Plant Proteins for Future Foods

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Protein calories consumed by people all over the world approximate 15–20% of their energy intake. This makes protein a major nutritional imperative. Today, we are facing an unprecedented challenge to produce and distribute adequate protein to feed over nine billion people by 2050, in an environmentally sustainable and affordable way. Plant-based proteins present a promising solution to our nutritional needs due to their long history of crop use and cultivation, lower cost of production, and easy access in many parts of the world. However, plant proteins have comparatively poor functionality, defined as poor solubility, foaming, emulsifying, and gelling properties, limiting their use in food products. Relative to animal proteins, including dairy products, plant protein technology is still in its infancy. To bridge this gap, advances in plant protein ingredient development and the knowledge to construct plant-based foods are sorely needed.

Keywords: plant proteins ; future foods ; animal alternatives ; nutrition

1. Introduction

With global populations projected to increase above nine billion people by 2050 [1], we face an unprecedented challenge to produce and distribute adequate food to all of mankind. Apart from first meeting our calorie needs, the second most important macronutrient needed for human survival is protein. Protein production is a major concern because traditional animal protein sources require an intensive amount of land and resources [2]. Plant-based proteins represent a promising solution due to their long history of crop use and cultivation, lower cost of production, and easy access in many parts of the world. Plant proteins are also more environmentally sustainable [3]. However, in addition to lower protein quality, plant proteins also have comparatively poor functionality, defined as poor solubility, foaming, emulsifying, and gelling properties, limiting their use in food products. Relative to animal proteins, including dairy products, plant protein technology is still in its infancy. To bridge this gap, advances in plant protein ingredient development and the knowledge to construct plant-based foods are sorely needed.

This review is an attempt to stimulate interest and presents a roadmap to accelerate plant protein science and technology, focusing on plant protein ingredient development and future food creation (**Figure 1**). In each area, the current state of the art is briefly presented, and new research directions are highlighted. The purpose of this review is not to replicate what has been done, but to inject fresh ideas and to foster new thinking. Readers interested in more detailed discussions about various plant protein topics are referred to prior excellent reviews [4][5][6][7][8][9]. This paper focuses on manipulating plant protein structures during (a) protein extraction, (b) fractionation, and (c) modification. To create novel plant-based foods, important considerations such as protein–polysaccharide interactions, the inclusion of plant protein-generated flavors, and some novel techniques to structure plant proteins are discussed. Finally, the attention to nutrition as a compass to navigate the plant protein roadmap is also considered.

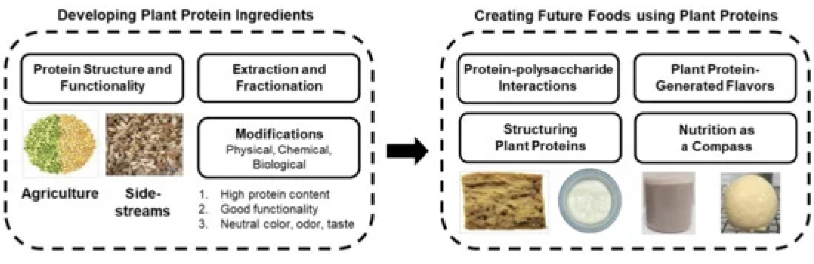

Figure 1. A roadmap to accelerate plant protein science and technology, focusing on plant protein ingredient development and future food creation.

Although the focus of this review on plant proteins is in the context of advanced nations, it is important to recognize that over one billion people may suffer from protein deficiency [10]. This problem may be more apparent in South Asia and

Africa. We therefore hope that this review will also stimulate scientists to consider how they may develop low-cost alternative protein sources for consumption by people in developing nations.

2. Creating Future Foods Using Plant Proteins

The previous sections have described the gaps in plant protein ingredient science and technology. However, we usually eat foods, and not individual ingredients. After obtaining highly functional plant proteins, the challenge is to transform these ingredients into delicious and nutritious foods. The following sections describe some important factors: the role of protein–polysaccharide interactions, the ability to structure plant proteins into fibers and gels, the inclusion of flavors derived from plant proteins, and nutrition to guide the development of plant-based foods.

2.1. Protein–Polysaccharide Interactions

Most foods are a complex mixture of various components. In addition to proteins, polysaccharides make up the predominant component in most plant-based ingredients. Polysaccharides are sugar polymers linked by glycosidic bonds and include a vast family such as starch, cellulose, pectins, agars, carrageenans, alginates and gums [11]. By capitalizing on the natural polysaccharides found in many plant protein sources, less-refined plant ingredients could be utilized, because polysaccharides also form the major building blocks in food products as structuring and stabilizing agents through their thickening, emulsifying, and gelling properties [12]. When used in combination with proteins, their functionality can be further expanded through mutual biopolymer interactions [13]. Hence, there is great interest in understanding and controlling protein–polysaccharide interactions to design plant-based foods such as plant-based milks, ice cream and pudding.

