Annual Review of Food Science and Technology

Alternative Protein Sources as Technofunctional Food Ingredients

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Annu. Rev. Food Sci. Technol. 2021. 12:17.1–17.25

The *Annual Review of Food Science and Technology* is online at food.annualreviews.org

https://doi.org/10.1146/annurev-food-062520-093642

Keywords

food system, resilience, sustainable, plant-based meat, vegan meat, plant milk

Abstract

Proteins obtained from alternative sources such as plants, microorganisms, and insects have attracted considerable interest in the formulation of new food products that have a lower environmental footprint and offer means to feed a growing world population. In contrast to many established proteins, and protein fractions for which a substantial amount of knowledge has accumulated over the years, much less information is available on these emerging proteins. This article reviews the current state of knowledge on alternative proteins and their sources, highlighting gaps that currently pose obstacles to their more widespread application in the food industry. The compositional, structural, and functional properties of alternative proteins from various sources, including plants, algae, fungi, and insects, are critically reviewed. In particular, we focus on the factors associated with the creation of protein-rich functional ingredients from alternative sources. The various protein fractions in these sources are described as well as their behavior under different environmental conditions (e.g., pH, ionic strength, and temperature). The extraction approaches available to produce functional protein ingredients from these alternative sources are introduced as well as challenges associated with designing large-scale commercial processes. The key technofunctional properties of alternative proteins, such as solubility, interfacial activity, emulsification, foaming, and gelation properties, are introduced. In particular, we focus on the formation of isotropic and anisotropic structures suitable

for creating meat and dairy product analogs using various structuring techniques. Finally, selected studies on consumer acceptance and sustainability of alternative protein products are considered.

1. INTRODUCTION

The global population is projected to rise to 9.8 billion by 2050, which is associated with an increasing demand for both macronutrients and micronutrients (U.N. 2019). In particular, the supply of a sufficient amount of protein is of great concern because many consumer-preferred and technofunctional proteins are currently of animal origin. Proteins are key structural and functional constituents of foods and are essential nutrients for enabling the body to maintain metabolic activities like maintenance, growth, and repair of its cellular machinery (Brody 1999). The production of proteins, however, is resource intensive and has a substantial impact on environmental sustainability (Reijnders & Soret 2003). Emerging data provide a conclusive link between diet, health, and the environment, which, coupled with existing planetary boundaries, requires an extensive transformation of the global food system (Caron et al. 2018). To accomplish this shift toward sustainable and healthy diets, alternatives to currently used proteins are urgently needed. In response, food science and technology research has focused on the exploration and exploitation of alternative protein sources that provide adequate human nutrition and have a lower environmental impact. These investigations include identification of new protein sources, development of novel or more efficient extraction procedures, and modification of technofunctional properties (solubility, emulsification, foaming, gelation) and organoleptic attributes (taste, mouthfeel, appearance) as well as development of innovative processing methods—especially for the purpose of texturization—and formulation approaches. In this review, we summarize the current state of knowledge surrounding this topic.

2. DEFINITION

Currently, different protein sources are considered as an alternative to existing and established ones. However, the term "alternative protein" is somewhat ambiguous and has regional connotations. It implies, for example, something different in Europe than it does in Sub-Saharan Africa, where the major share of dietary protein originates from plants and not from animals (Sans & Combris 2015). It is, therefore, reasonable to establish a definition for the term that does not simply imply non-animal-derived protein; a definition that would, for example, exclude insects. Hence, the following definition for alternative proteins is proposed: Alternative proteins are produced from sources that have low environmental impact to replace established protein sources. They can also be obtained from animal husbandry with good animal welfare.

In this review, we discuss protein sources from land plants, algae, fungi, and insects, as these sources have been shown to have low environmental impact and, moreover, in the case of insects, good animal welfare practices may be established (Clune et al. 2017, Dobermann et al. 2017, Dossey et al. 2016). This review does not cover the current progress in the development of cultured meat because the technology is still in its early stages and its potential environmental benefits have been questioned (Smetana et al. 2015). That may change in the future, however, at which point cultured meat could also be considered as a sustainable alternative protein source.

3. SOURCES

Raw materials containing alternative proteins can have either a high (e.g., legumes or insects) or low protein content (e.g., potato). For commercial use, the protein content is not necessarily a

decisive factor, as the total amount of available raw material plays an even more important role. If large amounts of a particular material are available, the total amount of protein from that source may exceed that of a material with a higher protein content. Moreover, cascade aspects, i.e., the use of other fractions of the material as predominant extraction targets, also matter to price and availability. For example, many alternative proteins are at present obtained from sidestreams of starch or oil production that were previously used in lower value applications such as animal feed (e.g., potatoes or soybeans).

Land plants and algae accumulate mainly globular storage and metabolic proteins (except for mycelia from fungus) (Wiebe 2002) and, to a far lesser extent, structural proteins analogous to the contractile filamentous proteins responsible for muscle structure and motility found in animals, especially those obtained from domesticated livestock (Madison & Nebenführ 2013). In plants, structural elements are mainly provided by polysaccharides, and movement is most often facilitated by a change in turgor pressure and not muscle contraction (Cosgrove et al. 1984, Morillon et al. 2001). Storage proteins accumulate in protein bodies and vacuoles (seeds) or in vacuoles only (tubers) and act as a nutrient reserve for the developing plant (Herman & Larkins 1999, Shewry 2003). In contrast, insects contain striated muscle tissue, but their overall size is too small for effective direct use in foods and, depending on the final application, extraneous compounds (e.g., chitin from the exoskeleton) need to be removed before further transformation into complex food matrices (Azzollini et al. 2019, Smetana et al. 2018).

The protein composition of land plants has been classified according to the fractionation scheme developed by Osborne (1907), which relates to interactions with a particular solvent. The scheme lists four general classes: albumins (soluble in water), globulins (soluble in dilute salt solutions), prolamins (soluble in aqueous ethanol mixtures), and glutelins (soluble in dilute acid/alkaline solutions or totally insoluble). Below, the general composition and properties of major alternative protein sources are summarized. Algae are excluded because their protein composition is not yet well described. Readers are also referred to the **Supplemental Material** and referenced reviews for a detailed description of protein composition (Day 2013).

- Cereal and pseudo-cereals: Proteins in wheat, maize, and rice are mainly composed of water-insoluble proteins (prolamins and/or glutelins) (Delcour et al. 2012, Shewry & Halford 2002, Veraverbeke & Delcour 2002). In contrast, proteins in oats, quinoa, and amaranth are composed of albumins and/or globulins (Janssen et al. 2017, Klose & Arendt 2012).
- Legumes and pulses: The major storage proteins in legumes and pulses are globulins and albumins, whereas the globulins are commonly the major storage proteins (often >50%), but this may vary depending on season and variety (Boye et al. 2010). The properties of these proteins have been reviewed earlier (Boye et al. 2010, Fukushima 1991, Lam et al. 2018, Singh et al. 2015). A minor fraction of storage proteins can be found in some legumes as 2S albumin fractions, which have an important role in cysteine delivery (Boulter & Croy 1997). Other albumins are metabolic proteins in the plant and may act as antinutritive compounds in legumes, such as enzyme inhibitors and lectins (Shevkani et al. 2019).
- Oilseed proteins: In oilseeds, such as rapeseed and sunflower, the major protein classes are 11S globulins followed by albumins, whereas pumpkin seeds may also contain a major glutelin fraction (Campbell et al. 2016, González-Pérez & Vereijken 2007, Pham et al. 2017). However, the concentration of each fraction varies among different pumpkin species. For example, *Cucurbita maxima* mainly contains albumins (Rezig et al. 2013).
- Potato proteins: The main protein in potatoes is the glycoprotein patatin, which is present in the tuber as an 88-kDa dimer, and various protease inhibitors with smaller molecular weights ranging from 5 to 25 kDa (Ralet & Guéguen 2000).

