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## **Review Article**

# Sesame seed protein isolate as a value-added by-product hydrocolloid: A review on functional properties and food applications

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## **Abstract**

Sesame (Sesamum indicum L.) is one of the most important oilseed crops cultivated worldwide. Sesame cake, as a by-product of sesame oil extraction contains approximately 50% protein. So, it has high potential for use as a protein source for a wide range of applications in the food industry. The sesame protein isolate (SPI), as a promising plant-based protein, can be used as a value-added hydrocolloid. Introducing the capabilities of the SPI can help expand its application in the food sector. This paper investigates those functional characteristics of the SPI that are important for use in food. The extraction, purification, and chemistry of the SPI are investigated. The solubility, water/oil binding capacity, rheological properties, and the ability to interact with polysaccharides are the most important characteristics of the SPI related to food application that are discussed in this paper. Moreover, all the potential applications of the SPI in the food industry are reviewed. This valuable plant protein has been used as an emulsifying/foaming agent, gelling hydrocolloid, film-forming/edible coating material, and also as a wall material for encapsulation purposes. The benefits and shortcomings of the SPI for all applications are discussed. The fields of use of the SPI in the food industry are much wider. This review paper is expected to open a new horizon in the use of this plant protein in the food industry by introducing the importance of SPI as a value-added by-product hydrocolloid.

**Keywords:** Emulsifying activity, Extraction, Protein isolate, Sesame cake, Solubility.

## Introduction

Hydrocolloids are natural macromolecular substances extracted from plant sources, animals, seaweed, and microorganisms. Hydrocolloids have a wide range of functions due to their high molecular weight and having many hydroxyl groups in their structure, which give them hydrophilic material with good surface-active properties (Yemenicioğlu et al., 2020; Liu et al., 2020). Hydrocolloids are used as food additives due to their useful functional properties. Some hydrocolloids can be used in the

food industry as thickening, gelling, foaming, emulsifying, water-binding, oil- and flavor-binding, texturizing, or stabilizing agents (Khalil et al., 2018). Recently, hydrocolloids have been used to form films and edible coatings for packaging purposes and to improve the safety and quality of food (Sason & Nussinovitch, 2021). Hydrocolloids have also been used for the encapsulation of food additives, bioactive compounds, and probiotics. The use of hydrocolloids as functional ingredients to modify the gastrointestinal fate of food has also been approved.

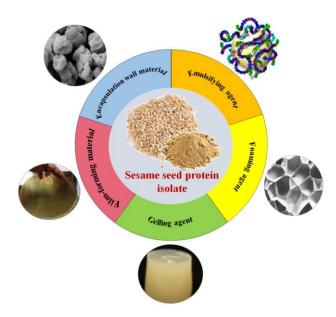
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They are used to control the hydrolysis of macronutrients (such as fats, proteins, and starches) in the gastrointestinal tract (McClements, 2020) or to protect probiotics during storage and passing through gastrointestinal conditions (Azizi et al., 2021). Based on their chemical structure, hydrocolloids are divided into two main groups of polysaccharides and proteins. Proteins have amphiphilic properties due to the presence of hydrophilic and hydrophobic groups in their amino acids. Due to their amphiphilic nature, proteins are suitable for adsorption at the oil-water interface, creating a stable viscoelastic film that makes them a useful hydrocolloid for emulsion and foam stability al., 2020). Protein-based (Yemenicioğlu et hydrocolloids have several advantages polysaccharide-based hydrocolloids, including their amphiphilic nature, strong surface-active properties, higher flexibility, linear chain structure, higher reactivity, and high nutritional value.

Various reports and forecasts indicate that the value of hydrocolloids in the global food market is expected to increase by 50% in the next decade. Therefore, extensive studies are needed to improve the efficiency of hydrocolloid sources and introduce various agro-industrial wastes as new sources of hydrocolloids (Nishinari et al., 2018; Yousefi & Jafari, 2019). Sesame (Sesamum indicum L.), belonging to the order Tubiflorae and family Pedaliaceae, is one of the most important oilseed crops (approximately 50% oil) in the world because of its high content of unsaturated fatty acids. The world's major sesame seed producers are China, Burma, India, Ethiopia, Nigeria and Sudan. Sesame cake, a by-product of sesame oil extraction, is typically used as animal feed. However, it is a good source of protein, and depending on the variety and extraction method, it contains about 50% protein (Laohakunjit et al., 2017). Sesame protein has high nutritional value due to having considerable amounts of essential amino acids such as methionine, cysteine, and tryptophan, which are limiting amino acids in most vegetable/seed proteins (Idowu et al., 2021; Mailer, 2016). Besides its nutritional importance, the sesame cake has a high potential for use as an inexpensive protein source in the food industry. Sesame protein isolate (SPI) is a suitable option for a wide range of industrial applications because of its facile extraction process, high molecular weight,

good thermal stability, and low water solubility. Figure 1 shows all the potential applications of SPI in the food industry reported in the literature.

The interesting functional characteristics of SPI, such as foaming capacity, whippability, emulsifying activity, film-forming potential, and fat absorption capacity, open a wide horizon for the application of this waste-originated hydrocolloid in the food industry. The main objective of this review is to provide comprehensive insights into the extraction methods and physicochemical and functional properties of SPI. In addition, the emulsifying, foaming, and gelling capacities, film-forming potential, and encapsulation activity of SPI are presented to explore the potential applications of SPI as a new plant-based hydrocolloid in the food industry. The current usage, which researchers have reported so far, and possible future trends of this hydrocolloid in food-related studies are also discussed in this review.



