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Legumes as an alternative protein source in plant-based foods: Applications, challenges, and strategies

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ABSTRACT

Since animal proteins may pose a threat to the global environment and human health, the development of alternative proteins has become an inevitable trend in the future. Legumes are considered to be one of the most promising sources of sustainable alternative animal proteins. Legume proteins are considered to exhibit excellent processing properties, including emulsification, gelation, and foaming, which have led to their widespread use in the food industry. Moreover, legume proteins are not only taken as substitutes for meat proteins, they also play an essential role in novel plant-based foods (meat, dairy, fermented food, and fat). However, there are few comprehensive overview studies on the application of legume proteins in plant-based foods. Therefore, this review provides a general overview of the main sources, functional properties, and applications in plant-based foods of legume proteins. In addition, challenges to the application of legume proteins in plant-based foods and specific strategies to address these challenges are presented. The review may provide some references for the further application of legume proteins in novel plant-based foods.

1. Introduction

Animal-based diets are considered unsustainable, inefficient, environmentally unfriendly and unhealthy. Modern animal husbandry mainly uses grains as feed, and these are equally available as food for humans, which undoubtedly lengthens the food chain thereby causing energy loss (Semba et al., 2021). It has been reported that the average animal protein produced requires 11 times more fossil energy, 4–26 times more water, and 6–17 times more land than plant protein (Pimentel and Pimentel, 2003). Animal husbandry, brought about by animal-based diets, is already greatly detrimental to the environment of the planet as a major source of greenhouse gases. Moreover, the high fat content of meat may be associated with dyslipidemia, certain cardiovascular diseases, and cancer (Willett et al., 2019). Nonetheless, meat consumption continues to increase worldwide and it is due to a combination of increased consumption levels and population growth. By 2050, the world population will grow to 9–10 billion people, and meat proteins may not be able to meet the protein intake required for human growth and optimal health (Semba et al., 2021). As a result, plant-based diets made from plant proteins are being encouraged, and it is considered a more sustainable and healthy option. However, the unique taste, flavor, and nutrition of meat and animal dairy have made them highly desirable to consumers. Based on the background above, plant-based foods that mimic the characteristics of meat and animal dairy based on plant proteins are rapidly expanding, including plant-based meats, plant-based dairy, plant-based fats, and plant-based fermented products (Sabaté and Soret, 2014). Global retail sales of plant-based meat and plant-based dairy products are projected to reach $162 billion by 2030, significantly higher than the 2020 figures of $29.4 billion (FAO, 2022).

Legume proteins hold a remarkably important position among plant proteins, associated with their balanced and comprehensive amino acid composition. For example, soy and pea proteins have Protein Digestibility Corrected Amino Acids Score (PDCAAS) of 1.0 and 0.89, respectively, similar to high-quality proteins such as eggs and milk (Semba et al., 2021). Other legume proteins are less, but most of them are in the range of 0.5–0.7. Moreover, legume proteins have excellent

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functionality such as solubility, foaming, water and oil retention capacity, which provides a stable foundation for their wide application in the food industry (Vogelsang-O'Dwyer et al., 2021). Besides its favorable nutritional and functional properties, legume proteins also show potential in terms of sustainability. According to reports, each 100 g of legume protein produces more than 90% less greenhouse gas emissions than the same weight of beef protein (Ritchie and Roser, 2020). If beef were replaced with legumes in the U.S. diet, emissions reductions of 46–74% could be realized, freeing up more than 40% of farmland resources (Harwatt et al., 2017). However, legume proteins have several drawbacks that are hard to ignore, such as a lack of sulfur-containing amino acids, beany flavor, and allergenic problems. These issues have seriously hindered the expansion of legume proteins on a larger scale in the food industry.

Overall, legume proteins show remarkable potential for plant-based food applications, yet a comprehensive review is lacking. Therefore, this review provides a detailed account of the application of legume proteins in plant-based foods, including plant-based meat, dairy, fat, and fermented products. In addition, the challenges encountered in the application of legume proteins to plant-based foods are documented and corresponding solutions are given.

2. Sources of legume protein

Legumes are considered to be an excellent source of protein with high protein content, balanced amino acid profile and low cost. The major legume crops are soy, pea, chickpea, mung bean, lentil and lupin. There is a large amount of literature available on them, so we will give a brief overview of the first three crops here. In addition, information on some legume proteins is summarized in Table 1.

Soy is undoubtedly the most important legume crop worldwide, and its food products are becoming increasingly popular around the world. Brazil is the largest producer of soy (133 million tons), followed in order by the United States (113 million tons), Argentina (48 million tons), and others (Singh and Krishnaswamy, 2022). Moreover, soy is a protein-rich crop (35–45%) and serves as an excellent animal protein substitute. This is mainly due to the comprehensive amino acid composition and excellent gelation properties of soy protein, which also make it an important component of plant-based meat products. In addition, soy protein is also used as an important additive in meat patties, sausages, breads, etc. To improve texture, flavor and nutritional properties (Zhang et al., 2021).

Peas and chickpeas, as the second and third most important legume crops, are among the most widely grown and consumed legumes in the world. In 2022, 34.54 million tons of peas and 15 million tons of chickpeas were harvested globally, with Canada, Russia, the United States, and India being the largest producers of peas, while the latter are mainly found in India, Turkey and Pakistan (FAO, 2022; Patil et al., 2024). Peas and chickpeas are not only high and similar in terms of yield, but they are also similarly rich in protein (peas, 23–31%; chickpeas, 18–29%) (Boukid, 2021a). The pea protein lacks methionine but has high levels of lysine, while the chickpea protein lacks sulfur-containing amino acids, whereas their other essential amino acid compositions are well balanced (Grasso et al., 2022; Schneider and Lacampagne, 2000).