From a search of the literature, a total of 49 articles relating to plant proteins with polysaccharides were found published between 1990 and 2021, with the majority (30 articles) published in the last five years. Although this is reflective of the present emphasis on plant protein research, the number of studies is still a small fraction of the entire plant protein research field. Hence, there is a great opportunity to explore deeper into this area. In addition to advancing basic knowledge, polysaccharides could help overcome some functional shortcomings of plant proteins, with the potential to replace animal proteins [14][15]. The following sections summarize the key movements in this area.

The solubility of plant proteins is relatively low; therefore, the addition of polysaccharides has been employed to improve overall biopolymer solubility, and this is often coupled with a processing or modification step. Some examples include simple complexation [16][17], sonication [18][19] and conjugation [20][21][22]. Most notably, some authors report that the biopolymer solubility improved close to the protein isoelectric point [18] with minimum protein solubility shifting towards more acidic regions [16]. This is likely due to a change in net biopolymer surface charges upon complexation and modification. The shift in the apparent biopolymer isoelectric point will be useful for developing acidic beverages with high protein content, to reduce the precipitation of plant proteins. This strategy deserves further examination such as including other types of processing methods.

In addition to solubility, polysaccharides also improve the viscosity [23][24], foaming [17][25][26][27], emulsifying [28][29][30][31][32][33][34][35][36] and gelling [37][38][39][40][41][42][43] properties of plant proteins. Although the alteration of biopolymer interfacial properties would have obvious effects on foaming and emulsifying properties, an interesting approach is to leverage on the poor solubility of plant proteins, to create insoluble plant protein–polysaccharide particles as Pickering emulsion stabilizers [32][33]. Furthermore, it is important to note that processing plays an important role. For example, the thermal treatment of plant protein–starch mixtures led to a mixed protein–starch gel network [44][45], whereas high-pressure processing resulted in starch granules remaining intact and ungelatinized, acting as a filler in the pressure-induced protein gel matrix [42]. High-pressure processing can also kinetically arrest protein–polysaccharide phase separation through pressure-induced gelation, because the transmission of hydrostatic pressure is quasi-instantaneous compared to thermal gradients found in conventional heat processing. This suggests promising directions to explore further.

Polysaccharides do not always improve plant protein performance [46]. In some cases, it may even worsen their properties (e.g., reducing solubility and foaming capacity due to the formation of insoluble electrostatic complexes [47]). More work is needed to understand how these situations occur. In addition to environmental factors such as pH, biopolymer concentrations and ratio, and ionic strength of the system, some intrinsic factors influencing protein–polysaccharide interactions include the shape of the plant proteins. For example, globulins are spherical and highly charged compared to the more extended and charge-diffused gliadins; these differences can affect the interactions and phase separation with various polysaccharides [48]. Another important deliberation is the natural state of the protein. As discussed previously, most commercially available plant protein isolates tend to be largely denatured, and it has been shown to affect the interactions and resultant properties with polysaccharides [49]. To round off this section, other active research areas include complex coacervation [50][51][52][53][54][55][56] and applications in encapsulation [57][58][59][60].

3. Structuring Plant Proteins

3.1. Formation of Fibrous Structures

Replicating the characteristics of muscle tissue comprising muscle fibers, connective tissue, and adipose tissue arranged into complicated hierarchical systems with viscoelastic textural characteristics has proven challenging. The physicochemical and sensory characteristics of traditional meats are largely determined by the structural arrangement of these tissues. During cooking, the thermally unfolded proteins are cross-linked into a continuous gel structure [61]. The firmness and elasticity of the gel are due to the increased hydrogen bonding during cooling. These structural components are responsible for adhesiveness, viscoelasticity and juiciness [62]. In the plant protein industry, the widely used plant proteins are soy, pea and wheat owing to their availability, cost, and processing functionality. Plant proteins are globular, which does not allow for the formation of a meat-like fibrous texture. Having said that, physical modifications such as extrusion and fiber spinning are required to convert native globules into fibers. Soy (140–375 kDa) and pea (150–400 kDa) have a high molecular weight and high surface hydrophobicity, which undergo structural alterations to form polymers during physical modification, and can therefore be texturized to produce products with textural qualities comparable to meat. Mung bean and chickpea isolates presented good gelling properties and formed heterogeneous and porous networks when mung bean flour was extruded to make meat analogue products [63]. Wheat has a significant amount of gluten and possesses special film-forming characteristics that result in meat-like fibers [64]. Wheat gluten is also used in combination with legume proteins, which contributes to meat-like chewiness [65]. Due to the physicochemical differences between animal and plant proteins, it is difficult to reproduce the complex structure of meat fibers, i.e., highly organized fine texture and the water-binding capacity of meat to give plant-based alternatives a meat-like mouthfeel. A potential method to create fibrous structures is thermomechanical processing.

