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Review

Proteins from Agri-Food Industrial Biowastes or Co-Products and Their Applications as Green Materials

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Abstract: A great amount of biowastes, comprising byproducts and biomass wastes, is originated yearly from the agri-food industry. These biowastes are commonly rich in proteins and polysaccharides and are mainly discarded or used for animal feeding. As regulations aim to shift from a fossil-based to a bio-based circular economy model, biowastes are also being employed for producing bio-based materials. This may involve their use in high-value applications and therefore a remarkable revalorization of those resources. The present review summarizes the main sources of protein from biowastes and co-products of the agri-food industry (i.e., wheat gluten, potato, zein, soy, rapeseed, sunflower, protein, casein, whey, blood, gelatin, collagen, keratin, and algae protein concentrates), assessing the bioplastic application (i.e., food packaging and coating, controlled release of active agents, absorbent and superabsorbent materials, agriculture, and scaffolds) for which they have been more extensively produced. The most common wet and dry processes to produce protein-based materials are also described (i.e., compression molding, injection molding, extrusion, 3D-printing, casting, and electrospinning), as well as the main characterization techniques (i.e., mechanical and rheological properties, tensile strength tests, rheological tests, thermal characterization, and optical properties). In this sense, the strategy of producing materials from biowastes to be used in agricultural applications, which converge with the zero-waste approach, seems to be remarkably attractive from a sustainability prospect (including environmental, economic, and social angles). This approach allows envisioning a reduction of some of the impacts along the product life cycle, contributing to tackling the transition toward a circular economy.

Keywords: bioplastic; protein; biowaste; valorization

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1. Introduction

The accumulation of plastic wastes is a globally recognized problem that involves an extremely negative impact on the environment [1]. The exceptionally low biodegradability of fossil-based plastics, together with the massive production scale associated with the plastic market over the past 60 years, has generated a huge accumulation of plastics in landfills and the oceans [2]. To illustrate the magnitude of the problem, considering that almost 400 Mt of plastic waste is generated every year [3], there is currently more than 1 ton of plastic/person alive in the world. However, in spite of the recent efforts made in this field to shift from a fossil-based to a bio-based circular economy model, only 20% of plastic is collected for recycling, of which only 3% is reused [4,5]. The rest is incinerated, landfilled, or disposed of into nature, an large part of which is ending up in the oceans [6]. In this sense, European Union Directive (EU) 2019/904 aims to prevent and reduce the impact of certain single-use plastic products on the environment, especially the marine environment, and on human health. Consequently, the future of the plastics industry needs to be driven by sustainability issues, where the bioplastic sector is a crucial building block for a circular economy scenario [7,8].

The most accepted definition of the term bioplastic, which has been controversial among plastic industrial associations and environmental organization, is given by Euro-

pean Bioplastics [5]. According to this association, any plastic material can be denoted as a bioplastic if it is either bio-based, biodegradable, or displays both properties. Consequently, bioplastics embrace a whole family of materials with different properties and applications, ranging from biodegradable fossil-based polymers, such as poly(butylene adipate-co-terephthalate) (PBAT) or polycaprolactone (PCL), to non-biodegradable bio-based polymers, such as bio-based polyolefins (e.g., bioPE and bioPP) or polyesters (e.g., bioPET) [9]. However, the ecofriendliest bioplastic group is formed by biodegradable and bio-based polymers. This group comprises biodegradable aliphatic polyesters produced by fermentation of biomass, including polylactates (PLA), polyglycolates (PGA), polyhydroxy butyrate (PHB), polyhydroxy valerate (PHV), etc., and polymers extracted from renewable sources, also known as agropolymers, which include polysaccharide-based polymers (e.g., starch, cellulose, and cellulose derivatives) and protein-based polymers that can be extracted from animal or plant sources [10,11]. Currently, a big amount of the food produced worldwide (~30%) is discarded by the agri-food industry, being considered as byproducts or wastes [12]. These food biowastes could be reused as raw materials for the emerging bioplastics sector since their proteins, carbohydrates, lipids, and other compounds can be used for this application [13]

