is also known as a tropocollagen molecule

and represents the basic building block of collagen molecules. Tropocollagen molecules have a molecular weight of 300 kDa, 1.4–1.5 nm of diameter and a length of about 300 nm (Rich and Crick, 1955; Balian and Bowes, 1977; Haug et al., 2004). Tropocollagen molecules are then chemically linked together to form fibrils. Collagen fibrils are formed when the fibrils align alongside each other, with each fibril shifted by one-quarter of its length relative to its neighbouring fibrils (Poppe, 1997; Shoulders and Raines, 2009). These fibres are stabilised by the formation of covalent cross-links between neighbouring collagen molecules (Eyre et al., 1984).

As collagen is hydrolysed to form gelatine, collagen fibres and fibrils dissociate into tropocollagen molecules as the hydrogen bonds and hydrophobic bonds that stabilise the structure are broken. After dissociation, the hydrolysis of tropocollagen molecules

yields three different forms: the independent  $\alpha$ -chain,  $\beta$ -chain and  $\gamma$ -chain. These chains differ in molecular weight, with the  $\alpha$ -chain having a molecular weight of 80–120 kDa, the  $\beta$ -chain 160–250 kDa, and the  $\gamma$ -chain 240–375 kDa (Poppe, 1997; Tu et al., 2015; He et al., 2022). This information is important for the manufacturers, as it helps them control the hydrolysis process to achieve the ideal molecular weight for the intended application. For example, gelatine viscosity is related to its molecular weight and the lower the molecular weight of gelatine, the lower its viscosity will be (He et al., 2024).

It is important to note that different animal species, from which collagen is derived, exhibit varying amino acid compositions, which may affect their respective properties, such as melting point, setting temperature and gel strength (Table 1) (Haug and Draget, 2009; Boran and Regenstein, 2010). Gelatine exhibits

**Table 1.** Amino acid composition of gelatine from bovine, porcine, chicken, fish and insect gelatine

Amino Acid	Bovine skin	Porcine skin	Chicken skin	Tilapia (warm-water fish)	Alaska pollock skin (cold-water fish)	Black soldier fly larvae (insect)
1	2	3	4	5	6	7
Hydroxyproline	83	91	121	79	55	1.77
Aspartic acid	46	46	21	48	51	102.52
Threonine	33	18	10	24	25	35.81
Serine	39	35	22	35	63	26.27
Glutamic acid	74	72	58	69	74	152.45
Proline	127	132	134	119	95	82.49
Glycine	342	330	337	347	358	46
Alanine	113	112	101	123	108	192.93
Valine	19	26	19	15	18	43.04
Methionine	4	4	7	9	16	11.51
Isoleucine	11	10	12	8	11	29.17
Leucine	24	24	26	23	20	38.47
Tyrosine	4	3	12	2	3	54.09
Phenylalanine	12	14	18	13	12	30.44
Histidine	4	4	30	6	8	32.36

**Table 1 – cont.**

1	2	3	4	5	6	7
Lysine	25	27	47	25	26	70.84
Arginine	47	49	56	47	51	30.52
Hydroxylysine	5	6	N/A	8	6	N/A
Cysteine	0	0	1.6	0	0	19.26
Tryptophan	0	0	0	0	0	N/A
Total imino acid	210	223	255	198	150	84.26
Total residue	1 012	1 003	1 032.6	1 000	1 000	999.94
Reference	Gómez-Estaca et al., 2009	Eastoe and Leach, 1977	Sarbo et al., 2013	Sarabia et al., 2000	Kimura and Ohno, 1987	Chua et al., 2023

properties related to both proline and hydroxyproline, as previously mentioned. For instance, proline and hydroxyproline play an important role in gelling strength, as the high amounts of these two imino acids cause the gelatine gel to have higher gel strength due to the increased hydrogen bonding within the network (Cho et al., 2006). Fish gelatine, particularly from cold-water fish, has a lower amount of proline and hydroxyproline, making it less effective in gelling compared to other sources (Haug et al., 2004). For example, hydroxyproline in codfish was only observed at 53 residues per 1000 whereas pig skin contains 90.7 residues out of 1000 (Eastoe and Leach, 1977).

Alternatively, warm-water fish gelatine, such as that derived from carp skin, shows a slightly similar proportion of proline and hydroxyproline to mammalian gelatine. As reported by Ninan et al. (2014), gelatine derived from carp skin contains imino acids ranging from 19.16% to 20.86%, which is slightly lower than found in bovine (22.91%) and pig gelatines (23.7%).

Poultry-based gelatine has also been shown to have a higher imino acid content compared to bovine and porcine gelatines. Additionally, the high alanine content in chicken skin gelatine may enhance its high viscoelastic properties by facilitating the formation of a triple-helix structure and stabilising the gel at lower temperatures (Sarbo et al., 2013).