In addition, both pea protein and chickpea protein are hypoallergenic, which is lacking in soy protein. Apart from this, pea and chickpea proteins also have the advantage of their high yield and nutritional value thus are considered as promising alternative protein sources for the future (Ding et al., 2020; Wangorsch et al., 2020; Xing et al., 2020). However, current product development for them is still in its primary stage, mainly for incorporation into other food products as nutritional supplements such as bread and meat products, and as animal feed (Venkidasamy et al., 2019). Fortunately, the progressive trend of plant proteins will allow them to be more intensively studied and developed for their applications.

3. Functional properties

3.1. Solubility

Protein solubility is one of the most important parameters in the field of food science (Grossmann and McClements, 2023). It is closely related to the foaming, emulsification and other properties of protein (Gao et al., 2023). The solubility of proteins can be defined as the dispersion level of proteins in solvents, which depends largely on the balance between protein-protein and protein-solvent interactions (Lam et al., 2018). In addition, the solubility level of protein is affected by temperature, pH and ionic strength. Carbonaro et al. (1997) conducted an in-depth study on the solubility of legumes. Studies have shown that the solubility of legume protein increases with the increase of pH above 7.0, but the solubility near the isoelectric point will reach the lowest state. In order to solve the problem of low solubility of some legume proteins, researchers use physical, chemical and biological methods to modify proteins to improve solubility. High-intensity ultrasound is an effective means to improve the solubility of legume protein (Cui et al., 2021). The principle is to use the ultrasonic process to produce microbubbles, and then the microbubbles undergo a collision, expansion, and crushing process, resulting in the exposure of hydroxyl groups to increase the solubility of the protein (Zhi et al., 2019). Jiang et al. (2010) placed soy protein isolate under extremely acidic and alkaline conditions to promote protein unfolding, and then adjusted the pH to neutral to refold the protein, thereby increasing the solubility of soy protein isolate under extremely acidic and neutral conditions. Enzymatic method is also an

Table 1
Protein nutritional characterization of legumes worldwide.

Common names	Latin name	Protein content (%)	amino acid composition (AAC)	Reference
Soybean, Soy	*Glycine max*	30–45	It provides all nine essential amino acids (EAA), but is deficient in sulfur-containing amino acids.	Zhang et al. (2021)
Pea	*Pisum sativum*	20–25	It provides all nine essential amino acids, and lower concentrations of sulfur amino acids and tryptophan, and higher concentrations of basic and acidic amino acids.	Shanthakumar et al. (2022)
Fava bean	*Vicia faba*	27–34	It provides all nine essential amino acids, but lack of sulfur-containing amino acids and tryptophan.	Nivala et al. (2021)
Mung bean, Green bean	*Vigna radiata*	25–28	It contains sufficient quantities of all amino acids including lysine, except methionine, cystine, and tryptophan.	Du et al. (2018)
Azuki bean, Red bean	*Vigna angularis*	21–23	It provides all nine essential amino acids.	Wang et al. (2022)
Kidney bean	*Phaseolus vulgaris*	23–25	Sulfur-containing amino acids (cysteine and methionine) were limiting but the proteins were rich in acidic amino acids.	Mundi and Aluko (2012)
Lentil , Hyacinth bean	*Lablab purpureus*	21–31	It contains all essential amino acids, but is usually deficient in sulfur-containing amino acids (methionine and cysteine) and tryptophan.	Hang et al. (2022)
Chickpea	Cicer arietinum	15–25	It contains all essential amino acids, but sulfur-containing amino acids as limiting amino acids.	Zia-Ul-Haq et al. (2007)

important method to improve protein solubility. There are two main methods: enzymatic hydrolysis and enzymatic cross-linking (Gao et al., 2023). Enzymatic hydrolysis can make the protein into a variety of small peptides, resulting in the exposure of hydrophobic groups so that the solubility gradually increased (Vogelsang-O'Dwyer et al., 2022). Enzyme cross-linking binds to proteins through non-hydrolytic enzymes (mainly TG enzymes) through cross-linking. In this process, non-polar and free amino exposure will be reduced, resulting in a decrease in the hydrophobicity of the protein and an increase in solubility. In addition to the above three methods, more and more researchers have begun to combine multiple methods to improve solubility. Wang et al. (2024) improved the solubility of protein by high-pressure homogenization assisted pH adjustment. The results showed that this method was better than single high-pressure homogenization or pH adjustment, which may be due to strong electrostatic repulsion and strong shear force. The protein structure is more extensive.

3.2. *Gelation*

Gelation, as an important property of proteins, can be simply defined as a continuous network of denatured molecules interconnected to form a continuous network under specific conditions. The gap of the network structure is filled with liquid as a dispersion medium (Totosaus et al., 2002). According to the formation form of gel, it can be divided into heat-induced gel and cold-induced gel (Zheng et al., 2022). Heat-induced gelation is the most common phenomenon in food science (Guo et al., 2021). The generation of thermal gel can be explained as follows: first, the heat causes the protein molecules to unfold, the protein molecules are denatured, and then the denatured protein molecules are associated and aggregated to gel (Ferry, 1948). The production conditions of cold gel are relatively mild. It can be produced by adding salt, enzyme and other additives at low protein concentration or low temperature (Wan et al., 2021). Tofu, which people often eat, is essentially a gel formed by salt induction (Zhang et al., 2021). It is worth noting that there are many ways to induce protein gelation. In addition to the most common heating, high pressure, acid induction, urea induction, etc can induce protein gelation. However, the gels produced by them are different. For example, the gels produced by pressure induction and heat induction are very different, which may be due to the influence of hydrogen bond structure (Angsupanich et al., 1999). Researchers have done a lot of research on the gelation of legume protein. Guldiken et al. (2021) studied the effects of salt and protein concentration on the gel formation ability of lentil, yellow pea and broad bean protein concentrate under neutral conditions. The results showed that the gel network became more orderly with the increase of protein concentration or the presence of NaCl or $CaCl_2$. Hu et al. (2013) used high-intensity ultrasound to treat soy protein isolate and improved the water holding capacity and gel strength of acid-induced soy protein isolate gel. Similarly, Wang et al. (2023) treated mung bean protein with ultrasound, and the results showed that the gel properties of mung bean protein could be improved by ultrasound treatment. The appropriate addition of polysaccharides can also improve the gel properties. Lu et al. (2023) studied the effect of carrageenan on soy protein isolate gel. The results showed that the appropriate amount of carrageenan promoted the molecular interaction in the network, and the gel strength was enhanced. Overall, a considerable number of methods for gelatinization of legume proteins have been developed, and it is important that selection of legume protein type and gelatinization methods are based on the characteristic needs of the gel product.