Agropolymers are considered the most ecoefficient bioplastic source in terms of the ratio between the added value of their potential applications and the environmental impacts associated with them [14]. They consist of a carbon backbone with different side groups that can form inter-/intra-molecular H-bonds. It is precisely the ability to temporarily disrupt these H-bonds and cause flow into new material shapes that allows forming plastic materials by conventional polymer processing techniques (e.g., casting, thermoforming, compression molding, extrusion, and injection molding) [15]. However, despite the unquestionable importance of bioplastics for enabling a more sustainable circular economy [16], they only cover approximately 1% of the global plastic market, accounting for 2.11 Mt in 2020 [5]. About 60% of the bioplastic market corresponds to biodegradable polymers and 20% to agropolymers (over 420 kt). Among them, starch and cellulose are abundant and low-priced raw materials [6]. Unfortunately, they typically require complex processing before they can be properly used as bioplastics. These processes, including fermentation or functionalization, typically increase their costs and, as a result, reduce their efficiency in the replacement of conventional plastics.

In contrast, an emerging ecofriendly and cost-efficient alternative to plastics is based on the use of protein which can be easily processed for many applications [17,18]. Moreover, protein may be inexpensively extracted from many sources that are also abundant in nature. Interestingly, global food biowastes represent about 1300 Mt/year, according to the Food and Agriculture Organization (FAO) of the United Nations [19]. This biomass may be regarded as a potential source that can be used in the protein-based bioplastic sector, competing with other uses (e.g., biofuel). However, some problems related to the collection of food biowastes, due to their extremely wide dispersion, still impose a barrier to their efficient application at large scales [20]. Other more interesting alternatives are currently being considered for the valorization of proteins, such as the use of agri-food co-products from the starch, oil, or biofuel industries; the extraction from industrial biowastes such as blood, bones, feathers, wool, hair, nails, etc., from poultry or cattle slaughterhouses; or microalgae from sewage plants [20,21]. However, the commercial use of protein-based bioplastics in 2020 is still residual (with an output lower than 30 kt) as compared to other agropolymers, particularly starch which accounts for almost 400 kt in 2020 [5]. Some authors have indicated that plastics production from proteins is economically feasible, reducing the wastes associated with industrial products [22].

As for the portfolio of bioplastic applications, food packaging remains the widest segment of the whole bioplastic family, with an output of almost 1 Mt in 2020, representing nearly half of the total bioplastics market. The other half is largely diversified finding applications as consumer goods, or in the textile, agriculture, automotive, and construction field, among others [5]. In particular, protein-based bioplastics may also accomplish some

of those applications, mainly in the fields of food packaging and plastics for agriculture. Moreover, they may also be used in more specific applications, such as in the development of absorbent and superabsorbent materials, in the controlled release of active agents (e.g., drugs, antimicrobial agents, nutrients, etc.), or biomedical applications (e.g., as scaffolds for tissue engineering) [23]. Therefore, despite the many advantages associated with the use of protein as bioplastics for a wide variety of applications, its high potential for the replacement of conventional plastics has not yet been sufficiently explored [20].

This review is focused on the potentials of protein co-products of the agri-food industry or protein fractions extracted from agri-food industrial biowastes, as well as their applications as substitutes for conventional non-biodegradable fossil-based plastics. The characteristic of these protein-based materials must be analyzed to assess the functionality required for each application. These properties can be typically divided into mechanical properties, thermal properties, and optical properties, correlating them to the microscopic (even molecular) structure of the materials [24].

2. Proteins from Industrial Biowastes and Co-Products

Every year, around one third of all food produced worldwide is either lost or wasted [25]. In Europe, that amount is reduced to one fifth, being 19% obtained from food processing and 11% from primary production [12]. Food biowaste is mainly composed of carbohydrates, proteins, lipids, and other compounds with great potential for high-value applications [13]. In this section, the main protein-rich biowastes and co-products from the agri-food industry that have been used in the development of plastic materials are presented. It should be highlighted that depending on the application pursued, proteins should be previously extracted and/or concentrated from the biowaste. Extraction can be carried out either through dry (e.g., air classification) or nondry (e.g., chemical treatment) conditions. Among the concentration procedures for obtaining protein concentrates or isolates are isoelectric precipitation or ultrafiltration. These preparation techniques are outside of the scope of this study, and readers interested in their description are referred to a recent review on this topic [26].