On the other hand, gelatine extracted from insects contains relatively low levels of imino acids compared to other species (Mariod and Fadul, 2014; Chua et al., 2023).

## TYPES OF COLLAGENS AND ITS IMPORTANCE TO THE GELATINE INDUSTRY

There are nearly 28 types of collagens, with type I being the most common (Ricard-Blum, 2011; Makareeva and Leikin, 2014). Type I gelatine is found in various parts of animals, including bones, skin, tendons, ligaments, and organs (León-López et al., 2019; Naomi et al., 2021). Type II collagen is exclusively located in cartilage, while type III collagen is commonly present in muscles, the walls of blood vessels, and skin (Cole, 2003; Pelley, 2007; Khoshnoodi et al., 2008; Xu et al., 2023). Type IV collagen is found in the basal lamina, an epithelium-secreted layer of the basement membrane. In contrast, type V collagen is abundant in the corneal stroma, interstitial tissues of the liver, lungs and placenta (Khoshnoodi et al., 2008; Leeming and Karsdal, 2019). These types of collagens differ in several ways:

1.  **$\alpha$ -chain composition**
2. **The repeat and length of “Gly-X-Y” amino acid repetition**
3. **The presence or absence of interruptions in this sequence**
4. **The occupation of proline and hydroxyproline at the X and Y positions, respectively** (Ricard-Blum, 2011; León-López et al., 2019).

Since gelatine is primarily produced from the skins and bones of animals, type I collagen is the predominant type used in its production. However, type II

collagen may also be present if the raw materials include cartilage parts of the animals.

## MECHANISM OF GELATION IN GELATINE

Gelation is a chemical reaction in which macromolecular chains link to form a branched polymer structure that traps and immobilises the liquid, creating a rigid framework (Lewis, 1996; Redaelli et al., 2017). The resulting structure varies in solubility, properties and appearance depending on the chemical nature of the starting materials. The mixture of soluble branched polymer and water is referred to as colloidal solution or sol. As the chain continues to link, a molecule spanning the entire system begins to form. These molecules, called networks or gels, consist of several branched polymers. The transition from finite molecules to infinite molecules is known as the sol-gel transition or gelation process. The point at which the gel begins to form is identified as the gel point.

Gelation can be induced through either physical or chemical linking. Physical linking is a reversible process in which a polymer solution transitions into a gel (de Carvalho and Djabourov, 1997). This type of gelation is exemplified in gelatine, where forces, such as molecular interactions, secondary interactions, or a combination of both, facilitate the polymer network connection. The linking in physical gelation is reversible and does not require additional initiators or chemical cross-linkers, which can be toxic.

Gelatine gelation involves dissolving gelatine in water at approximately 40°C, forming a colloidal solution. Upon cooling, the sol-gel transition occurs, leading to the formation of a cross-linked gel. During this process, gelatine regains the triple helix structure of collagen as some cross-links are established. The solution transitions into a gel at the gelling temperature ( $T_g$ ) and melts at the melting temperature ( $T_m$ ). When comparing  $T_g$  values across species, mammalian gelatine exhibits the highest  $T_g$ , followed by warm-water fish and cold-water fish (Meng and Cloutier, 2014).

Interestingly, the  $T_m$  of gelatine is (27–35°C) significantly lower compared to other gelling agents, such as gellan gum (~70–80°C) and agar (85°C) (Schröder, 2003; Williams and Phillips, 2003; Cui et al., 2013). This lower  $T_m$  makes gelatine a preferred gelling agent

in food products, as it enables easier flavour release at temperatures below normal human body temperature.

## RAW MATERIALS OF GELATINE

The raw materials used in gelatine production globally are estimated to come from 45% pig skin, 30% cattle hide, and 23% cattle and pig bones (Karim and Bhat, 2009; Boran and Regenstein, 2010). Other sources, such as fish and chicken, contribute only around 1.5% of annual gelatine production (Boran and Regenstein, 2010). In Europe, however, 80% of edible gelatine is derived from pork skin, with 15% from porcine and cattle bones, and 5% from fish (Gelatine Manufacturers of Europe, n.d.).

The widespread use of pork, and to a lesser extent, cattle in gelatine production raises concerns related to health, religious beliefs, and dietary restrictions. For instance, Muslims are prohibited from consuming pork, while Hindus are restricted from cattle products, both due to their religious beliefs. Additionally, due to the historical link between bovine-based gelatine and BSE outbreaks, some individuals are hesitant to consume products containing bovine-based gelatine (BIOHAZ et al., 2020). However, the FDA has confirmed that bovine-based gelatine is safe to consume as long as strict guidelines are followed (European Union, 2000).