- Insect proteins: The proteins in insects include sarcoplasmic proteins (water soluble), myofibrillar proteins (salt soluble), and connective tissue (soluble in acid or alkaline, or insoluble). From the data published so far, it can be concluded that albumins are equal to sarcoplasmic proteins, globulins are equal to myofibrillar proteins, and glutelins are equal to connective tissue proteins. Thus, insects have a major share of myofibrillar and connective tissue proteins.

Taken together, a broad range of new materials are currently alternative proteins, and it is likely that the continued search for functional and more sustainable proteins will continue to expand the portfolio of target materials that will be subjected to subsequent extraction.

4. EXTRACTION AND FRACTIONATION

Typically, there are several sequential unit operation steps needed to liberate proteins from the interior of cells, including a first mechanical step to break cells open, followed by suitable fractionation or purification steps (Tamayo Tenorio et al. 2018). Because proteins are often not the only valuable parts of a source material, other compounds may be extracted first using process operations that have been optimized for these compounds rather than the proteins (Sardari et al. 2019). As a result, the proteins may have been exposed to adverse conditions with respect to protein functionality (e.g., high temperatures, organic solvents, pH extremes, and mechanical forces). Furthermore, other compounds are often strongly bound to proteins (e.g., polyphenols, fatty acids, saponins, and pigments), and removal of these compounds is often required for protein ingredients to have a neutral flavor and color (Lusas & Riaz 1995). Finally, fractionation or isolation of specific classes of proteins may be needed to facilitate their use in some food applications. Consequently, the design of a process composed of cascading unit operations optimized for proteins as target compounds is essential to obtain functional protein ingredients that can then be used to build novel foods. In general, an extraction cascade for proteins comprises four key steps: (*a*) tissue disruption, (*b*) protein solubilization, (*c*) protein precipitation, and (*d*) protein concentration. Because the location of proteins and the nature of the structural entity in which they reside differ between legumes, plants, microbial cells, and insects, fractionation processes need to be tailored to the specific biological characteristics of the source material.

Shown in **Figure 1** is a general schematic of an extraction cascade used to obtain protein ingredients. Depending on whether proteins or other macromolecules such as lipids or polysaccharides are the primary extraction target, a separate scheme such as a mechanical or solvent lipid extraction may be superimposed. The schemes for other targets can be interwoven into the protein extraction process (e.g., pathway 2) or done completely separately using the original biomass (e.g., pathway 1). Alternatively, if proteins are used as a primary target (e.g., pathway 3), the sidestream that is obtained after the protein solubilization and solid–liquid separation step may be used as material for further downstream processing.

It is important to note that the choice of the cascade design has a major impact on the technofunctional properties of the ingredients obtained because the unit operations involved alter material properties to varying degrees (Swanson 1990). There is still a lack of systematic studies on the relationships between the processing, structure, and functionality of proteins, and research into these areas should be intensified.

As a first step, a tissue disruption is carried out. Depending on the mechanical properties of the cellular structure in which the target proteins are embedded, an irreversible structural change, i.e., a deformation or disruption, is needed to gain access to a majority of proteins. Because this typically involves the application of high-shear stresses, strong acids, strong bases, or hydrolyzing

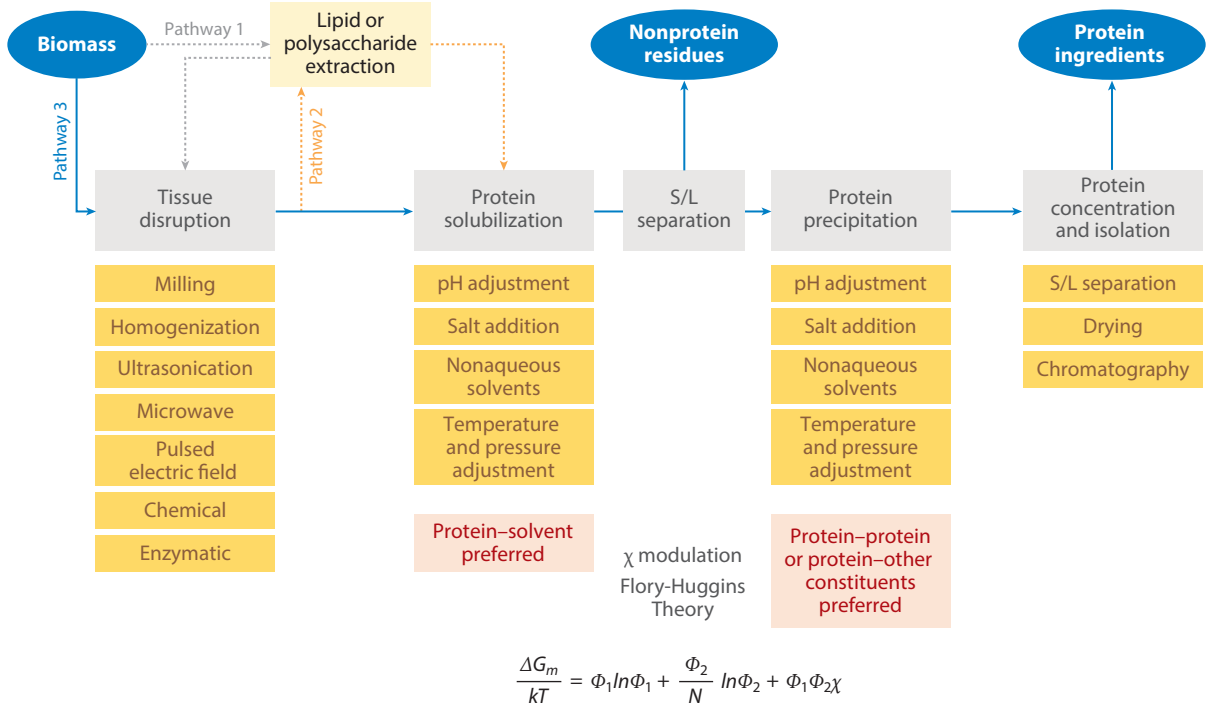

Figure 1

Schematic representation of the general design of protein extraction cascades, including tissue disruption, solid–liquid separation (S/L), protein solubilization (based on Flory-Huggins theory), protein precipitation, and protein concentration. Abbreviations: G_m, Gibbs free energy of mixing; k, Boltzmann constant; N, number of chain segments; T, absolute temperature; ϕ_i, volume fraction; χ, interaction parameter.

enzymes with a broad spectrum of activity, not only the cellular structure but the target protein molecules themselves may be modified. As such, this is one of the most crucial steps and should be designed with maximum efficiency in mind, i.e., keeping conditions as mild as possible while achieving a sufficient cellular disruption. Here, the quest for high yield can backfire, as harsh conditions may render the later-obtained proteins less functional. Some of the most common mechanical processes used include pressing, chopping, dry milling, wet milling, high-shear blending, and high-pressure homogenization (Goldberg 2008). Chemical and enzymatic processes can be used to support mechanical cellular disruption and include alkaline or acid treatments and enzymatic digestion. Moreover, freezing, drying, shock osmosis, and treatment with microwave or pulsed electric fields have been shown to facilitate the removal of proteins from cellular envelopes. The classical mechanical disruption approach subjects materials to combinations of impact and abrasion (e.g., in ball, pin, or hammer mills). The biomass may be either dry or, more commonly, hydrated. Alternatively, pressure fluctuations may be imposed (e.g., in high-pressure homogenizers or microfluidizers), which cause cavitation, creating high-shear gradients in the vicinity of collapsing bubbles that disrupt cellular envelopes.