2.1. Co-Products from Starch

Wheat gluten (known as vital gluten, containing 75–80% protein) and corn gluten (typically containing 55–70% protein) are produced industrially as a co-product either from starch or bioethanol industrial plants [27–29]. The most abundant amino acid residues present in wheat gluten are glutamate and glutamine (31.9%) and proline (14.1%) [30]. Corn gluten is abundant in methionine and cysteine but is very low in lysine and tryptophan [31]. Wheat gluten is mainly used in bakery products, while corn gluten is mainly used as animal feed. Nonfood applications for gluten have been pursued (e.g., thermoplastic materials) [32,33]. In this sense, most wheat gluten-based plastics have been processed through casting or extrusion [33–36], leading to insoluble films, plastics, and adhesives with good barrier properties for oxygen and carbon dioxide [37,38]. Corn gluten has not been studied as profusely as wheat gluten in the field of bio-based materials. However, some studies have reported its potential to form glassy dense material of high thermoplasticity [29,39].

Potato proteins (i.e., patatin, protease inhibitors, and different high-molecular-weight proteins) can be obtained from potato-based starch production, as well as from peels and damaged potatoes. Although the protein content of fresh potatoes is low (2%), a protein isolate (90%) can be obtained from the wastewater generated during their processing through alkaline precipitation [40,41]. Potato protein is rich in hydrophobic amino acid residues with branched (isoleucine (3.1%), leucine (6.7%), and valine (3.7%)) and aromatic (phenylalanine (4.2%) and tyrosine (3.8%)) side chains [42,43]. They also possess a lysine content (~6%) higher than the average found in most plant-based proteins [43]. Potato proteins have been used as food additives or for bioplastic production. Thus, films or sheets from thermoforming or compression molding have been produced from potato

protein flours mold, leading to bioplastics with adequate mechanical properties, sometimes reinforced with some animal proteins, such as gelatin [44,45]. Films obtained by casting have also proven to show significant barrier properties, highlighting its potential in the packaging industry [46].

2.2. Co-Products from Bioethanol

In addition to the abovementioned wheat gluten, zein can be also obtained as a co-product of the bioethanol industry. Zein is the alcohol-soluble protein of corn, and it is a prolamin predominantly present in the endosperm [47]. It possesses a negligible content in essential amino acids, such as lysine and tryptophan, which, together with its poor solubility in water, limit its use for human consumption [47]. Zein may be obtained as a byproduct from the production of ethanol, starch, or oil from corn, containing a high amount of glutamic acid (21–26%) and hydrophobic amino acids, like proline (10.5%) or leucine (21.1%) [47,48]. It has been mainly employed as a coating agent due to its ability to form films with water vapor barrier properties [49,50]. Zein-based films have also been produced through extruders provided with slit dies, where additives like oleic acid can be used to enhance elongation [51–53]. Furthermore, zein may be used as plasticizer in injection molded starch-based plastics [54].

2.3. Co-Products from Seed Oil

Soy oil is extracted from soybean (38–45% protein content) producing a protein-rich meal as a byproduct, which is, for the most part, discarded as industrial biowaste or used for feeding animals [55–57]. Soy proteins, mostly globulins, are rich in polar amino acid residues, such as glutamic acid (12.4%), also containing a considerable amount of lysine (3.4%) [43]. Soy protein-based bioplastics have been processed through several techniques, like casting, compression, or injection molding, resulting in materials with adequate mechanical properties but low water resistance [58–61]. It has been acylated successfully to further enhance its hydrophilic character, which may be well used in superabsorbent or horticulture applications [62,63].