3.3. *Emulsification*

Emulsifying properties are typical functional properties of proteins (Foegeding and Davis, 2011). Emulsifying properties of proteins are the properties that proteins can make two or more insoluble liquid dispersions, one of which is dispersed in another liquid in the form of small droplets to form a stable multiphase dispersion system (Friberg et al., 2003). In the field of food, the type of emulsion is divided into oil-in-water (O/W) or water-in-oil (W/O) emulsion according to the oil phase as the dispersed phase or continuous phase in the emulsion, with the former mainly being used for products including milk and cream, and the latter mainly for margarine (Lam and Nickerson, 2013; Zhou et al., 2021). Proteins, especially legume proteins, are natural emulsifiers. This is due to the amphiphilicity of the protein, that is, it has hydrophobic groups and hydrophilic groups (Kato and Nakai, 1980). When the protein reaches the interface, it will undergo a certain degree of denaturation, which will lead to the exposure of hydrophobic groups. Then the hydrophobic amino acids on the surface will be in the oil phase, and the hydrophilic amino acids will be in the water phase (Nishinari et al., 2014). Karaca et al. (2011) studied the emulsifying properties of soy, pea, chickpea, broad bean and lentil protein. The results showed that the emulsifying properties of different legumes were quite different. The emulsifying stability of pea protein was the worst. The emulsifying activity and emulsifying stability of soy protein isolate were the best among several legumes. Chickpea protein and lentil protein followed closely, and were expected to be alternatives to soy protein isolate to stabilize the emulsion. At present, soy protein is the most common emulsifier, and it has been studied the most. Rivas and Sherman (1984) found that the strength of the film formed by 7 S is higher than that formed by 11 S at the interface, which means that the emulsification performance is also better, and the strength of this film is not affected by salt concentration or pH. Improving the emulsification of protein has always been the research direction in the field of food science. O'sullivan et al. (2015) found that ultrasonic treatment could significantly improve the emulsion stability of pea protein, which may be because ultrasonic treatment improved the interface layer. The combination of protein and polysaccharide is also an effective method to improve emulsification. Han et al. (2023) added trehalose to soy protein isolate to prepare nanoemulsions. It was found that the addition of trehalose could significantly inhibit protein aggregation and improve its emulsifying properties, which he attributed to the fact that trehalose could cover the protein and reduce its hydrophobicity. In addition, Chen et al. (2013) studied the effect of oxidation on the emulsifying properties of soy protein isolate, and the results showed that moderate oxidation could improve the emulsion stability.

3.4. *Foaming*

In the food industry, foam is popular because it provides a unique texture and taste for food (Deotale et al., 2020). Protein as a foaming agent can promote the formation of foam and improve the stability of foam (Foegeding et al., 2006). This is closely related to the surface hydrophobicity of the protein. Higher surface hydrophobicity often results in higher foam volume (Amagliani et al., 2021). The principle can be simply explained as that the protein forms a tough film at the water-liquid interface to absorb and stabilize bubbles. Like other properties of proteins, the foaming properties of proteins are also affected by the environment such as protein concentration, pH, temperature (Damodaran, 2005). Koop et al. (2020) found that the foaming stability of protein was positively correlated with it within a certain protein concentration. At 4% protein concentration, the foaming performance is the best. Plant proteins, especially legume proteins, are alternatives to egg white proteins because of their unique sustainability and health (Zhou et al., 2021). The extraction method of bean protein often has a great influence on the foaming characteristics. Cui et al. (2020) extracted pea protein by alkaline extraction-isopoint precipitation method, and studied the effect of pH on the foaming properties at 8.5–10. The results showed that the foaming properties of pea protein became better with the increase of pH. Tontul et al. (2018) studied the effects of different drying methods on the foaming properties of chickpea protein. The results showed that the foam stability of the protein obtained by freeze-drying was 6.8 times higher than that obtained by refractive window method. Similarly, researchers have done a lot of research on improving the foaming properties of soy protein. Martínez-Velasco et al. (2018) explored the effect of high-intensity ultrasonic treatment on the foaming properties of Faba bean protein, and found that the protein after ultrasonic treatment produced larger foam volume and better stability. On this basis, Wang et al. 2022a, 2020b combined ultrasonic and pH-shifting treatment of chickpea protein, and found that this combined treatment can significantly improve the foaming properties of the protein. Shao et al. (2016) found that heat treatment (55 °C and 85 °C) could improve the foaming properties of soy protein isolate and soy protein concentrate, and the effect at 85 °C was better than that at 55 °C. In addition, mixing proteins with polysaccharides can also improve the foaming properties of proteins. YR Xie and Hettiarachchy (1998) found that soy protein isolate could improve the foaming ability under the action of xanthan gum, and the foam stability of soy protein isolate-xanthan gum complex was nine times that of soy protein isolate.