Second, the protein is solubilized. After cell lysis, some proteins may have already been released in the suspension media, e.g., if a wet cell lysis process was used. Typically, this amounts to less than 50% of the total protein content in the cell. In a dry milling process, these soluble proteins may be released by simply adding water. However, for alternative proteins, a substantial

portion is still bound to other compounds or is present as insoluble aggregates, and the simple addition of water is ineffective in terms of solubilization. Instead, environmental conditions need to be changed so that protein–solvent interactions become more favorable than protein–protein interactions, leading to dissolution (Schein 1990). A thermodynamic description of the solubilization of polymers has been derived by Flory-Huggins by extension of the regular solution theory (Flory 1953). Contained in its mathematical description is the characteristic interaction parameter χ—the so-called Flory-Huggins parameter—which is influenced by factors such as pH, ionic strength, the dielectric constant of the solvent, temperature, and pressure. These parameters thus constitute solubilization tools, i.e., if altered appropriately, they induce solubilization (Boire et al. 2013). Following that, insoluble nonprotein residues can be removed by solid–liquid separation, e.g., using filtration or centrifugation to leave the proteins in the filtrate or supernatant.

In the third step, the protein is precipitated. After solubilization, proteins need to be separated from other soluble compounds that may still be present in the solvent (e.g., sugars or minerals). Therefore, a precipitation step is needed. This precipitation step can also be used to selectively remove only a subset of proteins to obtain specialized fractions. The tools are the same as the ones described above for the solubilization, with the difference that conditions are changed such that protein–solvent interactions become unfavorable and protein–protein interactions become favorable, leading to the formation of larger protein aggregates that eventually phase separate.

Lastly, the protein is isolated and concentrated. Once proteins have been precipitated, several technologies are available to separate the protein aggregates from the fluid phase. Both batch and continuous filtration and centrifugation processes are available and pressure or vacuum filters, sedimentation centrifuges (decanters), or filter centrifuges are commonly used for this purpose (Svarovsky 2001). To concentrate the proteins in the precipitate obtained, vacuum evaporation, spray drying, freeze drying (lyophilization), or vacuum drying may be applied (Emami et al. 2018). To purify proteins and obtain isolates rather than concentrates, chromatographic methods such as gel filtration, anion exchange, or metal-affinity chromatography may be used (Bonner 2007). Because of economic considerations, the use of these latter techniques is rare for bulk ingredients and is typically used only to obtain specialty proteins.

Taken together, a review of the literature shows that there are various approaches used to produce protein ingredients and that an individual cascade design is required depending on the nature of the source material to optimize both the yield and functionality of the final protein ingredients. More recently, alternative (mild) downstream processing approaches based on dry fractionation—optionally coupled with electroseparation—have been developed (Schutyser & van der Goot 2011). The less-refined ingredients obtained using these approaches require fewer resources (energy, water) but often have different functionalities than conventionally extracted protein ingredients. Research is currently underway to better understand how certain material properties of these new less-refined ingredients may relate to their performance in product matrices to better facilitate product design.

5. TECHNOFUNCTIONALITY AND SOLID STRUCTURE FORMATION

One of the main features of food proteins is that they serve as technofunctional ingredients in creating and stabilizing food structures (e.g., as emulsifiers, foam stabilizers, and gelling agents). As mentioned above, the extraction procedures should be carried out such that the obtained ingredients are optimally suited for a specific food application. In the first part of this section, we review the solubility, emulsification, and foaming properties of alternative proteins, as they are key technofunctionalities, and in the second part, we take a closer look at the creation of solid food

structures that are important for the design of sausage, meat, or cheese analogs—a current focus of vegan food design.

5.1. Solubility Characteristics of Protein Fractions

Protein solubility in foods is analyzed as the protein fraction that cannot be separated from an aqueous suspension of a proteinaceous material by centrifugation or filtration under standardized conditions. The solubility characteristics of glutelins and prolamins are not discussed because of their low water solubility at neutral pH (Majzoobi et al. 2012, Thewissen et al. 2011). In general, it must be noted that many plant proteins are already aggregated and that aggregation might be further enhanced by industrial processing. A recent study reported that 37% of proteins in a commercial pea protein isolate were aggregated, which could have been fostered by protein extraction procedures (Chen et al. 2019).

Globulins. Globulins have distinctive properties that influence their solubility. 7S globulins (150–190 kDa) are trimeric and lack disulfide bonds but exhibit glycosylation, whereas 11S globulins (300–370 kDa) are often hexameric and have numerous disulfide bridges (González-Pérez & Arellano 2009). The solubility of globulins, especially 11S globulins, is strongly dependent on pH and ionic strength, and the following dependencies have been reported, in particular for globulins from legumes (Kimura et al. 2008, Lakemond et al. 2000):

- At low ionic strengths ($\leq$0.1 M), 7S globulins have a minimum solubility at pH 4.5–7 and a high solubility (>90%) above and below this region, whereas 11S globulins have a broader minimum solubility at pH 4–7.5.
- At high ionic strengths ($\geq$0.5 M), 7S globulins generally have a high solubility (>90%) at pH 3–9, whereas the solubility of most 11S globulins is low (<80%), up to pH 5–6 except for soy 11S, which has no clear minimum solubility and a high solubility (>80%) above pH 3–4.

The solubility characteristics of these proteins are related to various factors that influence their molecular interactions and the entropy of mixing, and vary from plant to plant: e.g., (*a*) intrinsic molecular weight differences between 7S and 11S globulins, (*b*) presence of hydrophobic as well as basic and acidic subunits on the protein surface that can be screened by counterions, and (*c*) association and dissociation of subunits into smaller or larger units (Gueguen et al. 1988, Kimura et al. 2008, Lakemond et al. 2000, Molina et al. 2004). In general, smaller molecular weights favor solubility (higher entropy), whereas more acidic subunits oppose minimum solubility effects at high ionic strengths. Low ionic strength and low/high pH favor dissociation because of strong electrostatic repulsion between the protein molecules, whereas high ionic strength and neutral pH favor the native state of globulins (Gueguen et al. 1988, Kimura et al. 2008, Withana-Gamage et al. 2015).

Albumins. Compared to globulins, albumins exhibit a higher solubility over a broad pH range because of their smaller molecular weight (10–18 kDa) and higher hydrophilicity. For example, the solubility at different pH values was reported to be higher from neutral to acidic pH for albumins compared to globulins from kidney bean, pea, chia, hemp seeds, buckwheat seeds, locust bean, pumpkin seeds, and sunflower seeds (Djemaoune et al. 2019, González-Pérez et al. 2005b, Julio et al. 2019, Lawal et al. 2005, Malomo & Aluko 2015, Mundi & Aluko 2012, Pham et al. 2017). Albumins of sunflower seeds and kidney beans have a protein solubility of >90% and >70%,

respectively, in pH 3–9 (González-Pérez et al. 2005b, Mundi & Aluko 2012). The solubility of sunflower seed albumins was also high in this pH range at higher ionic strengths ($I = 0.25$ M) (González-Pérez et al. 2005b). Hence, albumins have a unique solubility profile for food applications in the acidic pH range and show a solubility profile from acidic to neutral pH similar to most 7S globulins at higher ionic strengths.