The processing of rapeseed to obtain oil results in the production of a press cake with a high protein content (35%) [64], which cannot be used as a food ingredient due to the presence of antinutritional compounds (e.g., glucosinolates) [65]. Rapeseed and canola have been sometimes used interchangeably, although canola should be strictly employed for cultivars that have been genetically improved and contain a lower content of antinutritional compounds [66]. Main rapeseed proteins are globulin cruciferin (60%) and albumin napin (20%) [67,68], containing an important amount of glutamine/glutamate and aspartic acid/aspartate residues (18.14% and 7.25%, respectively) [69]. The protein-rich biowaste obtained in the manufacture of the oil is mainly used for low-value applications [70,71], although some research about high-value applications has been pursued. Plastic materials have been obtained from canola mostly through casting [72–74] or from rapeseed by compression molding or injection molding [75,76].

Sunflower cake after extracting sunflower oil has been used for the development of protein-based bioplastics [77–79]. The protein content of the cake after oil extraction is high (~35%); however, the lignocellulose content is also high (~40%) [77]. Within the protein fraction, globulins are the most abundant (~58%), followed by albumins (~20%), glutelins (~14%), and prolamins (~3%). Because of its high protein content, it has mostly used for animal feed. A protein extract can be obtained at alkali pH with a high content of globulin and albumin [79]. Films have been obtained from sunflower through casting or extrusion [79,80].

2.4. Wastes from Animal Farming

Both casein and β-lactoglobulin are milk proteins extensively used by the food industry. However, they are also noticeably present in the wastewater from dairy factory plants (casein) or in the whey from cheese production (β-lactoglobulin), which can be

revalorized as a source of protein for the development of bioplastics [81]. Both sources are rich in glutamic acid (13.9% and 15.5%, respectively) and lysine (4.6% and 7.1%, respectively) [43]. Materials obtained from whey are similar to those prepared from caseins, characterized by transparency and flexibility and a water resistance that can be increased by crosslinking [82,83].

Blood (18% protein content) represents up to 4% of the animal weight and, and only 30% is used by the food industry, which would imply that over 3000 ML were discarded into municipal sewers and landfills in 2016 [84–86]. Plasma obtained after a centrifugation/drying process possesses a protein content that lies around 70% [87], which consists mainly of albumin (50–60%) and globulins (40–50%) [88]. The protein content is highly dependent on the animal species, being higher for bovine and porcine blood (~19%) and plasma (~6.9%) compared to poultry (~13 and 3.5%, respectively). Lysine (~7%), aspartic (~9.1%), and glutamic (~9.7%) acid contents are high for all those species [89,90]. Blood and plasma may be used for nonedible applications, such as packaging [91]. Thus, blood meal has been successfully extruded and injection molded [92–94], while more recently the plasma fraction has proven its potential as the basis of superabsorbent materials [95–97].

Around one third of the fish caught globally is used to produce protein-rich marine byproducts for animal feeding. For instance, a fish meal containing 59.0–68.5% of protein may be obtained [98]. Moreover, during fish processing, around 20–80% of waste, depending on the level of processing and type of fish, is generated which can also be used as fish meal [99]. Fish biowaste can also be used for production of proteins, oil, amino acids, minerals, enzymes, bioactive peptides, collagen, and gelatin. Most of the fishmeal is consumed by the aquaculture industry, but it could be employed for the production of green materials, through compression or extrusion [99,100].

Collagen represents 30% of the animal protein content and may be obtained from different byproducts of the meat industry, mostly pig skin (46%), bovine hide (29%), and pork and cattle bones (23%) [101]. One third of collagen is glycine, which is also rich in proline and hydroxyproline residues (~23% of the overall amino acid composition) [102,103]. Gelatin is produced when collagen is cooked or denatured by heat, being relatively cheap and abundant [104–106]. Its excellent ability to form films for both food and biomedical applications has facilitated its processing through casting, extrusion or electrospinning, displaying flexibility, good moisture and oxygen barrier properties, and excellent biodegradability [83,107,108].

Keratin comprises a mixture of high-molecular-weight fibrous proteins whose properties are greatly influenced by the methodology (chemical, enzymatic, and ionic solution) employed for its extraction from different epidermal appendages (mainly, feathers and wool, but also nails, claws, beak, hair, or horns) [109]. The amino acid composition may vary depending on the source as well as on the animal breed or diet [110], being the cystine content usually high [111–114]. Transparent materials have been obtained primarily by casting, resulting in water-sensitive films with adequate UV barrier properties and thermal stability up to 200 °C [109,114]. The toughness of these materials can be enhanced through crosslinking [109].