**Figure 1.** The potential applications of the sesame protein isolate in the food industry which are reported in the literature.

## SPI extraction and purification

The SPI can be obtained by alkali or salt treatment of sesame cake. Figure 2 shows a diagram of the two methods. Moreover, SPI fractions can be extracted separately based on sesame protein solubility in different media. Alkali SPI extraction from defatted sesame meal is based on the precipitation at the isoelectric pH of protein. In this method, sesame protein is extracted by dispersing cold-pressed oil extraction residue in water at pH 10, followed by precipitation by lowering the pH to 4.5 (Gómez-Arellano et al., 2017). SPI can also be extracted using salt in ammonium sulfate or NaCl 1 M at pH 7, followed by precipitation at 10% HCl or pH 4.5 (Brewer et al., 2016; Onsaard et al., 2010). The final step in separating SPI from whey involved

neutralization to pH 7.0 and washing the protein by dispersing it in distilled water. The SPI obtained from both methods contains nearly 95% protein (Fasuan et al., 2018). Achouri et al. (2012) extracted the SPI by the salt method and studied the effect of NaCl concentration on the yield of SPI. They reported an increase in the yield of SPI from 12.5% at 0 M NaCl concentration to 54.6% at 1 M NaCl concentration. However, this increase did not affect the main components of the SPI profile.

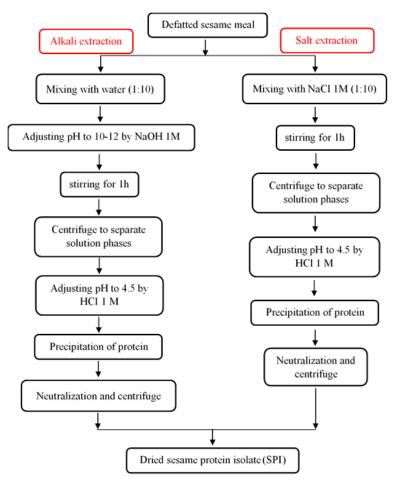


Figure 2. Flowchart for alkali or salt extraction methods of protein isolate from sesame seeds.

The optimum pH range for maximum extractability is reported to be around 7–10 and in the presence of 0.6-1 M NaCl (Achouri et al., 2012). The protein content of the SPI extracted by the alkali method ( $\sim$ 83%) was higher than that of the salt method extracted SPI (75.5%). This result indicates that the extraction process affects the protein content of SPI (Onsaard et al., 2010). The oil extraction method and

other pretreatments can also affect the yield and characteristics of the extracted SPI. For example, Abirached et al. (2020) reported that after supercritical fluid oil extraction, the yield of SPI isolated from sesame cake was enhanced using the alkali method. Moreover, roasting or boiling sesame seeds before oil extraction improves SPI production from sesame cake. Soaking the seeds in agitated

water helps extract the protein. This process prevents protein damage and preserves their functionality during oil extraction before SPI isolation. In addition, enzymatic pretreatments can break polysaccharide-protein complexes, improving the protein extraction yield (Zakidou & Paraskevopoulou, 2021; Latif & Anwar, 2011).

#### Chemical structure of SPI

The chemical structure of a hydrocolloid directly affects its functional properties. Therefore, it is necessary to know the chemical structure of SPI before investigating its functional properties. As reported by Sharma et al. (2016b), the SPI protein content is approximately 90.50% (Fasuan et al., 2018). Other researchers have reported the same amount of protein in the SPI. On average, the SPI contains about 86% protein, 6.9% moisture, 3.3% ash, 1.8% fat, and 1.6% crude fiber. In general, the protein content of isolated sesame protein is more than 80%; therefore, it can be called an isolate and not a concentrate (Fathi et al., 2018).

In terms of building arrangement, SPI is a globular protein. The secondary structures of globular proteins are often β-sheets, while located along with the oil-water interface, shift to the form of  $\alpha$ -helix, and form a viscoelastic membrane (Achouri et al., 2012). The main components of sesame seed protein are salt-soluble globulins (67.3%), water-soluble albumins (8.6%), dilute acid or alkali-soluble glutenins (6.9%), and alcohol/water mixture-soluble prolamins (1.3%) (Pal et al., 2020). Sesame protein with a high globulin content has been recognized to have strong functional properties for use as a hydrocolloid. Accordingly, the SPI isolated by the alkaline method is considered optimal, as it contains 41.3% albumin, 0.41% glutenin, 8.14% globulin, and 0.8% prolamin (Gómez-Arellano et al., 2017). Poveda et al. (2016) reported that the use of pure water or salt solution as a solvent had no significant effect on the performance of SPI, but Achouri et al. (2012) showed that increasing the concentration of NaCl improves the extraction yield and performance of the SPI due to an increase in the solubility of globulins. Sesame globulins are composed of two parts:  $\alpha$ -globulin (11S globulin) ( $\sim$  80%) and  $\beta$ globulin (2S albumin) (~ 20%) (Achouri et al., 2012;

Saatchi et al., 2019).  $\alpha$ -Globulin manifests as tetragonal pyramid crystals, whereas β-globin is obtained as a white, amorphous powder (Onsaard et al., 2010). SDS-PAGE (Sodium Dodecvl Sulfate-Polyacrylamide Gel Electrophoresis) is a laboratory technique that separates proteins based on their molecular weight by using an electric current and a polyacrylamide gel matrix. SDS-PAGE was used to measure the molecular weights of sesame globulin fractions. The 13 and 20-35 kDa polypeptide bands are related to sesame 2S albumin and 11S sesame protein globulins, respectively. In some cases, 2S albumin has been reported to be allergenic (Saatchi et al., 2019).