The fibrous structuring of plant proteins using thermomechanical processing can be classified under two main principles. The first principle is based on phase separation within a multi-phase protein mixture [66][67]. The dispersed phase acquires a spherical droplet morphology under interfacial tension and undergoes deformation–elongation–solidification to form anisotropic structures in the direction of the applied shear [68][69]. A continuous protein phase possessing intrinsic properties (e.g., molecular composition, structure, and conformation) can acquire anisotropy during structuring, but does not have a dispersed phase that may impart structural anisotropy to the protein system. For instance, Krintiras et al. reported that soy protein isolate could be dispersed in a continuous wheat gluten matrix to form anisotropic structures, after shearing in a Couette cell [70]. In recent years, Mattice and Marangoni and Chiang et al. developed a more affordable technique using less sophisticated equipment, known as protein mechanical elongation methods [68][71]. These methods demonstrated potential in forming anisotropic structures using zein or wheat gluten by stretching and orientating the fine fibrils of the proteins (**Figure 2**). Further studies are recommended to understand the suitability (e.g., self-assembled networks) of protein ingredients, and to optimize the process conditions.

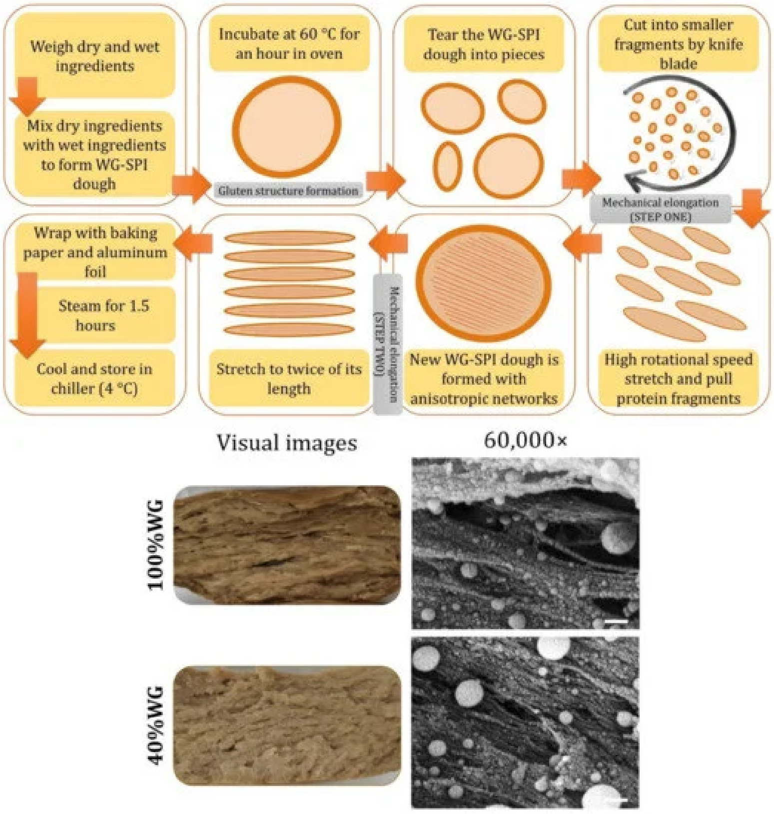

Figure 2. Schematic illustration of the mechanical elongation method in two steps to produce meat analogues and resultant microstructures. Adapted from [68] with permission.