In response to these concerns, alternatives to pig and bovine-based gelatine have been explored, such as fish, poultry and insect-derived gelatines (Sarbon et al., 2013; Mariod and Fadul, 2014; Abedinia et al., 2020; Qiu et al., 2022; Chua et al., 2023).

Fish gelatine offers several advantages over mammalian gelatine, including a lower melting point, and reduced gelling power. These characteristics make it particularly useful in applications where easy flavour release is desired, improving sensory qualities (Boran and Regenstein, 2010). Furthermore, fish gelatine is accepted by all religious groups and carries no disease risks associated with bovine gelatine, such as those linked to BSE outbreaks in the past. Additionally, fish processing can generate by-products, such as skin, tongue, stomach, liver, scales and cheeks, which can constitute up to 75% of the total weight of the catch and are ideal for gelatine production (Shahidi, 1994). Fishes species studied for gelatine production include

tilapia (Gong et al., 2024; Li et al., 2024), carp (Ninan et al., 2014; Tkaczewska et al., 2018; Xu et al., 2021), and codfish (Alves et al., 2022).

Poultry-based gelatine, particularly from chicken, duck and goose, has also been studied as an alternative to pig and bovine gelatine. Poultry gelatine is typically extracted from the head (Ee et al., 2021), feet (Kuan et al., 2016), skin (Kim et al., 2020), and bones (Qiu et al., 2022). Gelatine derived from chicken and duck has similar properties to mammalian gelatine, with higher levels of proline and hydroxyproline compared to pig-based gelatine (Abedinia et al., 2020), resulting in comparable gel strength. Additionally, chicken skin is used in gelatine film production due to its superior thermal properties and higher gel strength compared to cattle-based gelatine (Sarbon et al., 2013).

Insect-derived gelatine has recently gained attention due to its low cost, high availability, and inclusion of essential amino acids (Tang et al., 2019). Insects such as melon bug (*Coridius vidutus*), grasshoppers (*Sphegnarium histrio*) and silkworm pupae are consumed in Sudan, Thailand, and China, respectively (Mariod and Fadul, 2014; Tang et al., 2019). Studies on insect gelatine, such as those by Mariod and Fadul (2014), which focused on the melon bug (*Coridius vidutus*) and sorghum bug (*Agonoscelis pubescens*), as well as Chua et al. (2023), who worked with black soldier fly larvae (*Hermetia illucens*) for gelatine production, have found that insect gelatine contains lower levels of key amino acids, particularly glycine, proline, and hydroxyproline compared to mammalian gelatine. This difference may influence the chemical and functional properties of the gelatine, as these amino acids play a crucial role in the gelatine structure and gelling ability.

## PRODUCTION OF GELATINE

Gelatine is produced through the partial hydrolysis of collagen molecules. This hydrolysis reaction can occur through acidic, alkali, enzymatic, or a combination of these methods. The general process of gelatine production begins with cleaning the raw materials, followed by pre-treatment, gelatine extraction, filtration/purification/sterilisation, concentration, drying and milling (Haug and Draget, 2009).

Initially, raw materials are washed to remove impurities. For bony materials, additional steps such as

washing, crushing and rewashing, are involved, resulting in degreased crushed bone chips. These bone chips are then exposed to acidic conditions for several days to remove minerals, leaving behind spongy bone materials known as ossein (Haug and Draget, 2009).

The raw materials are subjected to either acidic or alkali pre-treatment, depending on the collagen source and the required quality of the final gelatine. Notably, no specific guidelines exist for optimal pre-treatment methods for different parts of various species. However, acidic pre-treatment is generally used for pig, fish and poultry species to produce the corresponding gelatine, while cattle species typically undergo alkaline pre-treatment (Baydin et al., 2022). Insect gelatine, on the other hand, lacks a widely adopted pre-treatment method, though some studies report using acidic pre-treatment and hot water extraction to produce gelatine from insects (Mariod and Fadul, 2014; Chua et al., 2023).

### Acidic pre-treatment

In this method, cleaned raw materials are immersed in a diluted, cold mineral acid, such as sulfuric acid or hydrochloric acid, for a period ranging from 8 to 30 hours, depending on the size and thickness of the raw materials, until maximum swelling occurs. (Haug and Draget, 2009; Sántiz-Gómez et al., 2019). The materials are then washed with water several times and neutralised until the required pH for the next extraction step is achieved. Gelatine produced by this method is classified as type A gelatine.