4. Application in plant-based foods

4.1. Plant-based meat

Due to the limited sustainability of meat, there has been a trend of transition from meat foods to plant-based meat worldwide. The main raw materials for plant-based meat are plant proteins, of which legume proteins occupy the majority due to their affordability and excellent processing properties (Kyriakopoulou, Dekkers and van der Goot, 2019). The key to plant-based meat is the mimicry of meat texture, flavor, and nutrition. Among them, the fibrous structure is crucial for the texture of plant-based meat, and the mechanism of its formation lies in non-covalent interactions and disulfide bonds between proteins (Zhang et al., 2023a). This fibrous structure mimics the texture of real meat, making plant-based meat more similar to real meat in terms of taste and appearance, thus increasing consumer acceptance (Zhang, et al., 2021, 2022). In addition, for legume proteins, it may not be possible to prepare plant-based meat products with a rich fiber structure by relying on a single protein alone, but rather a combination of different proteins or other compounds, such as wheat proteins, starch, edible gums, is required (Boukid, 2021b). Also, the combination provides a more complete profile of nutrients in plant-based meat products. Jiang et al. (2024) prepared different blends by combining pea protein, soy protein isolate, chickpea protein and wheat gluten, and found that the addition of wheat gluten helped in the formation of fibrous structures and that the mixture of pea protein, chickpea protein, and wheat gluten was more suitable for the preparation of plant-based meats and provided a more comprehensive amino acids. Among the many preparation processes, extrusion has become the most commonly used method for preparing plant meat due to its high productivity, energy efficiency, versatility and low cost, and the process parameters of extrusion play a key role in the formation of textures and structures (Andreani et al., 2023; Dekkers et al., 2018). In particular, high-moisture extrusion is considered to be the most promising technology due to its unique characteristics of low energy, environmental friendliness, high efficiency, and excellent product quality (Zhang et al., 2021). Plant proteins processed by high-moisture extrusion are able to achieve a fibrous structure that is very similar to meat and retains a significant amount of nutrients. Moreover, the texture of high-moisture plant meat is mainly affected by different protein raw material formulations and extrusion parameters. It has been shown that the optimal process conditions for the preparation of high-moisture plant-based meat based on isolated pea protein are 55 per cent moisture content, a barrel temperature of 175 °C and a screw speed of 200 rpm (Zhang and Ryu, 2023).

4.2. Plant-based dairy

In recent years, plant-based dairy products have become increasingly popular among consumers. Even though the nutritional value of plant-based dairy products is currently not as high as that of conventional dairy products, their unique property of being free of dairy allergens such as lactose, cholesterol, and casein is appealing to consumers (Adamczyk et al., 2022; Pua et al., 2022). The raw materials of plant-based dairy products include cereals (oats and rice), legumes (soys and peas), vegetables (potatoes), seeds (flax and hemp), and nuts (almonds, cashews) (Bridges, 2018). Among them, soy milk made from soy protein is one of the most important plant-based dairy products and is considered remarkably safe. Astolfi et al. (2020) compared 41 elements in 43 cow's milk and plant milk samples and found very low in toxic trace elements, and the idea that soy milk is the best alternative to cow or goat milk in the human diet. Processing legume proteins into plant-based dairy products may only require simple milling, or the application of ultrasound, enzyme treatments, etc., which results in the transformation of proteins from large particles to small particles (Pua et al., 2022). In addition, emulsions made from legume proteins and plant oils (mostly water-in-oil) can be used to make other types of plant-based milks, such as creams and ice creams. Wen et al. (2020) demonstrated that high internal phase Pickering emulsions made from the soy proteins and sunflower oils have high self-supporting power and stability, showing potential as solid creams.

However, for legume proteins, especially soy proteins, several technical issues may be addressed in order to prepare plant-based dairy products with organoleptic properties and nutritional values comparable to animal milk, such as beany flavor and nutrient loss (Yu et al., 2023). Several methods are known to eliminate or remove the beany flavor from soy milk, including high-temperature vacuum treatment (HTVT), Cornell heat-grinding (CHG), and Illinois pre-blanching (IPB) (Lopes et al., 2020; Sethi et al., 2016). In addition, during the production process of soy milk preparation, some key nutrients, such as proteins and isoflavones, are often lost in the residue of the substance known as soy bean residue, which makes these nutrients not integrated with the soy milk. To overcome this challenge, several innovative technological pathways are being investigated to enhance the extraction of these nutrients. The goal is to optimize and enhance the storage and application of active ingredients in the soy milk production process. As an example, the use of ultrasonic treatment can improve the stability of soy milk as well as enhance the extraction of proteins, oils and solids (Olías et al., 2023). In conclusion, there is still a need for a comprehensive understanding and study of different sources of legume proteins for the production of a tasty and flavorful plant-based dairy, including the nutritional composition and functional properties.

4.3. Plant-based fermented food

Nowadays, fermented foods have become an important part of different dietary cultures and foods fermented with plant ingredients are considered to have high nutritional and functional values, as well as health benefits (Marco et al., 2017; Torres et al., 2020). For legume proteins, most of the applications of their fermentation technology are in the further processing of plant-based dairy products, such as plant-based yogurt, plant-based butter. Moreover, fermentation technology is considered as an effective means to enhance bioactive components and reduce anti-nutritional components in soys. Based on the nutritional attributes, Ahsan et al., conducted a functional study of bioactive components in fermented and non-fermented soy milk. Their results showed that the bioactive components were more active during fermentation and received higher sensory evaluation compared to non-fermented soy milk. Microbial communities in plant-based fermented foods are influenced by composition, nutrient content, fermentation duration, and physical conditions (Mathur et al., 2020). Dai et al. (2023) derived that compared to low- and high-temperature-fermented soy whey, simulated natural-temperature-fermented soy whey bacterial community had higher species richness and diversity and produced tofu with high elasticity and low beany flavor compared to low and high temperature fermented soy whey. Fermentation with probiotics is an important trend in plant-based fermented foods, and Lactobacillus, as a common probiotic, can be effective in alleviating soy allergenicity (Dai et al., 2023). Researchers have demonstrated that treatment of β-associated soy globulin by lactic acid bacteria fermentation significantly reduces the immune response to the protein, and that fermentation at higher initial protein concentrations and control of final pH are beneficial in the production of hypoallergenic soy products to reduce immune reactivity (Yan et al., 2023). Overall, although there is now a wide range of evidence to support the health benefits of fermented legumes, researchers still need to analyze the molecular level and subsequent clinical trials in depth.