Algae proteins. The solubility of proteins extracted from microalgae and cyanobacteria as a function of pH has been studied for many different species. Most studies show that proteins obtained from microalgae have low solubility at pH < 5 (Grossmann et al. 2019c). In contrast, the limited studies that have been published about the solubility of macroalgae proteins suggest that these proteins have an overall low solubility (<50%) in the pH range 3–7 with higher solubilities in the alkaline pH range (Harrysson et al. 2018, Kandasamy et al. 2012, Suresh Kumar et al. 2014, Vilg & Undeland 2017). Data reported for microalgae proteins showed unique solubility profiles for two microalgae species. First, Grossmann et al. (2019d) reported an overall high solubility of ≥84% in the pH range 2–12 for *Chlorella protothecoides* proteins. Second, the proteins extracted from *Tetraselmis* spp. had a solubility that was independent of the ionic strength up to 0.5 M and the proteins were completely soluble above a pH ≥ 5.5, as reported by Schwenzfeier et al. (2011). The observed effects were mainly attributed to a high amount of hydrophilic amino acids and a high degree of glycosylation with charged polysaccharides (e.g., uronic acids).

Insect proteins. Solubility has been reported for proteins obtained from different species, including *Gryllodes sigillatus*, *Locusta migratoria* L., *Tenebrio molitor*, *Hermetia illucens*, *Acheta domesticus*, *Bombyx mori*, and *Schistocerca gregaria* (Felix et al. 2020, Gravel & Doyen 2020, Kim et al. 2017, Mintah et al. 2019, Ndiritu et al. 2019, Stone et al. 2019). The solubility of the proteins in whole (nonfractionated) meals was low (<30%) at pH 3–9, which might be partly related to intensive downstream processing, such as sterilization and roasting (Hall et al. 2017, Ndiritu et al. 2019, Purschke et al. 2018a, Stone et al. 2019). Solubility was enhanced at higher pH values and higher ionic strengths (Kim et al. 2017). By extraction and fractionation, dissolution behavior was improved, but reported solubility of extracts from *H. illucens* and *B. mori* were still below 60% at pH 3–9, which is lower than many other protein sources (Felix et al. 2020, Mintah et al. 2019). In contrast, proteins extracted from *T. molitor*, *Patanga succincta*, *Chondracris roseapbrunner*, and *L. migratoria* had higher solubilities, especially in the slightly alkaline pH range where solubilities were greater than 90% (Bußler et al. 2016, Gravel & Doyen 2020, Purschke et al. 2018b, Zielińska et al. 2018). Thus, although many insect protein fractions have a low solubility, some specific fractions may have solubility characteristics suitable for use in food applications.

Potato proteins. Patatin has a low (<50%) solubility in pH range 3.5–5.5, but it can be enhanced at low ionic strengths ($I = 0.15$ mM). In contrast, a high (>95%) solubility at acidic pH (pH 3) and neutral to alkaline pH was reported (Ralet & Guéguen 2000, van Koningsveld et al. 2001). Interestingly, the protease inhibitor fraction (16–25 kDa fraction) was reported to have a very high (>90%) solubility across a broad pH range from 2.5 to 12 (Ralet & Guéguen 2000).

Taken together, it is important to note the wide variety in solubility behavior of alternative proteins depending on the environmental conditions and composition of the protein ingredients under investigation. Again, a more systematic approach to recording and reporting these properties for emerging new alternative proteins should be adopted to facilitate their use in the formation of various food matrices.

5.2. Emulsifying and Foaming Properties

Two fundamental technofunctional properties of importance to food design are emulsion and foam formation and stabilization, both of which are discussed below.

Refined plant protein fractions. Soy proteins have been subject to in-depth investigations as plant-based emulsifiers and foaming agents (Day 2013), but emulsifying and foaming properties have also been reported for other plant proteins derived from, for example, pea, rice, lupin, chickpea, lentil, faba bean, sunflower, canola, and pumpkin (Aluko et al. 2009, Benjamin et al. 2014, Can Karaca et al. 2011, González-Pérez et al. 2005a, Ho et al. 2017, Karaca et al. 2011, Pozani et al. 2002, Romero et al. 2012, Tan et al. 2014a). Commercial soy protein isolates can stabilize emulsions away from their p*I*, but soy-stabilized emulsions are prone to flocculation because of charge neutralization and bridging effects. They are, however, quite resistant to coalescence because of their thick interfacial layer (Tang 2017).

In general, the low solubility and presence of aggregates because of heavy processing often prevents the formation of stable emulsions and foams with commercial plant protein concentrates/isolates, and milder processing techniques are generally recommended to enhance their functionality (Lam et al. 2017, Tan et al. 2011). However, the thicker interfacial layers formed by many nonfractionated plant proteins make them promising candidates for concentrated emulsions, which contain tightly packed droplets.

Many authors remove insoluble aggregates prior to emulsion preparation to enhance functionality but also to ensure that the droplet sizes produced are more uniformly distributed (Benjamin et al. 2014, González-Pérez et al. 2005a, Gumus et al. 2017, Ho et al. 2017). In doing so, the formation of stable emulsions for up to 14 days was reported for pea and soy protein with $d_{32} < 0.5$ μm, viscoelastic interfaces, and adsorption characteristics similar to other globular proteins (Ho et al. 2017). Similar observations were made for proteins derived from lupin, lentil, and canola, with lentil proteins showing promise to broaden the available plant-based protein emulsifier portfolio (Benjamin et al. 2014, Chang et al. 2015).

Globulins and Albumins. As most other proteins that are soluble in aqueous solutions, globulins and albumins are surface-active and able to stabilize emulsions and foams to a certain extent. However, no clear correlation between the solubility characteristics and emulsifying properties of albumins and globulins has yet been established. Albumins obtained from flaxseed, hemp, *Ginkgo biloba*, and kidney bean adsorbed rapidly at interfaces because of their higher solubility, lower molecular weight, and higher molecular flexibility, which allows them to quickly diffuse to and rearrange at oil–water or air–water interfaces (Deng et al. 2011, Krause & Schwenke 2001, Malomo & Aluko 2015, Mundi & Aluko 2012, Nwachukwu & Aluko 2018). In contrast, albumins from pea, canola, chickpea, and sunflower have overall low emulsifying properties compared to globulins, which has been related to their low molecular flexibility and/or low hydrophobicity (González-Pérez et al. 2005a, Lu et al. 2000, Papalamprou et al. 2010, Patil & Benjakul 2017, Tan et al. 2014a).

Even though some albumins are effective emulsifiers and capable of producing foams, their ability to stabilize foams and emulsions was reported to be lower compared to globulins for proteins from kidney beans, *Ginkgo biloba* seeds, cashew nut, and hemp seeds, especially at neutral pH (Deng et al. 2011, Lawal et al. 2005, Liu et al. 2018, Malomo & Aluko 2015, Mundi & Aluko 2012). This indicates that globulins are capable of forming stronger interfacial films, probably because of unfolding and interactions between the larger subunits.