### Alkaline pre-treatment

Alkaline agents, such as calcium hydroxide or potassium carbonate are used in this pre-treatment method. The cleaned raw material is placed inside a container or a pit with the alkaline agent and water at ambient temperature (around 24°C) (Haug and Draget, 2009). The mixture is agitated intermittently using mechanical means. This process can last from 20 days to 6 months, depending on the thickness and type of raw materials used. Ossein (from bone) requires more time than bone hides for this treatment. Once the treatment is complete, the materials are washed with water to neutralise the mixture. Finally, the materials are treated with diluted acids to achieve the correct pH for extraction. Gelatine produced through this method is classified as type B gelatine.

### Enzymatic pre-treatment

The enzymatic pre-treatment method involves the use of proteolytic enzymes such as pepsin, bromelain, or papain to hydrolyse collagen into gelatine (Norziah et al., 2014; Ahmad et al., 2019; 2021). This method has the advantage of shorter processing times compared to acidic and alkaline pre-treatments, but it generates higher production costs and less waste (Rather et al., 2022). The production is summarised as follows: the raw materials are cleaned to remove unwanted materials and then broken down into smaller pieces. They are immersed in an alkali solution such as 0.1 M (w/v) sodium hydroxide (NaOH), for several hours to remove non-collagenous proteins (Roy et al., 2017; Ahmad et al., 2021). After thorough washing with water to remove alkali traces and achieve a neutral pH, the materials are treated with mineral acid (e.g. hydrochloric acid) and washed with water again. The materials are then immersed in a proteolytic enzyme solution, such as pepsin, at a specific concentration in a controlled environment for several hours. The mixture is then incubated at a high temperature (90°C) for several minutes to inactivate the enzyme (Ahmad et al., 2021). The resulting mixture is ready for the gelatine extraction process.

### Extraction of gelatine to final gelatine product

After the raw materials undergo the pre-treatment of choice, they are placed in a vessel and covered with hot water. A series of extractions with hot water (usually 3 to 5 or 6 times) is performed, with each successive extraction occurring at a temperature of 5°C to 10°C higher than the previous one, ranging from 55°C to 100°C (Haug and Draget, 2009). The combined pre-treatment and extraction processes result in a gelatine mixture consisting of polypeptide chains with varying molecular weights and compositions. The three primary fragments observed in gelatine are:  $\alpha$ -chain,  $\beta$ -chain (two  $\alpha$ -chains covalently linked), and  $\gamma$ -chain (three  $\alpha$ -chain covalently linked) (Goudie et al., 2023).

The initial extraction, conducted at lower temperatures produces high-quality gelatine with a lighter colour, higher molecular weight, viscosity and gel strength due to less hydrolysis of the polypeptide backbone (Haug and Draget, 2009). Later extractions, which occur at higher temperatures, lead to more depolymerized gelatine with lower molecular weight,

reduced gel strength, and a darker colour. The colour of the gelatine is influenced by the Maillard reaction, where the carbonyl group of carbohydrates reacts with the amine group of amino acids in gelatine, forming conjugates. This leads to the formation of a Schiff base, which undergoes cyclization to form Amadori compounds. These compounds eventually lead to the production of coloured, insoluble polymeric compounds known as melanoidins, which cause the darker colour of gelatine (Kchaou et al., 2019). The ash content of gelatine at this stage is approximately 2–3%, and if lower ash content is required, excess salt is removed from the gelatine through ion exchange.

The gelatine solution is then concentrated, filtered and sterilised. The sterilisation is conducted using either direct steam sterilisation or the plate heat exchange method (Haug and Draget, 2009). After sterilisation, the solution is cooled to form a gel. For powdered gelatine production, the gel is extruded into “noodles” and placed on a conveyor belt for drying. The drying process involves using filtered, dehumidified and microbiologically clean air. The drying chamber conditions, including temperature and humidity, are carefully controlled, with the starting temperatures typically set at 30°C in the initial zone (Haug and Draget, 2009). Drying times vary depending on the quality, concentration, and thickness of the material and specific conditions used (Hinterwaldner, 1977). Once dried, the gelatine “noodles” are crushed and milled into blends containing particles with a specific diameter range. The moisture content of the resulting blend typically ranges from 8% to 12% (Haug and Draget, 2009).

### IMPORTANT PROPERTIES OF GELATINE

Gelatine appears as a solid, vitreous granule with a slightly faint yellow colour to dark amber, and is tasteless and odourless (Vergauwen et al., 2017; Rather et al., 2022). Commercial gelatine typically has a molecular weight ranging from 40 kDa to 90 kDa (Djagny et al., 2001). In terms of solubility, gelatine is completely soluble in water when the temperature is above 35–40°C (Kramer, 2001). Gelatine is also soluble in other aqueous solutions such as acetic acid and glycerol, but is insoluble in most organic solvents, such as acetone and benzene (Sigma Aldrich, 2020). As gelatine

is a protein, its amphoteric nature, electrically charged groups, and ionisation constants are important to understand and explain its behaviour when it reacts with other substances. Some important properties of gelatine include amphoteric behaviour, gel strength, viscosity, and setting and melting temperatures.