The second principle is based on the complex conformational changes and molecular interactions of the protein upon thermomechanical processing under high-moisture (40–80%) conditions such as extrusion [72]. As mentioned in the previous section, extrusion causes protein unfolding, with partial uncoiling of the secondary structure and a complete loss of tertiary structure [73]. Meanwhile, the hydrophobic and free SH groups that were initially buried inside the native protein are exposed. The shearing from the extrusion aligns the uncoiled protein molecules in the direction of flow, resulting in a three-dimensional network structure [66]. The texturization phase inside the cooling die initiates the solidification and formation of fibrous structure through the inter- and intra-molecular aggregation of the proteins. Many studies had been conducted on different sources of plant proteins. In recent years, Chiang et al. studied the effect of animal proteins (i.e., Maillard-reacted beef bone hydrolysate, MRP) with soy protein and wheat gluten to form extruded meat alternatives [74]. The inclusion of MRP not only increased the protein content of the meat alternatives, but also increased the sensory profiles of the meat alternatives. Further work could focus on using meaty flavors generated from plant proteins to increase the protein content and flavor profile of meat analogues.

3.2. Formation of Gels

In addition to anisotropic fibrous structures, many foods are structured in the form of homogenous gels. These include yogurts, cheeses, tofu, tempeh, etc. Protein gelation occurs via various mechanisms. Globular protein gelation involves protein denaturation, aggregation, and network formation, whereas casein gels proceed via the aggregation of casein micelles. There are different methods to induce gel formation, including heat gelation, cold gelation by pH change (acidification, pH shift, fermentation), the addition of salts, enzymatic cross-linking, or pressure-induced gelation. Various types of gels can also be formed, such as hydrogels, oleogels (oil gels), aerogels and emulsion gels. Readers are referred to the excellent review by Cao and Mezzenga for a comprehensive examination of food gels [75].

Plant protein gelation has long been utilized in traditional foods such as tofu and tempeh. One active area is the use of other plant proteins beside soy to produce traditional foods. Some examples include tofu and bean curds made from pea and various legumes [76][77][78]. Another active frontier is the development of plant-based yogurt and cheese analogues [79]. The wide variety of cheese styles with different textures and melt-stretch properties require different approaches to create. Some workers incorporate both tofu- and cheese-making steps involving the coagulation, pressing and fermentation of curds [80]. A recent study explored the use of zein to provide stretchability in plant-based cheese [81]. Traditional methods may not be optimal for plant protein ingredients; therefore, this presents opportunities to rethink the processes. For example, plant-based yogurt products presently adopt the traditional fermentation of plant-based milks [82]. Often, the acidified plant protein gels are weak and experience phase separation [83]. Sim et al. demonstrated a novel approach to structure plant-based yogurts using high-pressure processing [84], enabling separate operations to generate flavor and texture (**Figure 3**). In addition to creating plant-based animal alternatives, new product categories and unit operations should be explored.

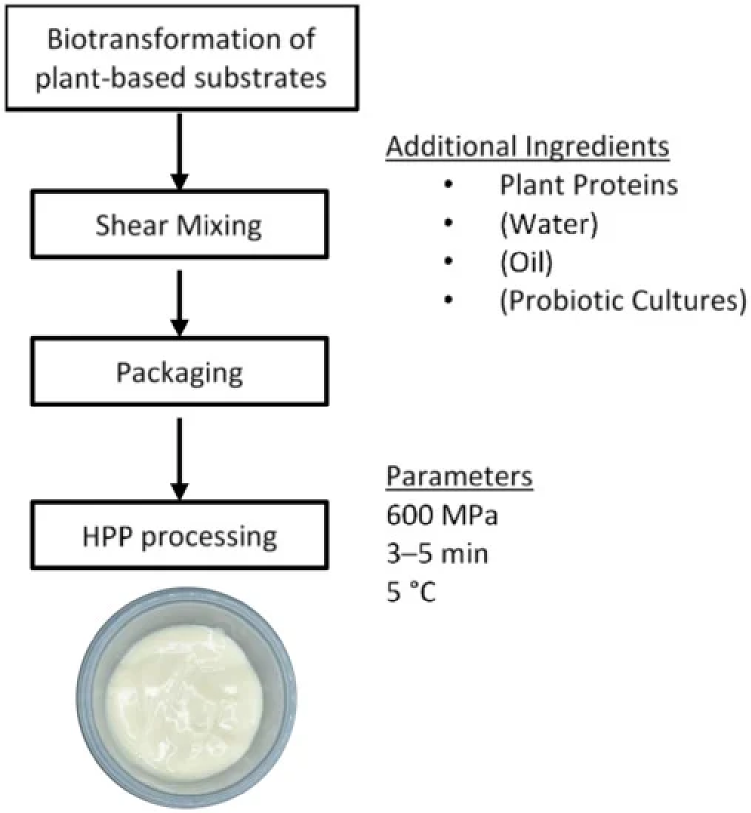

Figure 3. A proposed plant-based yogurt-making process using biotransformation for optimized flavor production and high-pressure processing (HPP) for consistent texture generation. From [84].