Additionally, foaming and emulsifying properties may differ depending on the nature of the used globulin fraction. For example, 7S soybean globulins (β-conglycinin) are more effective emulsifiers compared to 11S globulins (glycinin) from soybeans (Keerati-u-rai & Corredig 2010). In contrast, 11S soybean globulins are more effective in foam stabilization, which was attributed to increased interaction of the larger subunits at the interface (Ruíz-Henestrosa et al. 2007). The same behavior has been reported for pea globulins (Chen et al. 2019, Liang & Tang 2013, Xiong et al. 2018). Nevertheless, this is not universally true and some 11S globulins show higher emulsifying activity than 7S globulins, e.g., those in lima bean and faba bean (Chel-Guerrero et al. 2011, Kimura et al. 2008).

Prolamins and Glutelins. The low solubility of prolamins and glutelins is a major obstacle in preparing emulsions and foams, especially for high-molecular-weight fractions. The low solubility has been exploited to prepare nanoparticles from prolamins using antisolvent techniques, which can stabilize emulsions using the Pickering effect (Folter et al. 2012, Zhu et al. 2018). These particles may also lend themselves to prepare and stabilize foams. For example, native prolamin and its fractions extracted from wheat (i.e., gliadins) proved to be efficient foaming agents especially in the alkaline pH region (Thewissen et al. 2011).

Algae proteins. The emulsifying and foaming properties of proteins obtained from microalgae have recently been reviewed in different species by Grossmann et al. (2019c), but detailed information about the properties of macroalgae proteins is still missing. Interestingly, microalgae proteins obtained from *Chlorella sorokiniana* (stable at pH $\geq$ 5 and up to 500 mM NaCl), *C. prototthecoides* (stable in pH range 3–9 and up to 500 mM NaCl), and *Tetraselmis* spp. (stable at pH $\geq$ 5) exhibit particularly promising emulsifying properties for foods (Ebert et al. 2019, Grossmann et al. 2019a, Schwenzfeier et al. 2013).

Insect proteins. Insect proteins may also be used to stabilize interfaces (Gravel & Doyen 2020). Most studies report overall low foam capacities and foam stabilities for insect proteins (foam capacity <50%) with exceptions for *G. sigillatus* (foam capacity = 99%) and *L. migratoria* (foam capacity = 459%) (Purschke et al. 2018a, Zielińska et al. 2018). In contrast, proteins from insects seem to be more suitable to stabilize oil–water interfaces, as indicated by emulsifying capacities that are in the same range as other proteins, such as whey and soy proteins (Gravel & Doyen 2020). A study by Gould & Wolf (2018) found that proteins extracted from *Tenebrio molitor* at pH 7 had a lower interfacial tension and yielded smaller droplet sizes under fixed homogenization conditions (high-shear blending, 8,000 rpm for 2 min) compared to whey proteins. Moreover, the prepared emulsions were stable against coalescence and the addition of NaCl up to 330 mM for 2 months. This suggests that proteins from insects have the potential to act as functional ingredients if extraction is carried out appropriately.

Potato proteins. Although nonfractionated potato protein displays interfacial properties, improvements can be achieved through fractionation (Schmidt et al. 2018). Depending on the fractionation process used, patatin has a lower or higher emulsifying activity compared to the 16–25 kDa fraction (Ralet & Guéguen 2000, Schmidt et al. 2018). However, the 16–25 kDa fraction is less prone to creaming compared to casein and yields stable emulsions with slow coalescence rates in the pH range 4–8 and up to 1% NaCl (Ralet & Guéguen 2000). In contrast, patatin has an overall higher foaming capacity and foam stability compared to egg white, and the 16–25 kDa fraction in the pH range 4–8 and up to 1% NaCl (Ralet & Guéguen 2001).

5.3. Principles of Solid Structure Formation

In many solid foods, proteins either exist as a cross-linked network or are in a glassy state. These structures display isotropic (direction independent, e.g., homogeneous gels such as yogurt) or anisotropic (direction-dependent, e.g., fibrous or layered structures such as meat) structures, which are reviewed in this section.

Isotropic structures: gels. Isotropic structures can be formed by albumins, globulins, glutelins, and prolamins. Glutelins and prolamins self-assemble into a gel network upon hydration, whereas physical and chemical structuring is necessary for albumins and globulins. Heat gelation is induced by heating the proteins close to or above their denaturation temperature to achieve partial or complete unfolding, which enables proteins to form a three-dimensional network (Nishinari et al. 2014). Heat gelation has been studied for many refined alternative protein fractions but also for many other fractions (**Table 1**).

The gelation properties during heating are related to their molecular weight, reactive amino acid side chains, and specific denaturation temperature, which influence the subtle mix of intra- and intermolecular bonds. For example, patatin from potato has a lower denaturation and gelation temperature than β-lactoglobulin and ovalbumin, which is related to its high hydrophobicity and low amount of disulfide bonds, favoring the complete unfolding and exposure of hydrophobic groups (Creusot et al. 2011, Delahaije et al. 2015). Moreover, proteins isolated from lupin form weaker gels after heating compared to soy protein because of a higher amount of free sulfhydryl groups, which facilitate the formation of intramolecular bonds that in turn prevent the formation of a larger intermolecular three-dimensional gel network (Berghout et al. 2015, Sousa et al. 1995). Gel strengths thus decline in the order of soy > pea > lupin (Batista et al. 2005). Many plant proteins do not denature below temperatures of 70°C and often temperatures above 85°C are required (which is important when creating plant-based meat or egg analogs). It should be noted that the denaturation temperature is impacted by the presence of salts introduced during processing or extraction (Cai et al. 2002, Lakemond et al. 2000, Nieto-Nieto et al. 2014, Sirison et al. 2017).

Cold gelation involves the formation of a gel at or below room temperature through a pH shift (microbial fermentation or addition of acidifiers, such as glucono delta-lactone), the addition of multivalent cations (such as calcium), enzymatic cross-linking [microbial transglutaminase (mTG)], or static pressure, or by employing a combination of these techniques (**Table 2**). Cold gelation of proteins from whole soybeans by fermentation (e.g., yogurt-like products, tempeh) or addition of multivalent cations after heat denaturation (e.g., tofu) has a long history in traditional and modern food production and is also known for other whole pulses.

To date, many gelled foods made from alternative proteins contain further network-enhancing compounds such as hydrocolloids, as their stability and mechanical strength are otherwise still lacking. Gels produced from extracted proteins prepared by cold gelation induced by multivalent cations or a decrease in pH have been reported for many different plant proteins (**Table 2**). In general, cold gelation caused by a change in pH or the addition of multivalent cations is a two-step process. The proteins are partly denatured by heat or high pressure to expose functional groups at a concentration, pH, and ionic strength that minimizes protein–protein interaction (i.e., low concentrations, pH far below or above p*I*, low ionic strength). This is then followed by a change in pH or the addition of multivalent cations (often Ca^{2+} salts) to cause gelation. Alternatively, cross-linking without a heat pretreatment can be accomplished by the addition of mTG or by high pressure (**Table 2**).