### **Amphoteric behaviour**

Gelatine in solution displays an amphoteric behaviour, meaning it can act as both an acid and an alkali. In a basic environment, gelatine becomes negatively charged, which causes it to migrate toward the anode in an electrically charged field. In contrast, in an acidic environment, gelatine becomes positively charged and migrates toward the cathode. The pH at which the net charge of the intermediate is zero is called the isoelectric point (IEP). The IEP of gelatine depends on the production process and the raw materials used. Type A gelatine has an IEP value range of pH 7–9.5, while type B gelatine ranges from 4.7–5.4 (De Wael et al., 2010; Lee et al., 2016; Sigma Aldrich, 2020; Baydin et al., 2022). The IEP is significant because at this pH, gel strength, turbidity, syneresis and foaming properties are maximised while viscosity, swelling and gelation properties are minimised. For instance, at the IEP value, the positive and negative charges throughout the molecules are balanced, promoting helix formation. When the pH deviates from the IEP, it inhibits molecular helix formation (Osorio et al., 2007; Goudie et al., 2023).

### **Viscosity of gelatine**

Viscosity is an important characteristic of gelatine, influenced by its bloom strength, concentration and the temperature of the gelatine solution. The viscosity of the gelatine solution increases with 1) higher gelatine concentration and 2) lower temperature. However, viscosity is at a minimum when the gelatine is at its IEP point (Poppe, 1997).

### **Gel strength**

Gel strength is one of the most crucial properties of gelatine, related to its Bloom value. The Bloom value refers to the rigidity of a gelatine gel and is measured under standard conditions. Bloom strength can be measured using a texture analyser, which calculates the amount of force (in grams) needed to create

a 4 mm indentation in a jelly surface using a 12.7 mm diameter plunger at 10°C (Haug and Draget, 2009). The resulting value is referred to as the Bloom value. Higher force indicates a stronger gel. Commercial gelatine typically has a Bloom value ranging from 50 to 300 bloom (Gelita, n.d.; Schrieber and Gareis, 2007b). This can be further classified as low bloom (50–100), medium bloom (100–200), and high bloom (200–300). A higher bloom value indicates stronger gelling power, higher melting temperature, and shorter gelling time in the final product. It also corresponds to a neutral odour and taste, with a lighter colour.

### **Setting and melting temperature**

The temperature at which the sol-gel transition occurs is known as the gelling point, while the temperature at which the gel-sol transition occurs is the setting temperature. These two temperatures can be measured using a temperature sweep. The melting point of a 10% gelatine solution can range from 27–32°C, depending on factors such as the type of pre-treatment of raw materials and the Bloom strength of the gelatine (Poppe, 1997). The setting point of a 10% gelatine gel solution typically varies from 24°C to 29°C. Generally, the temperature difference between the setting and melting temperatures of gelatine with a concentration of 5–25% is around 5°C (Schrieber and Gareis, 2007a).

## **FUNCTIONAL PROPERTIES OF GELATINE**

### **Gel forming**

The ability to form thermoreversible gels in water is one of the most important properties of gelatine. When gelatine is mixed with an aqueous solution, such as water, it forms a colloidal solution rather than a typical one. This behaviour arises because gelatine consists of polymer chains of varying lengths, rather than monodisperse protein molecules (Haug and Draget, 2009). When a gelatine solution is placed in a hot solution, it assumes a random coiled structure. Upon cooling, it partially regains its triple-helical structure, also known as the collagen fold, due to several cross-linkages (Hayashi and Oh, 1983). When the gelatine concentration is sufficiently high, the solution loses fluidity, and a gel forms. The formation of these cross-linkages during gel formation is the slowest part of the process, therefore,



the gel's strength increases over time as more cross-linkages are established (Slade and Levine, 1987).

Gelation of gelatine is an example of physical linking, involving molecular interactions, secondary interactions or a combination of both to create a polymer network. This interaction is sensitive to various factors, including changes in pH and temperature. A notable advantage of physical linkage gelation processes is that they do not require additional chemicals or initiators, as is the case with chemical linking gelation processes. These characteristics make thermoreversible gelatine gels particularly advantageous compared to other gelling agents, such as pectin, which requires both acid and sugar to form an irreversible gel (Schrieber and Gareis, 2007b).