4.4. Plant-based fat

In animal meat products, the fat content usually ranges from 15% to 35%. Excessive intake of long-chain saturated fatty acids and cholesterol from animal fats has been reported to lead to a range of serious metabolic disorders such as cardiovascular diseases (Ren et al., 2022). Therefore, the development of vegetable-based fat substitutes has become particularly critical. The adipose tissue in animal meat products is composed of liquid oils and solid fats in a network of connective tissue, which gives meat products their unique plasticity and elasticity (Du et al., 2023). Dreher et al. (2020) proposing structured plant-based fat substitutes as an effective way to form adipose tissue networks, they successfully generated a variety of structurally distinct lipid systems by emulsifying a fat crystal network composed of a mixture of liquid vegetable oils and solid plant-derived fats with excess soy isolate proteins, followed by transglutaminase-induced cross-linking. Colloidal systems formed from plant polymer compounds are an excellent processing technology for the preparation of plant-based fat substitutes, and common colloidal systems include hydrogels, oleogels, and emulsion gels (Du et al., 2023). Among them, emulsion gels have demonstrated excellent lipid reduction and oxidative stability and are relatively simple to prepare, and researchers have already replaced animal fats by preparing emulsion gels using protein or polysaccharide-based (Yan et al., 2023). For example, Hu et al. (2022) prepared emulsion gels assembled from soy protein, soy oil, and agar that mimic the appearance of beef adipose tissue but have a much softer texture than real meat adipose tissue. The high viscosity and softness of emulsion gels limit their formation into three-dimensional cubes (Huang et al., 2022). However, nowadays, studies have successfully prepared soy isolate protein-konjac glucomannan composite emulsion gels, which have improved the texture and rheological properties of emulsion gels, thus developing three-dimensional plant-based fat substitutes (Wei et al., 2024). The current trend in plant-based fat substitution is that a great deal of research is still needed to expand the properties of plant-based polymers for flexible application in various types of colloidal systems. In addition, a schematic diagram for the production of various plant-based food products from legume proteins has been summarized in Fig. 1.

5. Challenges and corresponding strategies in food applications

5.1. Allergenicity

5.1.1. Problem description

For legume protein products, allergenicity is a well-documented and significant problem that has hampered their development, especially for soy proteins. In 1934, the first human allergic reaction to soy was recorded (Duke, 1934). After wards soy was listed as one of the top 8 allergens, these account for 90% of all allergic reactions, and soy has been detected in at least 16 allergens (Meinlschmidt et al., 2016). Almost all allergic reactions in soy are caused by protein-mediated causes, mainly β-conglycinin and glycinin (Cordle, 2004). Soy allergy generally triggers mild symptoms, but may also result in severe reactions, including small intestinal colitis, eczema, or other IgE-mediated clinical symptoms and pathologies (Chizoba Ekezie, Cheng and Sun, 2018). Fortunately, however, studies have shown that soy protein has a high concentration threshold for causing allergies (about 100 times as high), so its allergic reactions may be relatively low (Cordle, 2004). Although other legume proteins are not as severe as soy proteins in causing allergies in humans, there are allergens present in all of them in varying degrees. For example, prolamin and 7 S globulin in lentil protein, 7 S globulin and 11 S globulin in chickpea, and prolamin and 7 S globulin in mung bean are considered to be major allergenic proteins (Zhang et al., 2024). Pea protein is generally considered to be a very hypoallergenic food source, which has led to its being widely marketed under the "hypoallergenic" label (Taylor et al., 2021). However, the truth is that pea protein allergies have been well documented. For example, vicilin, convicilin, and non-specific lipid transfer protein (nsLTP) have been

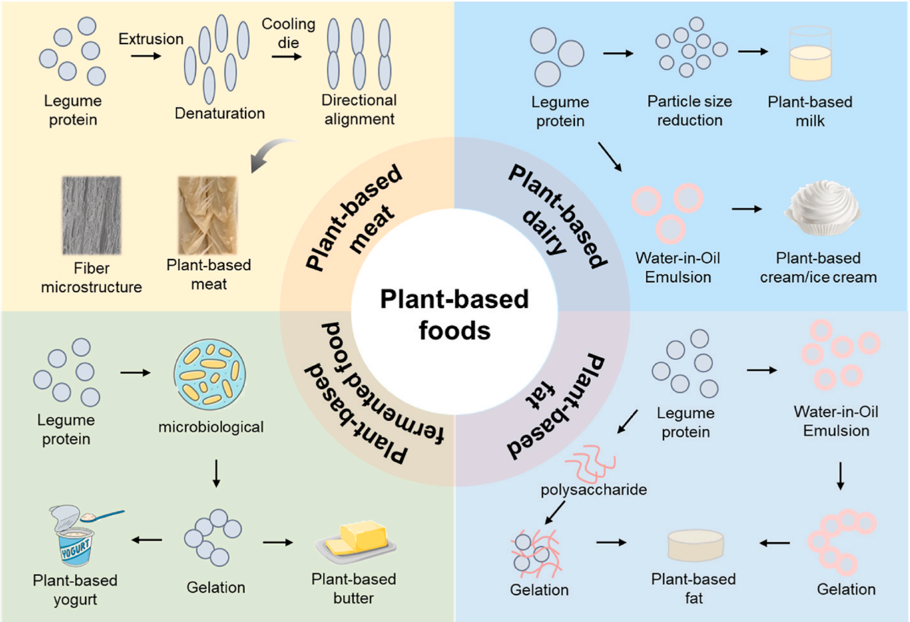

Fig. 1. Schematic diagram of the production of various plant-based foods from legume proteins.

detected as major allergens. Overall, the allergenicity of legume proteins is widespread, and a priority safety issue that warrants long-term research.