Table 1 Overview of heat-set gelation properties of alternative proteins and protein fractions

Protein fraction	Findings	Reference
Refined plant protein fractions		
Soy (isolate)	c_{gel} = 9–11% at 95°C for 15 min	Hua et al. 2005
	T_{gel} = 84°C at pH 7	Renkema & van Vliet 2002
Lupin (isolate)	c_{gel} = 18% after heating at 95°C for 30 min at pH 7	Berghout et al. 2015
Pea (isolate)	c_{gel} = 16% after heating at 95°C for 30 min at pH 7.6	O'Kane et al. 2005
	c_{gel} = 14.5% after heating at 95°C for 10 min at 0.3 M NaCl	Sun & Arntfield 2010
	$c_{protein}$ = 19.6%, pH 7.1, 2.0% NaCl, and 93°C (optimal process conditions)	Shand et al. 2007
Lentil (isolate)	c_{gel} = 8–14% after different treatments	Jarpa-Parra 2018
Chickpea (isolate)	c_{gel} = 11.5%	Papalamprou et al. 2009
Canola (isolate)	T_{gel} = 79.5°C at 7.5% at pH 9	Schwenke et al. 1998
Sunflower (concentrate)	c_{gel} = 10% after heating at 100°C for 20 min at pH 8	Salgado et al. 2012
Oat (isolate)	c_{gel} = 15% at 110°C for 15 min at pH 7	Nieto-Nieto et al. 2014
Albumins		
Napin (canola)	T_{gel} = 95°C (pH 7; 1°C/min; c_{gel} = 12.5%) G′ = 862 Pa after heating to 98°C at 1°C/min and pH 7	Schwenke et al. 1998 Tan et al. 2014b
Sunflower	Heat stable above 100°C	Pérez et al. 2005b
Great northern bean	c_{gel} = 18% after 1 h at 100°C	Sathe & Salunkhe 1981
Chickpea	T_{gel} = 65°C (3°C/min, pH 7)	Papalamprou et al. 2009
African locust bean	c_{gel} = 12% at pH 7 after 1 h at 100°C	Lawal et al. 2005
Globulins		
β-Conglycinin (soy)	T_{gel} = 73°C (9%, pH 7.6) forms weaker gel than glycinin	Renkema et al. 2001
Glycinin (soy)	T_{gel} = 90°C (9%, pH 7.6) forms firmer gel than β-conglycinin	Renkema et al. 2001
Legumin (pea)	T_{gel} = 88°C (pH 7.6, 0.5°C/min); c_{gel} = 8.4% at 100°C for 30 min at pH 7.6; 1.8% higher minimum gelling concentration compared to soy glycinin	O'Kane et al. 2004b
Vicilin (pea)	c_{gel} = 10–14% at 100°C and pH 7.6 for 30 min	O'Kane et al. 2004a
Red bean	T_{gel} = 80°C (pH 7.6, 3.5°C/min; c_{gel} = 10%)	Meng & Ma 2002
Cruciferin (canola)	T_{gel} = 70–72°C (pH 6–9, 1°C/min; c_{gel} = 12.5%) G′ = 1,435 Pa after heating to 98°C at 1°C/min and pH 7 T_{gel} = 80.9°C (pH 7.4, 1°C/min; c_{gel} = 5%)	Schwenke et al. 1998 Tan et al. 2014b Withana-Gamage et al. 2015
Pumpkin	c_{gel} = 14–18% at 100°C and pH 2–10 for 60 min	Pham et al. 2017
Oat	c_{gel} = 10% at 100°C for 20 min at pH 9.7 and 0.2 M NaCl	Ma et al. 1988
Prolamin		
Gliadin (wheat)	13% dispersion forms gel at 50°C and pH 9.3	Sun et al. 2009
Insects		
Tenebrio molitor	T_{gel} = 61.7 ± 1.1°C at 15% and pH 7	Yi et al. 2013
Alphitobius diaperinus	T_{gel} = 58.2 ± 2.1°C at 15% and pH 7	
Zophobas morio	T_{gel} = 51.2 ± 1.5°C at 15% and pH 7	
Acheta domesticus	T_{gel} = 56.2 ± 0.7°C at 15% and pH 7	
Blaptica dubia	T_{gel} = 63.2 ± 0°C at 15% and pH 7	

(*Continued*)

Table 1 (*Continued*)

Protein fraction	Findings	Reference
Microalgae		
Arthrospira platensis	$c_{gel} \approx 1.5\%$ at 90°C and pH 7	Chronakis 2001
Chlorella sorokiniana	$T_{gel} = 61$°C; $c_{gel} = 9.9$ g/L at pH 5.6	Grossmann et al. 2019b
Tubers		
Patatin (potato)	$T_{gel} = 30$°C–53.9°C (pH 3–6.6 at $I = 15$–200 mM; $c_{gel} = 8\%$)	Schmidt et al. 2019

No literature data have been identified for glutelin and proteins extracted from fungi and macroalgae.
Abbreviations: c_{gel}, concentration that induces gelation; T_{gel}, temperature to induce gelation, G′ = shear storage modulus.

Anisotropic structures: fibers. One major challenge in creating foods with alternative proteins is the formation of fibrous structures that resemble that of muscle tissue with its highly ordered elongated structures. The techniques that have been developed to produce fibrous structures from globular proteins are shown in **Figure 2**. For information about cell-culturing techniques, including mycoproteins, and 3D printing technologies the reader is referred to Dekkers et al. (2018) and Dick et al. (2019).

Extrusion. Co-rotating twin-screw extruders with a length-to-diameter ratio >20 are the most common equipment used to produce fibrous meat analogs. The proteins are aligned in the extruder and further structured in the die (Samard et al. 2019). An extruder typically consists of four main sections: a feeding zone (conveying elements; $T = 25$°C), mixing zone (kneading and paddle blocks; $T < 100$°C), melting zone (conveying and kneading/reverse elements to increase mechanical energy input elements; $T > 100$°C), and a die (Osen et al. 2014). These sections can be further divided, often resulting in more than seven independent compartments (Pietsch et al. 2017).

Depending on the moisture content and the die configuration, low-moisture texturized vegetable protein (LM-TVP) and high-moisture texturized vegetable protein (HM-TVP) are obtained. LM-TVP is mainly used as a meat extender, whereas HM-TVP serves as a meat analog. LM-TVP is obtained at low moisture contents (<50%) and a short shaping die. Because of the sudden pressure drop at the die exit, the hot melt expands through flash evaporation (**Figure 2**), resulting in cooling and reduction of water (<−20%) followed by a drying step. Second, HM-TVP is produced at higher moisture contents (50–70%). The melt is pressed into a long slit cooling die, which has a temperature of 20–80°C (Osen et al. 2014, Pietsch et al. 2017). In the die, the melt temperature and flow rate decrease without expansion. The high moisture content prevents extensive agglomeration in the extruder, allowing new intra- and intermolecular bonds to be formed during cooling. Depending on the raw material, a mixture of hydrophobic interactions and hydrogen and disulfide bonds are responsible for setting the structure (Chen et al. 2011, Osen et al. 2015). Here, at low temperatures and screw speeds, protein unfolding is incomplete, whereas too-high temperatures and screw speeds lead to insufficient cooling (lower viscosity) and evaporation, preventing the formation of anisotropic structures (Pietsch et al. 2019). For example, anisotropic structures were obtained with soy protein concentrate at $T > 120$°C and specific mechanical energy inputs >170 kJ/kg, and anisotropy was observed in an extruded pea isolate at a temperature of 140°C and screw speeds of 150 rpm (Osen et al. 2015, Pietsch et al. 2019).