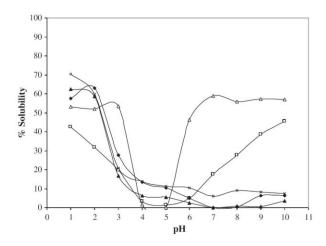
Fasuan et al. (2018) compared the amounts of essential amino acids in SPI with those in reference proteins (hen's egg), pea, and soy protein. The results showed that glutamic acid had the highest percentage of amino acids, and leucine had the highest percentage of essential amino acids in SPI. Sulfur-containing amino acids (methionine and cysteine) are limiting amino acids in the SPI. However, their content is higher than that of other plant proteins. The SPI protein also has higher levels of arginine than the reference proteins. The number of amino acids in sesame protein is very similar to those of reference proteins, soy, and peas. The ratio of essential amino acids to total amino acids in SPI is approximately 39%, which is higher than the minimum adequacy reported by the FAO for infants (26%) and adults (11%) (Fasuan et al., 2018). Di et al. (2022) showed that germination caused adjustments in the protein profile of sesame protein, and the excessive molecular weight proteins were decomposed into low molecular weight proteins, thereby causing changes in the shape and functional properties of sesame protein.

## **Functional characteristics of SPI**

Functional properties are defined as "physical, or chemical properties of proteins that influence the behavior of proteins in food during processing, storage, and consumption" (Owens, 2010). These properties depend on variables such as pH and salt content of protein dispersion, whipping, seed germination, and protein conformation, which may affect the functions of protein as a hydrocolloid in food products. The importance of these properties depends on the type of food product in which the protein is to be used. For example, meat products require protein isolates with high oil and water holding capacities, whereas proteins with high emulsion and foaming properties are suitable for salad dressings and soups (Sharma et al., 2021). Accordingly, some important functional properties of the SPI, such as protein solubility, rheological properties, oil/water-binding capacity, and interaction of the SPI with carbohydrates have been investigated in this section.

### **Solubility**

Protein solubility is an important functional property. Many protein-based formulations, such as foams, emulsions, and gels, usually require good protein solubility. This directly affects the texture, color, and organoleptic properties of food products. The good solubility of SPI is one of its strengths compared to other common protein isolates, such as flaxseed and soybean proteins. Its solubility has also been studied under different conditions, such as irradiation and NaCl presence (Elsorady, 2020; Achouri et al., 2012; Hassan et al., 2018). Ordinarily, SPI is insoluble in water at neutral pH (Azizi et al., 2021). A U-shaped curve has been reported for the solubility of SPI at different pH values. Figure 3 shows the pH-solubility profile of SPI in different solutions and its comparison with soy protein isolate (Achouri et al., 2012). The lowest solubility is related to the isoelectric pH (reported to be about 8%) (Fasuan et al., 2018), which is related to the lowest electrostatic repulsion and ionization of molecules. Above and below this pH, the increased net charge of the protein increases the electrostatic repulsion, and thus the ionic hydration improves its solubility. The solubility of about 94% at pH 12 has been reported by Sharma et al. (2016b). In the presence of NaCl, an increase in SPI solubility is reported due to the rise in the ionic strength of the dispersing medium and salting-in effect (Fasuan et al., 2018). Increasing the NaCl concentration to 1 M due to the salting-out effect of NaCl and high adsorption of chloride ions by the proteins decreases the repulsive interactions between protein molecules; therefore, the solubility shows a drastic decrease (Fasuan et al., 2018; Achouri et al., 2012; Khalid et al., 2003).



**Figure 3.** pH-solubility profiles of sesame protein isolate (SPI) in water ( $\square$ ), 0.2 M NaCl ( $\spadesuit$ ), 0.6 M NaCl ( $\blacktriangle$ ), and 1 M NaCl (x) compared to soy protein isolate in water ( $\Delta$ ) (Achouri et al., 2012).

Moreover, the same results have been reported by Khalid et al. (2003) about the solubility of the SPI, which is influenced by pH and salt concentration. The solubility of SPI fractions has also been studied separately. Albumin fraction showed the highest solubility in the pH range, while globulin had better solubility at alkaline pH, which may be due to the increase in the net charge of protein as the pH increases. Glutelin has the lowest solubility due to the lowest electrostatic repulsion, which facilitates protein aggregation. The prolamin is insoluble in the aqueous solution (Idowu et al., 2021). Hassan et al. (2018) suggested that gamma irradiation up to 1.0 kGy, unfolds the protein molecules, which causes an increase in protein solubility, emulsifying, and foaming properties by increasing the hydrophobicity and molecular flexibility. Accordingly, higher dose levels decreased protein solubility progressive denaturation of the SPI (Hassan et al., 2018). Ultrasonic pre-treatment of the SPI by Yang et al. (2021) caused the transformation of the SPI from soluble to insoluble aggregates due to the unfolding of hydrophobic groups from the inner part of proteins, which could decrease the amount of highly charged proteins (Yang et al., 2021). Di et al. (2022) studied the effect of germination of the solubility of sesame protein and reported that solubility increased after germination. Also, with the extension of germination time, the solubility of sesame protein continued to increase (up to 3.58 times on the 4th day). This may be because the hydrolysis activity of proteases was enhanced after germination, which hydrolyzed the proteins into shorter peptides and free amino acids, resulting in an increase in protein solubility. Sibt-e-Abbas et al. (2020) indicated that the solubility of the SPI is higher than those of flaxseed and canola protein isolates but lower solubility of the SPI was reported by Achouri et al. (2012) and Elsorady (2020) compared to soy and flaxseed protein isolates.