### **Emulsifier and stabiliser**

Stabilisers and emulsifiers are often used together in food production. An emulsifier is a substance added to facilitate the mixing of two substances that would otherwise separate, such as oil and water, by forming an emulsion. A stabiliser, on the other hand, is a substance added to maintain the emulsion's consistency and integrity throughout its shelf life and daily consumption.

Gelatine can function as both an emulsifying agent and a stabilising agent. For example, when oil and water are mixed, the oil breaks into tiny droplets that distribute within the water, forming a temporary mixture. Over time, however, these droplets separate, reforming distinct layers of oil and water. Adding an emulsifying agent like gelatine can help form and stabilise the emulsion, slowing or preventing the separation of the oil-water mixture.

Emulsifiers work by creating a barrier or thin film around droplets, imparting the same electrical charge to prevent the droplets from merging (Vaclavik and Christian, 2014). In this process, a surfactant emulsifier is required. Gelatine is considered a surfactant because its molecules have both hydrophilic (polar) and hydrophobic (non-polar) ends. The hydrophilic part of gelatine is attracted to the polar phase (e.g., water), while the hydrophobic part is attracted to the non-polar phase (e.g., oil). This reduces the interfacial tension between the oil and water phases, stabilising the droplets and preventing coalescence.

The emulsifier capabilities of gelatine are influenced by several factors, including its source, molecular

weight, and extraction method (Zhang et al., 2020). Compared to other emulsifying agents with surface-active properties, such as gum Arabic, gelatine is considered less efficient because it produces large droplets during homogenisation (Karim and Bhat, 2009). However, its performance can be improved through structural modification of gelatine or by combining it with another surfactant (Zhang et al., 2020).

### **Adhesive and cohesive agent**

Gelatine, often referred to as “animal glue”, has been utilised in various fields, including food production and the conservation and restoration of artefacts (Schrieber and Gareis, 2007b; Mosleh et al., 2023). For instance, in cereal bar production, a gelatine solution is applied to completely coat the surface of the materials that need to be bonded, enhancing adhesive forces. As the gelatine cools and forms a gel, cohesive forces increase, effectively binding the two surfaces. Common raw materials used for this purpose include rabbit skin, cattle skin, cattle bones and fish skin gelatine (Mosleh et al., 2023).

### **Thickener**

A thickener or thickening agent is a substance added to increase a mixture's viscosity without significantly altering its other properties. Common examples of thickening agents include gelatine, agar, pectin and gum Arabic. These agents are often used to control the texture of various food products. The rheological properties of thickening agents depend on several factors, such as temperature, the concentration of the active compound, dissolution, degree of dispersion, and more (Himashree et al., 2022). Hydrocolloids, such as gelatine, are particularly popular as thickening agents in food production, primarily due to their water-thickening ability, a characteristic shared by all hydrocolloids.

### **Film former**

Edible films or coatings have been widely used in food production due to their numerous advantages, including improving shelf life, altering molecular interactions between food and its environment, and preventing rapid deterioration under certain conditions (Fakhouri et al., 2014; Lu et al., 2022). These films or coatings can be prepared using various methods, such

as coating, solution casting, and layer-by-layer assembly (Suhag et al., 2020; Lu et al., 2022; Abbadessa et al., 2023).

The coating method is commonly employed, involving the application of an edible coating in liquid form onto the surface of the food through dipping or spraying to extend shelf life. Gelatine is suitable for this purpose due to its strong swelling behaviour in water and excellent gas barrier properties (Lu et al., 2022). However, gelatine has relatively poor mechanical resistance and high permeability to water molecules (Tyuftin et al., 2022), limiting its application in packaging. To overcome these limitations, several strategies have been proposed to enhance its performance. These included combining gelatine with other active ingredients, such as carbohydrates and phenolic compounds (Khan et al., 2020; Zhang et al., 2023).

### **Clarifying agent**

Gelatine is also used as a clarifying agent in food products, such as beverages, to enhance their clarity. For example, in wine production, tannins – compounds responsible for the “dry mouth” sensation, particularly in red wine – play a key role. When exposed to oxygen, tannins undergo oxidation and polymerisation, forming brown, light-diffracting colloids (Jackson, 2014). While this reaction typically occurs slowly, oxidation can alter the hue and diminish the intensity of the wine’s colour over time. The addition of gelatine during wine production addresses this issue. Positively charged gelatine interacts with negatively charged tannins to form large tannin-gelatine precipitates (Jackson, 2014). These precipitates are subsequently removed via filtration or centrifugation, reducing excess tannins. This process results in wine with a smoother mouthfeel, reduced astringency, and a lower tannin oxidation rate. Gelatine with lower bloom strength is commonly used for this purpose due to its superior distribution and ease of homogenisation in cold beverages (Poppe, 1997).