Notably, most traditional foods structured by proteins are in the form of either hydrogels or emulsion gels. This is unsurprising, because proteins tend to have biological activity only in aqueous environments. In the design of future plant-based foods, a main challenge is mimicking the texture and mouthfeel of animal-based fats, because most plant-based lipids exist in the form of liquid oils. Solid fats also contain saturated fats of which excessive consumption has been associated with elevated cardiovascular disease risk and could lead to other health complications [85]. In contrast, oleogels have been found to reduce postprandial insulinemia and lipidemia [86][87][88][89]. By structuring plant oils into oleogels, plant-based lipids could be made to behave similarly to animal fat, for example, in plant-based meats [90]. The behavior of oleogels in structured plant proteins will be an important area of study, for example, to develop unique marbling in high-value meat analogues such as Wagyu beef. Proteins do not easily form networks in oil; therefore, it is challenging to use proteins as oil structurants. As such, most approaches use indirect methods such as foam-templated, emulsion-templated, hydrogel-templated, and solvent exchange procedures [91]. More work is needed on protein-based oleogels, especially using plant proteins.

2.3. Flavors Generated from Plant Proteins

Flavor is one of the sensory attributes that affects a consumer’s eating quality and food purchasing decision. Numerous studies on meaty flavor chemistry have discovered thousands of volatile compounds from meat or model systems consisting of meat ingredients. Due to the rising trend in alternative proteins, there is interest in developing this meaty flavor from non-meat sources such as plant proteins. These meaty flavors can be generated via the Maillard reaction, a process whereby free amino compounds (e.g., amino acids or peptides) and reducing sugars (e.g., pentoses or hexoses) are reacted together under specific conditions to produce melanoidins [92]. The plant proteins are broken down into amino acids and peptides through enzymatic hydrolysis to generate these meaty flavors.

The most abundant flavor compounds formed during the Maillard reaction are aliphatic aldehydes, ketones, diketones, and lower fatty acids [93]. However, heterocyclic compounds containing oxygen, nitrogen, sulfur, or combinations of these atoms are much more numerous and play a significant role in the flavor development of thermally processed foods. The development of a meaty flavor is often influenced by reacting sulfur-containing amino acids (e.g., cysteine) with reducing sugars, where pentoses such as ribose or xylose are preferably used [94]. The chemical reaction between cysteine and reducing sugars is believed to be the main pathway for the formation of meaty flavor for most food products. The dicarbonyl compounds formed during the Maillard reaction catalyze the Strecker degradation of cysteine to generate mercaptoacetaldehyde, acetaldehyde and hydrogen sulfide as the primary degradation products [95]. These Strecker degradation products then start a series of reactions that lead to the formation of meaty flavor compounds.

There have been published reports using several plant proteins to generate meaty flavors such as pea protein [96], quinoa protein [97], flaxseed protein [98], soybean protein [99][100], etc. Xylose was widely reported as the reducing sugar used for the Maillard reaction, except for a combination of sugars (ribose, xylose, arabinose, fructose, glucose and galactose) used by Zhou et al. when reacting with pea protein hydrolysates [96]. Based on the gas chromatography–mass spectroscopy analysis, several aroma compounds such as furans, pyrazines, ketones, aldehydes, and others were detected from the Maillard reaction products (MRPs). Both Wei et al. [98] and Fadel et al. [99] reported the identification of 2-methyl-3-furanthiol, an odorant compound characterized with a meaty, sweet and sulfurous aroma in the MRPs [92]. This compound was formed by the Maillard reaction of cysteine and reducing sugar in a model system. However, these authors also reported the addition of sulfur-containing compounds such as cysteine, taurine and thiamine together with the protein hydrolysates and reducing sugars in heat treatment for the Maillard reaction [96][97][98][99][100]. Further work could be conducted to avoid these sulfur-containing compounds and only use the free amino acids or peptides from the plant protein hydrolysates to react with the reducing sugar.

3. Conclusions

This review has presented a roadmap to accelerate plant protein science and technology, focusing on plant protein ingredient development and the creation of delicious and nutritious plant-based future foods. The areas for further improvement include plant protein extraction, fractionation, and modification. More research is also needed in understanding plant protein–polysaccharide interactions, developing different structuring techniques, incorporating plant protein-generated flavors, and improving plant protein nutritional value. An area that needs future attention is the potential impact that different forms of fractionation and improved functionality may have on its nutritional quality. Finally, although the focus has been on plant proteins, it is vital to note that we usually eat whole foods and not individual ingredients; hence, other components that make up future foods will also be needed to be considered.

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