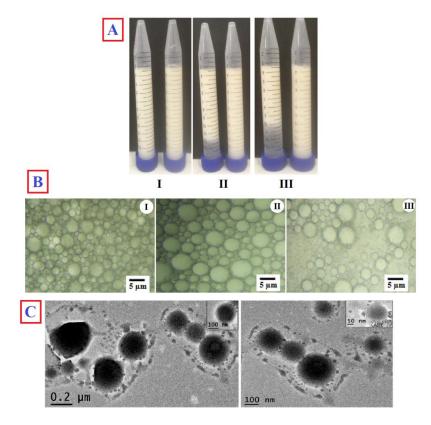


Figure 4. A) Creaming behavior of emulsions prepared with sesame protein concentrate (SPC) (left) and SPC-malodextrin conjugates (right) after storage for 0 day (I), 1 day (II) and 30 days (III) at pH 7 (Saatchi et al., 2019); B) Micrograph of homogenized sesame oil emulsions stabilized by the SPI at 8000 rpm (I, III) and 11400 rpm (II) at different concentration of sesame protein isolate (SPI) 1.5% wt (I) and 2.5 wt % (II, III) (Brewer et al., 2016); C) TEM images of the nanoemulsion formulations stabilized by the SPI (Dey et al., 2018).

#### Water binding capacity and oil binding capacity

Water binding capacity (WBC) refers to the ability of protein particles in food to absorb and retain water against gravity. The WBC can reflect the texture and viscosity of the matrix. The amount of water associated with proteins is closely related to their amino acid profile and increases with the number of charged residues, conformation, hydrophobicity, pH, temperature. ionic strength and protein concentration (Idowu et al., 2021, Lawal et al., 2021). The high WBC of the SPI can be attributed to the presence of hydrophilic (polar) amino acids at the protein-water interface (Sibt-el-Abbas et al., 2020). WBC of the SPI fractions has also been studied.

Ordinarily, the proteins with high solubility (albumin) have low WBC. Prolamin as the least soluble protein, has the highest WBC, followed by glutelin and then globulin (Idowu et al., 2021).

Conversely, oil binding is mainly attributed to the physical entrapment of oil and the number of hydrophobic (nonpolar) side chains on the protein's amino acids that bind hydrocarbon chains on the fatty acids. It is a reflection of the thickening and emulsifying properties of proteins. Oil binding capacity (OBC) is an important property in food formulation because fats improve the flavor and mouthfeel as well as emulsion characteristics of the food commodities (Lawal et al., 2021; Sibt-E-Abbas

et al., 2020). Several non-polar amino acids are present in the SPI, resulting in higher absorption of oil. Accordingly, the OBC of the SPI is observed to be more than the WBC. This is due to the fact that the SPI is not soluble in water at neutral pH (Sharma et al., 2016b). Tirgarian et al. (2019) have reported that the products with higher WHC show lower OBC.

Toasting (Lawal et al., 2021) and ultrasonic pretreatment (Yang et al., 2021) and treatment of the SPI with electromagnetic fields with subsequent thermos-denaturation (Bugayets et al., 2020) led to better WBC and OBC values in comparison to the untreated SPI, which was because of the partial unfolding and denaturation of protein structure and unmasking of the non-polar residues from the interior of the protein molecule. The WBC and OBC of the SPI have been reported to be lower than those of soy protein isolate (Achouri et al., 2012), while they were higher than those of canola protein isolate and flaxseed protein isolate due to the quantity and conformation of polar side chains (Sibt-E-Abbas et al., 2020). Hafez (2018) reported that by considering the greater WBC and OBC of SPI, it can be suggested that the suitability of sesame processing wastes for the preparation of bakery products like free gluten crackers for celiac diseases.

#### Rheological properties

The viscosity and consistency of proteins' dispersions are critical in fluid and gel-like foods. The viscosity depends on the protein-solvent and the protein-protein interaction. The rheology of the continuous phase directly affects the stability of a food emulsion (Gómez-Arellano et al., 2017, Brewer et al., 2016). Abel Gómez-Arellano et al. (2017) studied the effect of changes in protein concentrations, temperature, ionic strength, and pH on the viscosity of the SPI dispersions. In low concentrations of the SPI, the shear viscosity exhibited Newtonian behavior. Accordingly, the low viscosity of dispersion can be useful in the production of highly protein-enriched beverages without causing viscosity problems. Moreover, an increase in the ionic strength and deviation of pH from the isoelectric point and a decrease in temperature induced an increase in the protein dispersion viscosity, and their behavior became shear-thinning (Gómez-Arellano et al., 2017). The

SPI also exhibits a shear-thinning behavior (pseudoplastic) as shear rate increases, because the hydrogen bonds and other weak bonds break and lead to the separation of aggregates of protein networks.