### **Water Binding Capacity (WBC)**

Water binding capacity (WBC) refers to the ability of a substance to retain water after being subjected to external forces (Peters et al., 2017). Gelatine is well-suited for this purpose due to its inherently hydrophilic nature, which arises from a high proportion

of hydrophilic amino acids (those carrying hydroxylic groups), compared to hydrophobic amino acids (Núñez-Flores et al., 2012). These hydrophilic amino acids, with their free hydroxylic chains, form hydrogen bonds with water molecules, effectively “binding” the water as the gel sets at lower temperatures.

### **Protective colloids action**

A colloid is a mixture with particle diameters ranging from nanometres to micrometres, which remain evenly distributed within the solution (Lee, 2019). This phenomenon, known as colloidal dispersion, ensures that the dispersed substance does not settle out of the solution when left undisturbed. The dispersed substance is referred to as the dispersed phase, while the substance that forms the base fluid is referred to as the continuous phase.

Gelatine is classified as a lyophilic colloid, meaning it has particles that strongly interact with the solvent, particularly hydrogen bonds. These interactions form a sheath of solvent molecules around the particles, physically preventing them from aggregating. In contrast, lyophobic colloids have little to no attraction between the dispersed phase and continuous phases. An example of a lyophobic colloid is an oil-water emulsion, where the two phases do not naturally form a stable mixture and cannot be simply prepared through mixing.

When a lyophilic colloid, such as gelatine, is added to a lyophobic colloid, it forms a protective layer that coats the surface of the lyophobic particles. This layer stabilises the mixture by preventing coagulation or separation of the lyophobic compound (Zsigmondy, 1909; Cohen, 1914). For example, an oil-water emulsion is a lyophobic colloid that may coagulate over time. When a lyophilic colloid, such as gelatine, is added, it surrounds the oil droplets, forming a protective sheath that prevents the oil from separating out of the water, thereby effectively stabilising the emulsion.

Gelatine is one of the most efficient colloids available. Its protective action can be quantified using the gold number system, proposed by Zsigmondy (1909). The gold number represents the minimum amount (in milligrams) of lyophilic colloid needed to prevent a colour change in 1 mL of bright red colloidal gold solution to violet when 1 mL of 10% sodium chloride solution is added. The gold number value indicates the

level of protective action: the higher the gold number, the lower the protective effect, as a larger amount of colloid is required to prevent coagulation. Gelatine has a gold number in the range of 0.005 to 0.01, which is relatively low compared to other commonly used colloids, such as gum Arabic (0.15 to 0.25) and egg albumin (approximately 0.1 to 0.25) (Zsigmondy, 1909).

### Formation of foam

Foam is described as gas bubbles dispersed in a liquid continuous phase. It is also known as the colloidal dispersion of gas in a solid or liquid medium, frequently in water (Langevin, 2023). Like emulsion droplets, the gas bubbles in foam need to be protected. However, there are some differences between the two phenomena. For example, the size of the bubble in foams is larger compared to the droplets in emulsions (Bergeron and Walstra, 2005). Additionally, the density difference between the two phases is greater in foams than in emulsions, which increases the tendency for gas bubbles to escape as the continuous phase dries out due to gravity.

Foams are created in the presence of gas, liquid and foaming agents. Forces are required to integrate the gas bubbles into the liquid phase and trap them within it. At this stage, the bubbles formed are often too large and several changes may occur, leading to the rupture of the lamellae between the foam bubbles, resulting in coalescence (Walstra, 1989). A foaming agent, such as gelatine, is then added to the liquid phase, to reduce the surface tension by adsorbing to its surface. Gelatine also coats the surface of the gas bubbles, stabilising the foam. This step is crucial because if the newly formed foam is not stabilised, it will burst out, causing the loss of gas bubbles. Furthermore, gelatine can be whipped to double or triple the volume of the initial sol, enhancing its ability to cover the foam produced (Djagny et al., 2001; Brown, 2011).

## APPLICATION OF GELATINE IN THE FOOD INDUSTRY

Gelatine is widely used in food production due to its multiple functional properties. Two of the most notable properties of gelatine that make it desirable for food production are its ability to form a thermoreversible gel and its “melt in the mouth property”, which

results from its melting point ( $<37^{\circ}\text{C}$ ) being lower than normal human body temperature. Some of the key application areas for gelatine are discussed below.

### Confectionery

Confectionery is one of the common areas where gelatine is utilised. Typically, confectionery is made from sugar, water, corn syrup, and other substances that provide colour, texture, or flavour. Gelatine is often added in confectionery production because it can foam, gel and solidify when used, as seen in products like soft jelly and marshmallows.