5.1.2. Strategies

In order to decrease the allergenicity of legume proteins, a numerous measures have been developed nowadays, mainly including thermal techniques, enzymolysis, fermentation, high-pressure treatment, ultrasound (Pi et al., 2021). The principle almost always lies in the utilization of thermal or non-thermal techniques to induce denaturation of proteins leading to masking or destruction of epitopes, thus reducing allergenicity (Meinlschmidt et al., 2016). Thermal treatments cause proteins to denature and make structural changes, however with uncertainties associated with being thermal. Since thermal treatment may mask or destroy epitopes, but may also expose or create new epitopes thereby increasing allergenicity, or the epitopes may remain unchanged resulting in no change in allergenicity (Chizoba Ekezie, Cheng and Sun, 2018). Additionally thermal treatments have a high potential to alter the nutritional properties and sensory aspects of proteins, which makes this method of allergenicity reduction unattractive. Therefore, researchers are actively exploring new non-heat treatment methods to reduce the allergenicity of legume proteins. Meinlschmidt et al. (2016) demonstrated that enzymatic digestion is an effective method to reduce the allergenicity of soy proteins and that papain is the optimal hydrolyzing enzyme. However, the altered protein functional properties and the production of bitter peptides brought about by enzymatic hydrolysis have prevented the intensive application of this method. In addition, high pressure treatment resulting in structural changes and reversible folding is believed to definitively reduce the allergenicity of legume proteins. Li et al. (2012) showed a 48.6% reduction in the allergenicity of soy protein by treating it with high pressure at 300 MPa for 15 min. However, the high-pressure treatment was found to be unable to destroy the epitopes sufficiently, resulting in a non-satisfactory reduction of allergenicity. Overall, there are various methods for allergenicity reduction of legume proteins, but all of them may have drawbacks, resulting in the absence of a completely feasible technique to present day. Therefore, combining multiple methods may be a more effective approach to reducing the allergenicity of legumes than a single method (Pi et al., 2021).

5.2. Digestibility

5.2.1. Problem description

Besides the nutrient content of proteins, protein digestibility is also an important criterion for determining whether it meets human nutritional requirements. Especially in the case of plant-based food products that are used as a substitute for animal proteins, a comparison of digestibility with animal proteins is of particular importance. In general, the digestibility of legume proteins is lower than animal proteins, which is attributed to the structural differences between the two as well as to the presence of antinutrients in legume proteins, such as phytic acid, trypsin inhibitors, and lectins (Mulla et al., 2022). In addition to the comparison of the proteins themselves, the comparison of the plant-based foods in which they have been processed is also essential. Zhou et al. (2021) compared plant-based beef and beef in an in vitro simulated stomach and intestine, respectively, and found that the plant-based beef had a higher digestion rate in the stomach, while it was digested more slowly than beef in the intestine. The reason for this may be due to the different structures of the two, as plant-based beef is composed of globular proteins while beef is constructed of fibrous proteins. The research of Xie et al. (2022) further backed up this result and found that plant-based meat released more bioactive peptides upon digestion. Beyond the comparison to animal proteins, soy proteins seem to consist of greater advantages compared to other plant proteins. For example, soy protein (95%) is more digestible compared to wheat protein (91%) (Schaafsma, 2000). Additionally, differences in the types of legume proteins may also lead to differences in digestibility, with common legume proteins ranking in order of digestibility as soy, lentil, chickpea, and common bean (Ohanenye et al., 2022). The high digestibility of soy protein is thought to be due to the fact that it has the lowest level of β-sheet structure, which is negatively correlated with the digestibility of legume proteins (Carbonaro et al., 2015). Overall, although the digestibility of legume proteins is better among plant proteins, however there are still differences when compared to animal proteins, both in the protein itself and the products.

5.2.2. Strategies

Several methods have been developed to improve the digestibility of legume proteins with a focus on their structure (internal factors) and the presence of anti-nutritional factors (external factors). In traditional daily life, cooking, roasting, and milling are used to varying degrees to improve the digestibility of legume proteins. For example, Ma et al. (2011) treated different legume proteins using heating (boiling at 90 °C for 20 min) and roasting (baking at 80 °C for 1 min), and found that both heat treatments resulted in a significant reduction in trypsin inhibitor activity in lentil, chickpea and pea proteins. Moreover, novel methods have been applied to improve protein digestibility such as ultrasound, high pressure treatment, pulsed electric field, and microwave. By using ultrasound in an aqueous medium, temperature and pressure increase and shear energy is generated in cavitation, which may disrupt the non-covalent interactions and disulfide bonds of proteins, thereby converting aggregated proteins into uniformly small sized entities (Karki et al., 2010). Furthermore, ultrasound also inactivates anti-nutritional factors such as trypsin inhibitors, all of which enhance the digestibility of legume proteins. Other techniques similarly inhibit or inactivate anti-nutritional factors or disrupt protein structure to enhance digestibility. In general, both traditional and novel methods enhance the digestibility of legume proteins, but each has its own advantages and disadvantages. Traditional methods require a low technical threshold, low cost and do not require sophisticated instrumentation, but are time consuming and do not result in a limited increase in digestibility (Ohanenye et al., 2022). In contrast, the novel methods require a higher level of technology and are more costly, but are generally more efficient and provide a higher digestibility of the products.