Two effects are currently discussed that describe anisotropic fibrous structure formation in the die: (*a*) the laminar flow results in a temperature and velocity gradient in the melt starting from the cool die wall, which eventually fixes the laminar protein dispersion stream lines in a layered, fibrous structure in the direction of the flow (**Figure 2**) (Osen et al. 2014), and (*b*) a

Table 2 Overview of cold-set gelation properties of alternative proteins and protein fractions

Protein fraction	Findings	Mechanism	Reference
Refined plant protein fractions			
Soy (isolate)	c_{gel} = 6–9% at pH 7 after heating at 105°C for 30 min with 10–20 mM $CaCl_2$	Multivalent cations	Maltais et al. 2005
	c_{gel} = 6% with mTG at 10 units/g of protein	Enzymatic	Tang et al. 2006
	c_{gel} = 9% after high-pressure treatment at >400 MPa and >15 mM $CaCl_2$ addition	Static pressure and multivalent cations	Speroni & Añón 2013
Pea (isolate/ concentrate)	Gel point at pH 6.6 of predenatured proteins at 85°C for 60 min at 2%	pH	Mession et al. 2015
	Onset of gelation at pH 6.35 of predenatured proteins at 60°C for 60 min at 10%	pH	Klost & Drusch 2019
	c_{gel} = 18% treated with mTG resulted in gel with strength > 2 N/cm	Enzymatic	Schäfer et al. 2007
	c_{gel} = 16% after 250 MPa treatment for 15 min	Static pressure	Sim et al. 2019
Rapeseed (concentrate)	Optimum protein concentration 10% for mTG based gel	Enzymatic	Hyun & Kang 1999
Oat (isolate)	c_{gel} = 5% protein at pH 5.2 after heating at 115°C for 15 min	pH	Yang et al. 2017
Albumins			
Pea albumin	No gelling observed after 5-h treatment with mTG	Enzymatic	Djoullah et al. 2015
Globulins			
β-Conglycinin (soy)	c_{gel} = 9% forms self-supporting gel after high-pressure treatment at 400 and 600 MPa	Static pressure	Speroni & Añón 2013
Glycinin (soy)	c_{gel} = 6% exhibited a G′ of 1,120 Pa and a hardness of 15.1 g after heating at 95°C for 30 min and pH 4.92	pH	Zhu et al. 2011
	c_{gel} = 2% to form self-supporting gel with transglutaminase	Enzymatic	Chanyongvorakul et al. 1994
	c = 9% did not form self-supporting gel after static high-pressure treatment and Ca^{2+} addition	Static pressure	Speroni & Añón 2013
Legumin (pea)	c_{gel} = 3.5% at pH 6.6 reached after 7.7 min with predenatured proteins (85°C for 60 min)	pH	Mession et al. 2015
Vicilin (pea)	c_{gel} = 2% at pH 5.9 reached after 19.5 min with predenatured proteins (85°C for 60 min)	pH	Mession et al. 2015
Pea globulin, whole	c_{gel} = 6% with mTG at pH 7 after 5 h	Enzymatic	Djoullah et al. 2015
Legumin (broad bean)	c_{gel} = 3% to form self-supporting gel with transglutaminase	Enzymatic	Chanyongvorakul et al. 1994
Oat	c_{gel} = 10% with 0.2-M NaCl forms gel after mTG treatment at pH 7.5	Enzymatic	Siu et al. 2002

No literature data have been identified for prolamins, glutelins (which form hydrated networks by self-assembly), or proteins extracted from insects, microalgae, macroalgae, or tubers.

Abbreviation: mTG, microbial transglutaminase; c_{gel}, = concentration that induces gelation; G' = shear storage modulus.

phase-separation mechanism induced by temperature changes causes the formation of anisotropic fibers. Calculations by Sandoval Murillo et al. (2019) showed that shear strains obtained during laminar flow in the die are too low to obtain layered structures, and the authors proposed instead a spinodal phase-separation mechanism under the influence of the temperature gradient, which

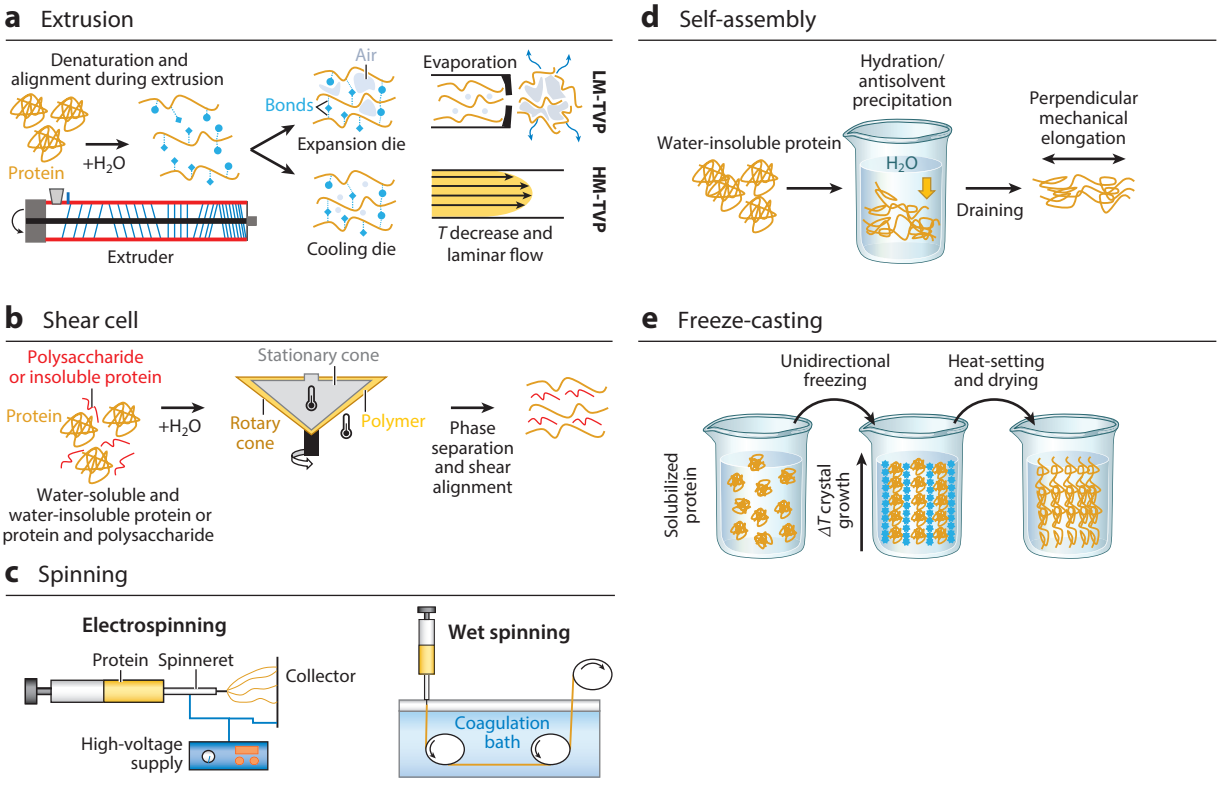

Figure 2

Schematic overview of techniques used to produce layered and fibrous structures for meat analogs including extrusion, shear-cell, spinning, self-assembly, and freeze-casting technology. Abbreviations: HM-TVP, high-moisture texturized vegetable protein; LM-TVP, low-moisture texturized vegetable protein.

yields protein-rich and water-rich segments, a process that might be favored by the presence of polysaccharides as well.