In the production of soft jelly, gelatine is added to the sugar and glucose syrup before cooking. The cooking time is typically short enough to prevent gelatine hydrolysis. Type A gelatine is commonly used due to its low viscosity (Poppe, 1997). By adjusting the gelatine concentration or modifying its gelling strength, different firmness and textures of jelly can be achieved.

Marshmallows are a type of aerated confectionery known for their light and soft texture. In marshmallow production, gelatine is added to the sugar mixture and whipped to form foam. Gelatine reduces the surface tension of the foam and stabilises the air-liquid interface by forming a protective sheath around the foam. As the gelatine sets, it helps prevent the phase separation in the marshmallows caused by drainage (Schrieber and Gareis, 2007b).

### Dairy products and desserts

Gelatine is commonly used in dairy products as a texturizing agent, with stabilising, emulsifying, and foaming properties. In yoghurt production, gelatine is frequently added to prevent syneresis. Syneresis is the expulsion of the liquid component of the gel (i.e., whey in the yoghurt) to the surface, which can cause undesired appearance and sensory issues, such as an unpleasant mouthfeel (Lee and Lucey, 2010; El Bouchikhi et al., 2019). Gelatine helps prevent syneresis by inhibiting the whey from being expelled from the casein gel. During gelling, the added gelatine forms a matrix with the casein gel, which is stabilised by hydrogen bonds. This matrix prevents the casein from clustering together and expelling the whey. Interestingly, the addition of 1.5% gelatine can nearly eliminate syneresis formation in yoghurt (Fizman et al., 1999).

Gelatine is also incorporated into ice cream production to improve the texture and melting properties. Ice cream can be described as a foam consisting of air cells, each coated with a layer of fat globules, which are themselves coated with emulsifiers, along with a matrix of ice crystals (Brown, 2011). In ice cream production, gelatine is added to reduce the surface tension of water and is whipped to generate foam. Gelatine binds with water and forms a thin film around the fat globules, helping to stabilise the air bubbles within the ice cream lattice. Additionally, gelatine plays a role in controlling the heat shock phenomena in ice cream, where the ice crystal can grow too large enough as the temperature increases, negatively affecting the mouth-feel (Brown, 2011). When gelatine is used, the ice crystals remain separate within the lattice, preventing them from combining into larger crystals, even with slight temperature changes.

### **Meat products**

Gelatine is used for gelling purposes in canned meat, cured meat, aspic meat, and jellied meat products. Its ability to absorb water and provide form and structure to products that might otherwise fall apart is highly valuable. For example, in the production of canned meat products, gelatine is used to retain the juices that might otherwise be lost during pasteurisation or cooking (Poppe, 1997; Schrieber and Gareis, 2007b). Generally, gelatine with high gel strength is preferred because it can withstand the high-temperature process mentioned earlier.

The water-binding capacity of gelatine is also beneficial for fresh meat. Fresh meat often loses some of its natural juices after slaughter or when separated from the main body. This occurs because actomyosin, the protein responsible for retaining water in the meat, begins to release water (Schrieber and Gareis, 2007b). This loss of moisture can result in less juicy meat when cooked. To enhance water retention, it is advised to add gelatine to meat.

### **Drinks and beverages**

The beverage industry also employs gelatine for the clarification of beverages such as juice, wine, and beer. Gelatine is effective in removing substances that can cause turbidity or bitterness, such as tannins, thereby enhancing the clarity and taste of the beverage

(Jackson, 2014). Gelatine can also be used in conjunction with other clarifying agents, such as bentonite, to further improve its clarifying capabilities (Jafari et al., 2024). Typically, a low-bloom gelatine is preferred for this purpose (Poppe, 1997).

## **CONCLUSION**

Gelatine has become a crucial ingredient in food manufacturing due to its versatile properties. As its use expands across various industries and demand for halal gelatine grows, particularly in Muslim-majority countries, we expect an increase in gelatine production over time. While concerns exist regarding the sources of gelatine, alternatives such as fish gelatine have been proposed for use in food production. For instance, warm fish gelatine has been shown to exhibit properties similar to mammalian gelatine, while cold fish gelatine has suboptimal characteristics in comparison. Chicken gelatine is also being considered as an alternative, as it possesses superior properties compared to bovine gelatine. Future research could explore other potential sources of gelatine, such as poultry products and insects, as viable substitutes for mammalian gelatine. Additionally, modifying gelatine properties through the incorporation of substances like gellan gum, pectin, and salts may also offer new opportunities to enhance its functionality, especially in food production.

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## **DECLARATIONS**

### **Data statement**

All data supporting this study has been included in this manuscript.

### **Ethical Approval**

Not applicable.

### **Competing Interests**

The authors declare that they have no conflicts of interest.

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