Shear-thinning behavior of proteins in dispersions is due to the continuous orientation of the molecules in the direction of flow to reduce the deformation of proteins and frictional resistance by hydration in the direction of flow (Saini et al., 2018; Moreno-Santander et al., 2020; Brewer et al., 2016). Di et al. (2022) confirmed that with the extension of germination time, the apparent viscosity of the protein dispersions reduced first after which it increased, which can be applicable to the adjustments within the protein shape after germination. The decrease in the viscosity is due to an increase in the mobility of surface-active groups to adsorb at the interface. Conversely, the discount in surface hydrophobicity may also cause a growth in viscosity. The use of sesame milk as an emulsifier in the food industry has been suggested by Karshenas et al. (2018). The relation between the shear stress and shear rate for mayonnaise was nonlinear with non-Newtonian flow behavior and a higher elastic modulus was reported at the low angular frequencies than the viscous modulus. Shear-thinning flow behavior helps the souse to pump and flow more easily (Karshenas et al., 2018; Karshenas et al., 2019).

#### **Interaction with polysaccharides**

The SPI has poor functional properties at slightly acidic and neutral pH levels. In order to overcome this limitation, proteins can be conjugated with polysaccharides. Conjugation through the Maillard reaction under mild and secure conditions would be valuable for the food industry because no external chemicals have been employed to obtain new ingredients with improved functional properties. Saatchi et al. (2021) conjugated the SPI and maltodextrin and reported that the conjugation enhanced the solubility of the SPI and provided a soluble substance. The tendency of the protein to form aggregates decreased in the continuous phase, and its solubility did not show a sharp decrease at the isoelectric point. It was also found that conjugation of maltodextrin to the SPI increases the viscosity and adhesion of the emulsion.

The surface of the polysaccharide-modified protein can increase the interaction between the protein and water at all pH values compared to that of nonconjugated SPI. The emulsifying activity index (EAI) and emulsifying stability index (ESI) of conjugated proteins increase because of the extended secondary structure of the molecules and quicker adsorption of the molecules to the oil-water interface, and hence the conjugated compound formed smaller droplets in O/W emulsion. This improvement is due to the amphiphilic structure of the conjugated polymers, and it is also affected by enhanced steric repulsion, which causes a decrease in the aggregation and coalescence of the droplets. It should be noted that thermo-denaturation during the conjugation process causes partial unfolding and unmasking of the nonpolar residues from the interior of the protein molecule, which enhances the emulsifying properties of the SPI (Saatchi et al., 2021). Saatchi et al. (2019) prepared sesame protein concentrate (SPC)maltodextrin conjugates and evaluated their emulsifying potential.

The emulsifying capability of SPC-maltodextrin conjugates was higher than that of SPC, and emulsions with smaller droplet sizes were obtained. Figure 4A shows the stability of the emulsion at pH 7 during an extended storage period (30 days). No visible flocculation and creaming were observed for SPC-maltodextrin conjugate stabilized emulsions, which indicated that this conjugate could produce efficiently stable emulsions. Moreno-Santander et al. (2020) reported that the gum-gel based on the SPI presents an elastic modulus higher than its viscous modulus. Mechanical properties of gels were improved with the increase of xanthan gum and carrageenan in formulations with low protein content. So, conjugated SPI can be introduced as a promising source of hydrocolloid to develop different food emulsions.

## Food applications of the SPI as a hydrocolloid

## **Emulsifying agent**

Proteins are surface-active molecules due to their amphiphilic structure. They are adsorbed to the surface of oil droplets in emulsions and create electrostatic repulsion, and eventually stabilize the emulsion. In fact, proteins can enhance the thermodynamic stability of oil-in-water emulsions by orienting lipophilic residues to the oil phase and hydrophilic residues to the aqueous phase and thus simultaneously happens the reduction of the surface tension and increasing of steric stabilization. Brewer et al. (2016) showed that the SPI-stabilized sesame oil emulsions have non-Newtonian liquid behavior. According to their results, the droplet size increased by increasing protein concentration but the polydispersibility index (PDI) decreased and better size distribution was achieved at higher SPI concentration and higher shear rate (Fig. 4B). The ability of a protein to create an emulsion is defined as EAI, which determines the number of interfacial areas that might be stabilized per unit amount of protein. Also, the stability of the identical emulsion over a particular time is remarked as ESI. The emulsion properties of proteins may be influenced by molar mass, hydrophobicity, conformation, pH, temperature, and ionic strength of the molecule (Sibt-e-Abbas et al., 2020).

Similar to the protein solubility, the emulsifying property of the SPI is pH-sensitive, which can ensue the explanation that emulsion activity depends upon the hydrophilic-lipophilic balance of the SPI which is affected by pH. The lower solubility of sesame protein hydrolysates at pH 4.0 is accountable for the lower EAI and ESI at that pH (Tirgarian et al., 2019). The SPI shows an increase in emulsion stability (ES) with the increase in pH. This might be because an increase in the pH ends up in increased coulombic repulsions between neighboring droplets. These factors result in a reduction in interface tension, which may cause higher ES (Sharma et al., 2016b; Cano-Medina et al., 2011; Fasuan et al., 2018). The results of Lawal et al. (2021) suggested that while the sesame flour may not form an emulsion readily, the stability of the emulsion after its formation is comparatively high. The stability of the protein film formed at the interface of the emulsion relies on the interactions of the proteins in oil and aqueous phases (Lawal et al., 2021). Sibt-e-Abbas et al. (2020) have compared hydrocolloid properties of the SPI to flaxseed and Canola protein isolates and reported that the most emulsifying capacity (EC) and ES belong to the SPI. He et al. (2020) studied on emulsifying properties of the SPI and reported that its EAI is better than that of the soybean protein isolate (He et al., 2020). It is reported that protein denaturation, toasting, and gamma irradiation up to 1.0 kGy enhance the emulsifying properties of the SPI attributable to the increased elasticity and hydrophobic surface because of unfolding in protein molecules (Sibt-e-Abbas et al., 2020; Lawal et al., 2021; Hassan et al., 2018). Moreover, the addition of NaCl up to 1M increases the EAI and ESI of the SPI. This increment may be related to the increase in the solubilization of the protein as the ionic power is increased and, accordingly, the emulsion capacity will be improved (Fasuan et al., 2018; Elsorady, 2020).