5.3. Beany flavor

5.3.1. Problem description

Beany flavor is a typical problem with legume proteins, which has hampered the development of legume proteins and their products. The beany flavor of legume proteins originates from polyunsaturated fatty acid derivatives, which are catalyzed by LOX (lipoxygenase, EC 1.13.11.12) to produce hydroperoxide derivatives and are once again degraded to volatile compounds including alcohols, aldehydes, ketones, acids, amines (Wang et al., 2021). In detail, these volatile compounds include fatty alcohols, fatty aldehydes, fatty ketones, furans and their derivatives, and aromatic compounds, of which there are more than 20 (Roland et al., 2017). These volatiles usually have extremely low thresholds, generally in the range of parts per million (ppm) or even parts per billion (ppb), such as (Z)-3-Hexenal, an important source of beany flavor in soy, which has a threshold of 0.00012 ppb (DeMan et al., 1999). The beany flavor of legume proteins has seriously threatened their use in food Industry, especially in plant-based foods. As the main proteins in plant-based meat products, legume proteins (mainly soy and pea proteins) produce an unpleasant beany flavor that diminishes consumer acceptance of products. Similarly, the main problem with plant-based dairy products based on legume proteins is the beany flavor, such as in soy milk where the main source of off-flavor is the endogenous enzyme lipoxygenase (Duarte et al., 2022).

5.3.2. Strategies

Beany flavor being a major hindrance to the application of legume proteins in the food industry, researchers have sought some ways to tackle it, which are mainly based on three perspectives: biological, physical, and chemical. Among them, biotechnology includes genetic engineering, enzyme treatment, and fermentation, all of which are considered to be effective ways to completely remove beany flavor (Wang et al., 2021). Breeding new soy varieties deficient in LOX (LOX1, LOX2, LOX3) through genetic engineering techniques is effective in blocking the degradation of unsaturated fatty acids (Yang et al., 2016). However, other oxidative degradation pathways (auto-oxidation and photosensitized oxidation) of unsaturated fatty acids may still result in the appearance of beany flavor. Enzymatic treatments are designed to degrade volatiles into their corresponding acids, thus removing beany flavor (Sethi et al., 2016). Fermentation has a similar mechanism to enzyme treatment, which uses microorganisms such as Lactobacillus, yeast, and edible tamarinds to convert beany flavor compounds into other substances (Tao et al., 2022). Physical techniques include heat treatment, flavor masking, and pulsed electric fields. Lv et al. (2011) found that thermal treatment of soymilk by subjecting it to hot water at 80–100 °C significantly reduced the activity of LOX and maintained it at 38–57%. However, while diminishing the beany flavor, the aroma substances of the legume proteins themselves were also significantly reduced, which resulted in the loss of the original sensory flavor of the legume proteins. The use of β-cyclodextrin for beany flavor masking is a very effective and economical technique. Specifically, the special hydrophobic cavities of β-cyclodextrin are utilized as a cage-like supramolecular structure to encapsulate the beany flavor compounds (E.-J. Lee et al., 2020). Chemical methods mainly including pH adjustment, reducing agent treatment, organic solvent treatment. However, chemical methods usually suffer from inefficiency, chemical residues, and alteration of the original flavor of the food, thus chemical methods are rarely used or combined with other methods (Xiang et al., 2023). In general, most of the methods are essentially to reduce the conversion of unsaturated fatty acids by eliminating or inhibiting the activity of LOX, while a few are to directly remove or mask the beany flavor compounds. It is also worth noting that the processing conditions during the processing of plant-based foods can also have an impact on the beany flavor. Zhu et al. (1996) measured LOX activity at different extrusion temperatures and found that an increase in extrusion temperature significantly inactivated LOX, thereby preventing the continuation of the beany flavor.

5.4. *Texture*

5.4.1. *Problem description*

The texture of plant-based foods is one of the most important indicators of consumer acceptance, especially for plant-based meat products, as its texture is expected to be similar to meat. However, the texture and mouthfeel of current plant-based meat products still fall short of meat. The texture of plant-based meat is usually described as fiber degree (degree of texture), tensile strength, hardness, elasticity, juiciness, and tenderness (Zhang et al., 2023b). Among them, the fiber degree is considered to be the most important metric, which is typically expressed by the ratio of the vertical and parallel shear directions of the texture analyzer, along with microscopic observation. Compared with plant-based meat, meat has a more aligned fiber orientation, a denser and more compact structure, and a layered structure (Zhang et al., 2023a). The specific fiber structure of real meat also gives it a high-water holding capacity, which results in excellent juiciness and tenderness (Frank et al., 2022). Besides a machine judgment, sensory evaluation provides a direct determination of the acceptability to the consumer. The texture of plant-based meat is judged by the oral processing and perception of human. The first of the mouth usually perceives hardness, and as the physical state of the plant-based meat changes, the water or oil within it is released thus being fully considered for juiciness and tenderness (Cordelle et al., 2022). Lee et al. (2021) used 20 trained personnel to perform sensory evaluations of plant-based meat and beef, and found that while plant-based meat was harder, the general acceptability was comparable to that of beef, with similar elasticity and chewiness. In addition, plant-based dairy products are often compared to animal dairy. However, the distribution of dispersed-phase proteins, lipids, starch, and other particles in continuous-phase water could lead to undesirable textures such as chalkiness and grittiness in plant-based dairy products (Moss et al., 2023). Besides, plant-based yogurts have been described as having a thin and a watery mouthfeel (Greis et al., 2023). Similarly, the texture of plant-based fats made from legume proteins, including lubricity and friction properties, may differ from animal fats (Nourmohammadi et al., 2023). However, the lack of relevant studies has resulted in the evaluation of plant-based fats without systematic and in-depth analysis.