Shear cell. The shear cell technology was first published by Manski et al. (2007) and employs a cone-in-cone or Couette-type design with a rotating and stationary plate (**Figure 2**) to produce fibrous structures. The advantages of this technique are milder conditions with a much lower specific mechanical energy input and the ability to control the flow parameters (Grabowska et al. 2016). During shear cell processing, a mixture of thermodynamically incompatible proteins (e.g., soy protein isolate and gluten) or a protein and polysaccharide (e.g., soy protein isolate and citrus pectin) are sheared at water contents of >50% and temperatures >100°C and subsequently cooled down (Dekkers et al. 2016, Schreuders et al. 2019). The phase-separated polymers form fibrous structures during shearing, whereas the dispersed phase gets elongated into weak fibers in the direction of shear and finally solidifies embedded in the continuous phase (Dekkers et al. 2018).

Self-assembly. Water-insoluble protein fractions, i.e., prolamins and gliadins, self-assemble because of their hydrophobic nature into a dough-like structure upon rehydration or antisolvent precipitation, which can be mechanically stretched and kneaded into anisotropic and fibrous-like structures. This effect is well-known in the production of seitan from wheat gluten. However, it

has been reported that also other prolamins, such as zein, can form fibrous structures after rehydration at 60°C and subsequent stretching at elevated temperatures or using antisolvent precipitation (Mattice & Marangoni 2020). Additionally, the production of fibrous, rod-like structures has been described for specific protein fractions, such as β-lactoglobulin, when heating the protein at low pH (Jung et al. 2008, Veerman et al. 2002).

Spinning. Two spinning methods have been described to obtain fibrous protein structures: wet-spinning and electrospinning. Wet-spinning for fibrous meat analogs was first introduced by Boyer (1954). A concentrated protein solution is pumped through a spinneret or extrusion die to form fibrous structures into a coagulation bath consisting of salt, acid, or alkali to denature the proteins. The filaments are collected and stretched on rolls and subsequently washed and treated further with, for example, binders to assemble a meat-like macrostructure. In contrast, electrospinning employs a high electrical field to produce fibers. The protein solution is pumped through a spinneret that is connected to a high-voltage generator, forming a droplet at the tip. If the voltage exceeds a critical value, a charged fluid stream is ejected with solvent rapidly evaporating forming an endless fiber that can be collected at a grounded receptor plate (Ghorani & Tucker 2015). This method has been successfully implemented for many proteins, including soy, pea, and zein, and also for needleless electrospinning devices with higher throughput, but large-scale production of protein fibers has not yet been accomplished (Dekkers et al. 2018, Kutzli et al. 2020).

Freeze-casting. Freeze-casting of proteins to obtain fibrous meat-like structures was developed in the 1970s and has been mainly described for soy proteins (Kim & Lugay 1978). The proteins are dispersed and frozen using a unidirectional removal of heat (Lawrence & Jelen 1982). The proteins are freeze concentrated and pushed by the directional growth of ice crystals into an anisotropic structure. The water is subsequently removed by, for example, freeze-drying and then the fibers are heat set (Kim & Lugay 1978).

6. CONSUMER ASPECTS AND SUSTAINABILITY

Foods made from alternative proteins are gaining market share, and high-performing initial public offerings of start-ups in this space have confirmed opportunities for investors in this market (Bashi et al. 2019). Plant-based food sales, for example, increased by 17% in 2018 with the current market having a size of approximately $2.2 billion (Good Food Inst. 2018). However, compared to the size of the meat market, which is estimated at $1.7 trillion, this is still quite low (FAO 2019). This will likely continue to change given that interest in alternative proteins is increasing.

It is interesting to note that the development of foods based on alternative proteins is not necessarily driven by the desire of the consumer to move toward a more sustainable food supply (Hartmann & Siegrist 2017). In fact, the low awareness of the environmental impact of meat production and the issues surrounding the sustainability of the anticipated growth in production was confirmed by a recent review of the literature, where only between 18% and 34% of consumers in the United States, Germany, the Netherlands, Portugal, and Australia agreed to statements on the negative impact of animal protein consumption. Apparently, few consumers are aware of the impact that their food choices have on the environment and climate change. Meat lovers, for example, have been reported to generate more than twofold carbon dioxide equivalents per day (7.19 $kgCO_2e$/day) of emissions compared to consumers who adhered to a vegan diet (2.89 $kgCO_2e$/day) (Scarborough et al. 2014). Moreover, it has been estimated that between 8% and 51% of all man-made greenhouse gas emissions are due to livestock farming, albeit there is a considerable debate about reported numbers (Herrero et al. 2011).

The current lack of information is one of the factors contributing to a low willingness in consumers to change food patterns. For example, only between 19% (Netherlands) and 38% (Belgium) of survey participants reported a willingness to reduce their meat consumption because of environmental concerns. When informed about the impact on the environment, willingness to change dietary patterns increased. Other factors such as concern for animal welfare or the desire to reduce the intake of animal proteins due to health concerns and incidences of noncommunicable diseases associated with current dietary practices, especially in Western societies, seem to have—at least at present—a larger impact than environmental concerns (Gómez-Luciano et al. 2019). Familiarity and repeated exposure have also been found to be of importance, as consumers get used to the flavors and textures of animal alternatives. A recent study reported that supermarkets can also play a role in facilitating this transition, as they can be enablers or limiters by increasing or limiting availability and/or marketing of products based on alternative protein sources (Gravely & Fraser 2018).

A brief review of pertinent literature shows that research on consumers' motivation to decrease meat consumption and increase consumption of foods made from alternative proteins is gaining traction, likely because foods with broadly acceptable sensory performances are now reaching the market. Population studies are generally still rare and most studies to date have been carried out in Western countries rather than in developing countries where the major part of the projected growth of meat consumption is projected to occur.

7. CONCLUSION

Progress has been made in discovering new sources for proteins and extracting them to obtain new technofunctional alternative protein ingredients that facilitate the formation of various food structures, especially fibrous ones. There are continuously new molecular and processing approaches emerging that form the basis of new food design using these alternative proteins. It is likely that with an enhanced understanding of the fundamental properties of alternative proteins in combination with improved extraction cascades, new food matrices can be developed that not only mimic existing foods but provide consumers with new experiences such as novel textures, flavors, and appearances. However, there are still discussions and uncertainties about the overall sustainability improvements of current value-chain approaches. Constant improvements in cultivation and harvesting techniques coupled with efficiency gains in processing and simplification in formulation approaches will undoubtedly lead to food systems that in the long run allow foods to be produced with a lower environmental footprint. This is to some degree facilitated by changes in consumer choices leading to increases in production volumes. There are, however, still considerable gaps in the knowledge base with respect to process–property–functionality relationships, and, consequently, product development is often done using time-consuming trial-and-error approaches. Moreover, more research is needed to find out how consumers' choices in this food category can be influenced, i.e., how consumers could be nudged toward making more sustainable food choices. This would in turn lead to increased market shares. The subsequent scale-up efforts combined with manufacturers' desire to minimize costs and maximize profits would lead to efficiency gains, which may have positive impacts on environmental footprints.

DISCLOSURE STATEMENT

The authors are not aware of any affiliations, memberships, funding, or financial holdings that might be perceived as affecting the objectivity of this review. The authors have received industrial and governmental research funding in the area of alternative proteins, which did not influence the writing of this article.

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