The SPI has also been used for the preparation of nanoemulsions. Dev et al. (2018) studied the performance of the SPI as a surfactant for the preparation of  $\omega$ -3 PUFA-enriched nanoemulsions. The presence of SPI in the nanoemulsion formulation increases its stability in the gastrointestinal tract and the durability of PUFA. Also, the emulsion system was stable against the accumulation of particles due to increases in the thickness of the interfacial capping along with the simultaneous ascend of the steric repulsion. Figure 4C shows the TEM images of the nanoemulsion droplets coated with the SPI layer. The covering of a thick layer on the surface of droplets is detectable. Chatterjee et al. (2015) have reported that the proteolytic activity of pepsin produces several small surface-active peptides which expand the EAI and ESI of the SPI and cause an increase in surface stabilizing activity of SPI.

To evaluate the emulsifying properties of the SPI fractions, Idowu et al. (2021) showed that at most of the pH values, lower oil droplet sizes (higher emulsion quality) were obtained for prolamin and albumin fractions than for glutelin and globulin fractions of the SPI because of hydrophobic characteristic and presence of less folded and open structure of prolamin and albumin. Over a wide range of pH, the albumin had similar solubility. This suggests that albumin is a better emulsifier than globulin or other fractions of the SPI. It should be noted that a change in pH affected the emulsionforming ability of the SPI fractions (Idowu et al., 2021).

### Foaming agent

Foams are hydrocolloid systems with two phases: the continuous phase of the liquid and the dispersed phase of gas or air (Tirgarian et al., 2019). A prerequisite for protein foaming is its solubility. Foaming capacity represents the ability of a protein to form a thick, flexible, and viscoelastic film in the interfacial layer, which reduces the surface tension and increases the volume of the protein solution in combination with air. To stabilize the foam in food products such as beer and bakery products, the proteins must interact with each other around air bubbles inside the matrix. An ideal foaming agent can hold air and maintains its volume and shape over a period against gravitational forces and ultimately produces stable foams (Tirgarian et al., 2019). The SPI may find application in those foods that require good foaming properties, such as salad dressing (Fasuan et al., 2018). The SPI has higher foaming properties compared to sov protein isolate (Achouri et al., 2012) and flaxseed and canola protein isolates (Sibt-e-Abbas et al., 2020; Elsorady, 2020). The foaming properties of proteins are influenced by gamma irradiation up to 1.0 kGy and ultrasonic pre-treatment, which caused an increase in the foaming capacity (FC) and foaming stability (FS) of the SPI and promoted interfacial membrane formation, which is due to the enhanced protein unfolding and structural flexibility (Yang et al., 2021; Hassan et al., 2018). On the other hand, the toasting decreased the foaming capacity and stability of the SPI due to protein denaturation (Lawal et al., 2021). By moving from neutral to alkaline pH, and adding NaCl at a concentration up to 1.0 M and a rise in ionic strength of the dispersing medium, improvement at the FC and FS of the SPI is reported (Elsorady, 2020; Sharma et al., 2016b; Fasuan et al., 2018) which is interrelated with the increased solubility due to an increase in net charge on the protein and speeding up the protein spreading to the air-water interface. The lowest FC is at the protein's isoelectric point. A study on the foaming properties of the SPI fractions shows that glutelin and prolamin are more stable than albumin and globulin at pH 3 and 9, due to the weak surface membrane produced by albumin and globulin, which cannot prevent the air bubbles from coalescence (Idowu et al., 2021). Germination remedy has stepped forward the foaming potential of sesame protein with growing germination time

(Di et al., 2022). After four days, the foaming potential of sesame protein reached about 2.7 times that before germination. The hydrophobicity and solubility of sesame protein had been extended after germination, at the same time as the molecular weight of the protein turned into decreased, which can also additionally enhance the formation of foam. These small peptides are believed to be unsuitable for forming stable foam due to the decreased interactions among proteins.

These outcomes indicated that germination improves the foaming potential of sesame protein. Zakidou and Paraskevopoulou (2021) in an interesting study, compared the foaming behavior of sesame protein extract with two commercial samples, i.e., cow milk and a soy beverage, suitable for cappuccino coffee drinks' preparation. The SPI exhibited satisfactory foam characteristics comparable to those of commercial soy beverages but inferior to or similar to those of cow's milk. The SPI incorporation on cassava starch foams used for packaging applications improved the foaming ability of starch batter, which can result in cell walls being more resistant to collapse during water evaporation during the time of thermal expansion process (Machado et al., 2017). Figure 5 shows the cross-section of starch-SPI foams. This research group proposed that, since low density is desirable, the SPI incorporated starch foams are promising for further practical use of foam-based packaging materials.