5.4.2. *Strategies*

Texture improvement of plant-based foods is mainly carried out in two ways: selection of raw materials and modification of processing techniques. For legume protein, its protein structure is mostly spherical, which is greatly different from the rod-shaped protein of meat fiber. Therefore, in order to simulate the fibrous structure of meat, substances with chain-like structure are compounded and added to legume protein, including wheat protein, starch and edible gum (Zhang et al., 2023b). In addition, fungal proteins with natural filamentous structure seem to be an attractive option (Malav et al., 2015). The addition of these exogenous substances not only improves the fiber structure of plant-based meat, but also enhances its water-holding capacity thus allowing it to become juicier. Besides the fiber structure, the fat naturally contained in meat is also a major problem that needs to be addressed in plant-based meat. Strategies available nowadays involve the direct addition of plant oils, including sunflower, soy, and coconut oils, during the processing of plant-based meat (Cho et al., 2023). However, the addition of oil during processing may weaken the fibrous structure of plant-based meat, thus a balance needs to be considered. It is noteworthy that most of the currently commercialized plant-based meat products are formed by adding plant fats after the protein is textured and then re-compacted and restructured by adding plant fats (Chen et al., 2022). Moreover, while plant fats here can be added directly to plant oils, a more advanced method is to use plant-based fats made from plant proteins or plant oils. Plant-based meat products made by the latter method appear to have a more similar texture to meat, yet as mentioned above plant-based fats still need to be further explored in research. For current plant-based meat products, most of them are produced by extrusion. High-moisture extrusion is an advanced manufacturing technique in extrusion, which produced products with 50–80% moisture and rich fiber structure, which is comparable to meat (Zhang et al., 2023a). In addition, according to different raw material characteristics and product requirements, adjustments to extrusion parameters including extrusion temperature, moisture content, screw speed, could produce plant-based meat products with different textural characteristics. For plant-based dairy products, the addition of thickening agents such as locust bean gum and pectin to legume proteins may improve their textural characteristics (Moss et al., 2023). In addition, the introduction of fermentation process in the processing of plant-based dairy would improve its product texture significantly. Mefleh et al. (2022) fermented chickpea proteins for the production of plant-based milk by three fermenters, Streptococcus thermophilus (ST), co-cultures of ST with Lactococcus lactis (STLL), and co-cultures of ST with Lactobacillus plantarum (STLP), and found that the fermentation increased the consistency, viscosity, and creaminess of products. Furthermore, the challenges for the application of legume proteins in plant-based foods and the corresponding solution strategies have been summarized in Fig. 2.

6. Conclusion

In conclusion, legume protein is considered as a high-quality sustainable protein that exhibits comprehensive nutritional profile,

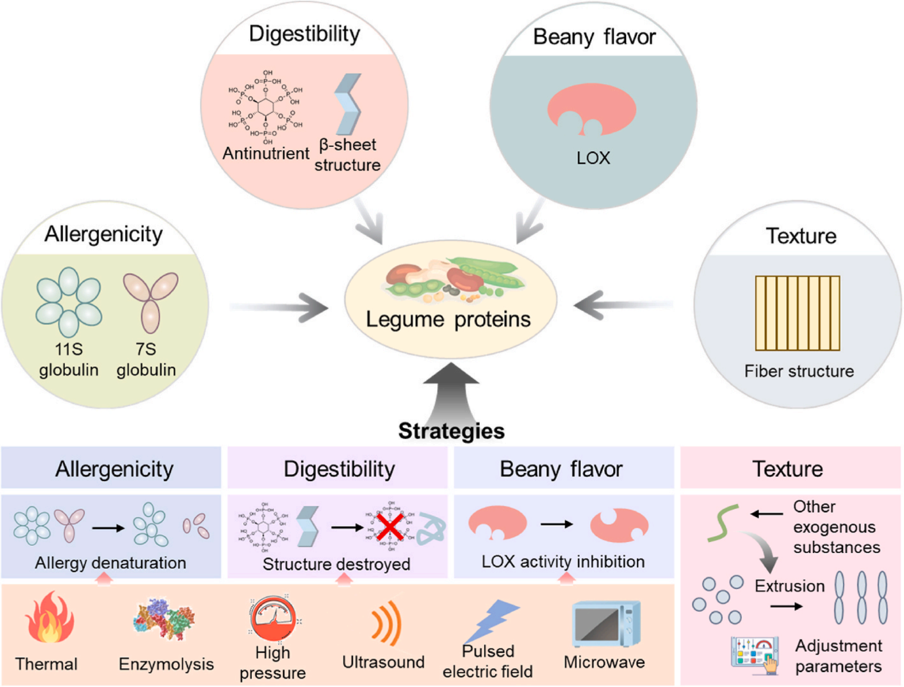

Fig. 2. The challenges for the application of legume proteins in plant-based foods and the corresponding solution strategies.

excellent processing characteristics, and environmental friendliness. As for the development of plant-based food, it has an effective role in reducing the burden on the environment and resources of the planet and in strengthening human health. The combination of legume proteins and plant-based foods may open up new opportunities for the development of the food industry. However, many of the current challenges with legume proteins need to be further explored and addressed. These challenges are mainly protein allergenicity, digestibility, beany flavor and product texture. Besides these, the innovative processing methods, relevant policies and standards, cost, and novel protein exploration are certainly critical challenges for the application of legume proteins in plant-based foods. In particular, it is worth noting that among the legume proteins, only soy and pea proteins have been well understood in the field of plant-based foods. The other legume proteins are considered to be poorly developed and therefore in need of more in-depth research focusing on their structure, functional properties, safety, and nutritional properties.

CRediT authorship contribution statement

Xin Zhang: Conceptualization, Writing – original draft. **Zhaonan Zhang:** Writing – original draft. **Ao Shen:** Writing – original draft. **Tianyi Zhang:** Writing – original draft. **Lianzhou Jiang:** Visualization, Investigation. **Hesham El-Seedi:** Visualization, Investigation. **Guohua Zhang:** Visualization, Investigation. **Xiaonan Sui:** Supervision.

Declaration of competing interest

The authors have no conflicts of interest to be declared.

Data availability

Data will be made available on request.

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