## **Gelling agent**

Gelation is an important property that affects the texture of foods. The ability of a protein to form a gel is measured by the least gelling concentration (LGC) and indicates the minimum amount of protein required to form a gel at which the gel remains in the inverted tube. The lower the LGC, the higher the ability of the protein to form a gel. Gel formation usually occurs at higher temperatures than protein denaturation. The structure of the gel improves the binding of water, flavor compounds, sugar, and other substances to the food texture. The SPI can be a choice in the use of the formation of gel or as an additive to other gel-forming materials in food products (Elsorady, 2020; Olasunkanmi et al., 2017). The protein, lipid, and carbohydrate contents of protein isolates cause different LGC of them. Sibt-e-Abbas et al. (2020) has studied the gelling properties of oilseed protein isolates and reported that the LGC for SPI is higher than flaxseed and canola protein isolates. Also, defatted sesame flour, especially roasted flour, shows higher gelation properties (lower LGC) than those of defatted soy flour (He et al., 2020). The gelling ability of the SPI is pHdependent, as the minimum LGC is observed at pH 12 and the maximum at pH 9, indicating that the gelling ability of the SPI increases with alkaline pH (Sharma et al., 2016b). Lawal et al. (2021) also reported that processed sesame flour is a good gelling agent and can be useful in food products such as puddings, sauces, and soups. In a study on the gelling formation capacity of the SPI fractions, globulin and albumin have been reported to be better than prolamin and glutelin, due to the smaller number of hydrophobic clusters (Idowu et al., 2021). In one case, factors affecting disulfide bonding contributing to the gelation of sesame globulins have been investigated. It was observed that hydrophobic interaction provides more stability to the SPI gels (Bröckel et al., 2013).

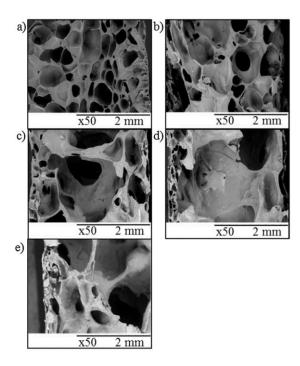


Figure 5. Scanning electron micrographs of cross-sections of the cassava starch foam trays produced with different SPI added: a) 0%; b) 10%; c) 20%; d) 30% and e) 40% (Machado et al., 2017).

Rheologically, the gel is a viscoelastic substance for which, as a function of frequency, storage modulus (G') is larger than loss modulus (G"). The gels obtained from the SPI were opaque, characteristic of aggregate or particulate. The clots had a macroscopic size and scattered light. The resulting gel offered a higher G' than the G". Moreno-Santander et al. (2020) showed that the concentration of protein is directly related to the firmness of the gels. Saini et al. (2018) reported the increase of both G' and G" with the temperature above 60 °C, suggesting the developed viscoelastic properties due to the better solubility of the SPI at higher temperatures, as gelation depends on the solubility and hydration of proteins.

#### Film-forming and coating material

Today, environmental concerns have increased interest in producing packaging materials based on natural polymers. Proteins, polysaccharides, or a combination thereof are used to produce biodegradable and edible packaging materials in the form of films or coatings. If a thin layer forms directly on the surface of the food, it is called a coating, and if a separate layer is formed and then the food surface is covered with it, it is called a film. Edible films and coatings protect food products from physical damage, mechanical impact, pressure, moisture, microbial contamination, etc. Due to the SPI's high molecular weight, good heat stability, and low water solubility, it can be considered as a good coating and film-forming material. The film-forming potential of the SPI and morphological structure of the produced films strongly depend on the purity of protein and also the film-forming conditions (Fathi et al., 2018). The physical characteristics, like thermal, mechanical, and water barrier properties of the SPI films, are better in comparison to other protein-based edible films such as peanut proteins, soy protein, lentil protein, and faba bean protein, and can be used for packaging or coating applications of fruits and vegetables. Moreover, the alkaline pH range is chosen for the films because the SPI is insoluble at acidic pH, so it is not possible to prepare films at acidic pH values. With increasing pH and temperature, proteins are denatured and wider structures of proteins are formed, which increases the chain-to-chain interactions in polymers and creates stronger films with less permeability to gases

and water (Sharma & Singh, 2016a). The effectiveness of this film depends on the water vapor pressure gradient because it contains hydrophilic parts of the protein. On the other hand, the water vapor permeability for food packaging films should be as low as possible. In order to improve these properties of the film, some treatments have been suggested. The treatment with organic acids (Sharma et al., 2018b), TiO2 nanoparticles (Fathi et al., 2019), and UV radiation (Fathi et al., 2018) creates crosslinks between protein molecules and reduces the solubility and WVP of films, and increases hydrophobicity, water, and oxygen barrier properties, and ultimately causes to improve the structure, morphology and mechanical properties of films.

Most of the plant protein-based films are susceptible to photo-cross-linking. Increasing the crystallinity, tensile strength, and water barrier properties of most protein-based edible films after exposure to UV radiation has been reported (Díaz et al., 2016; Schmid et al., 2015). Fathi et al. (2018) investigated the photo-cross-linking effect of UV radiation on the SPI-based edible films. The effects of different UV light types (UV-A, UV-B, and UV-C) were compared on the film-forming solution and pre-formed films. A more compact film with high crystallinity was achieved after UV radiation. The solubility and WVP decreased, and the hydrophobicity of the films increased after UV exposure. Mechanical properties were also improved, and the UV-C irradiated filmforming solution exhibited the highest tensile strength and Young's modulus. This research group attributed the improving effect of UV radiation on the physical properties of the SPI films to disulfide bonding and also other types of bonds, such as covalent bonds between aromatic amino acids present in the SPI. UV radiation is absorbed by double bonds and the aromatic rings of some amino acids, such as tyrosine, phenylalanine, and tryptophan. Producing free radicals in those amino acids leads to the formation of intermolecular covalent bonds between the radical aromatic residues. Having a higher amount of these amino acids makes the SPI a suitable protein for crosslinking induced by UV radiation. In another study, this research group incorporated TiO2 nanoparticles into the SPI films to produce nanocomposite films (Fathi et al., 2019). The mechanical and thermal

properties of the films improved after TiO2 incorporation, and the SPI-TiO<sub>2</sub> nanocomposites exhibited good photocatalytic activity. However, the TiO<sub>2</sub> addition decreased the transparency of the SPI

films, and more opaque films were achieved by increasing TiO<sub>2</sub> concentration. Figure 6 shows the appearance of the SPI-TiO<sub>2</sub> nanocomposite films.



Figure 6. Appearance of the sesame protein isolate (SPI)-TiO2 nanocomposite films and their calculated opacity values (Fathi et al., 2019).

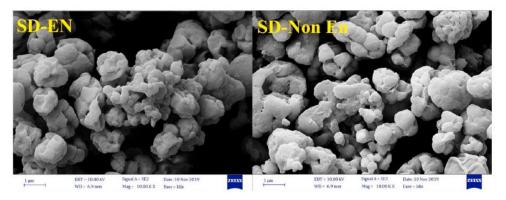


Figure 7. FE-SEM images of the SPI- Lacticaseibacillus rhamnosus microcapsules obtained by spray drying (SD) method with Microbial transglutaminase (MTGase) treatment (En); and without MTGase treatment (Non En) (Azizi, et al., 2021).

There are some reports on the use of SPI for the coating of fresh-cut fruits. Sharma et al. (2018a) have used a bilayer coating based on the SPI and guar gum on the surface of mango fresh-cut pieces to study on increase in shelf life during storage. An increase in shelf life and nutritional value of freshly chopped fruit samples and a reduction of wastage was observed. They also applied the native SPI and succinic acid, citric acid, and malic acid cross-linked SPI coating for shelf-life extension of pineapple fresh-cut pieces (Sharma et al., 2018a). They reported that the coatings from cross-linked proteins were more effective as compared to native SPI-based coatings in terms of preserving firmness,

total phenol content, carotenoids, and total soluble solids of pineapple pieces.

#### **Encapsulation wall material**

Encapsulation is defined as entrapping food additives, nutraceuticals, enzymes, bacteria, or other substances in small capsules. Carbohydrates, proteins, and lipids can be used as encapsulation agents. The SPI, due to the facile and cheap extraction, foaming/emulsifying potentials (Cano-Medina et al., 2011), fat absorption capacity (Achouri et al., 2012), high molecular weight, and good heat resistance, is a good candidate to be used as a wall material in the encapsulation of bioactive

compounds. As the first research group, Azizi et al. (2021) introduced the SPI to encapsulate the Lacticaseibacillus rhamnosus and suggested that the SPI has great potential to use as a wall material for encapsulating probiotics. The evaluation of the physical and morphological properties of the capsules revealed that it is suitable for use in food products. The SPI-stabilized microcapsules showed acceptable viability and survival after exposure to temperatures different and simulated gastrointestinal conditions. An interesting point of this research was to evaluate the possibility of enzyme cross-linking of the SPI. Microbial transglutaminase (MTGase) is an enzyme whose ability to create cross-links in proteins has been approved (Yokoyama et al., 2004). MTGase facilitates the formation of intra-molecular and extramolecular bonds between glutamyl and lysyl residues in the target proteins (Zhu et al., 2019). Depending on the sesame variety, the SPI contains lysine and glutamine at concentrations of 5.06 and 16.54 g/100 g, respectively (Fasuan et al., 2018). Concerning this hypothesis, Azizi et al. (2021) used MTGase-induced cross-linking to increase the rhamnosus integrity of L. entrapped microcapsules. Figure 7 shows the FE-SEM images of the spray-dried SPI microcapsules with and without MTGase treatment.

#### **Conclusion**

The main focus of this review paper was to explore the potential food applications of sesame proteins. The extraction/purification methods and also the chemical structure of sesame proteins were investigated. Moreover, the functional properties of the SPI were discussed in more detail, and all the applications of this protein that are reported in the literature were categorized. Improved knowledge of this protein can increase the motivation to use this new protein source as an alternative to other crops, such as soybean proteins. The information on functional characteristics of the SPI allows its use in the food industry where solubility, emulsifying/foaming properties, and gelling capacity are needed. The use as film-forming material or edible coating of foods, and also the use as encapsulation wall material, are of emerging potential of the SPI that was introduced. This paper showed that the by-product of the sesame oil

industry, as a source of an excellent plant protein, can have better valuable applications in the food industry, instead of being used in animal feed, and it is necessary to increase attention to this cheap protein source.

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## **Conflicts of interest**

Non.

#### **Disclaimer**

Non.

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