2.5. Microalgae from Sewage Plants

Different microalgae species can completely remove nitrogen and phosphorus from wastewater, being a useful tool for sewage plants [115]. These aquatic microorganisms possess a high protein percentage, with species like *Arthrospira platensis* or *Chlorella* sp. containing around 55 of protein in dry weight, and a lysine content similar to that of soy protein (~3.5%) [43]. Moreover, they offer the advantage of not requiring any soil to develop and allowing the use of nonpotable water as a culture medium when grown in wastewater [116]. The use of microalgae in plastic materials production is also interesting as scalable production seems to be more cost-effective because no prior treatment is needed before their processing [117]. Bioplastics materials have been obtained from microalgae biomass, although mainly blended with petroleum plastics or bioplastics [118,119].

3. Processing of Bio-Based Materials

The production of biodegradable materials is one of the most promising and studied pathways to handle the extremely high amount of biowastes and byproducts that the agri-food industry produces every year [58,97,120–123]. In this sense, the processing techniques and parameters selected strongly influence the end-use of the material developed. Commonly, these new materials are processed by traditional techniques used for synthetic plastic. However, a specific redesign is needed since these green materials require a different range of processing parameters due to their different composition and properties [124] which would influence their final characteristics, which definitely should be different from those of common synthetic materials [125,126].

The processability of a protein-based raw material is typically achieved either by its solubilization in an adequate solvent followed by a wet technique (casting or electrospinning) [127–129] or by a dry technique (e.g., extrusion and injection molding), which previously requires its blending with a low-molecular-weight component acting as plasticizer [130]. In the latter case, the plasticizer content is important for optimum control of the processing parameters, which are crucial to modulate the final properties of the materials. Furthermore, the amount of plasticizer alters the glass-transition temperature (T_g), a key processing parameter to consider during its processing [131,132]. The most extensively reported techniques employed in the production of protein based-materials are described in the following subsections: compression molding, injection molding, extrusion, three-dimensional (3D) printing casting, and electrospinning.

3.1. Compression Molding

Compression molding has been used since the early twentieth century for manufacturing plastics [133], although its batch process nature has resulted in a major industrial limitation [133]. During compression, the pre-cured or melt polymer is enclosed into a mold cavity and subjected to a large pressure [134]. In many cases, protein is mixed with a plasticizer to obtain blends that are then confined into the mold and compressed [135]. To perform the process correctly, the mold temperature must be slightly higher than the T_g of the protein blend. Therefore, temperatures over 60 °C should be generally selected [97,136]. This processing technique does not require high flowability; therefore, the obtained materials can be reinforced satisfactorily with fibers [27,137–145]. In literature, numerous studies have used different protein sources with this technique, such as soy [146], gluten [33,147–152], cottonseed [153,154], egg white [155], sunflower [156], corn [39,130], or whey [120,157]. As highlighted elsewhere, the properties of the compression-molded materials depend greatly on the processing temperature, which is usually around 100–120 °C [39,130,148]. Pressure, commonly around 10 bar for 2–10 min, is less influential than temperature [120,153,157].

3.2. Injection Molding

When intricate complex geometries and/or dimensional precision are required, injection molding is broadly used in polymer manufacturing, provided that the production is at a large scale [124]. This processing technique is commonly carried out through two stages: first, protein flour and the plasticizer (which is essential in this case) are conveniently mixed, and subsequently, blends are introduced into the injector feeding or cylinder, where the sample is heated if required. Then, the injection pressure is applied by means of a plunger, forcing the blend to flow through a nozzle into the mold cavity. After injection, the pressure is reduced and maintained constant for a period required to allow physical and/or chemical crosslinking of protein segments (i.e., the holding stage) [18,158]. The main control parameters are temperature (of the cylinder and mold), pressure, and time (injection and holding) [124]. In these terms, the mold temperature has been pointed out as the most influential parameter in this technique, where pressure exerts a lower impact [159]. Several studies have highlighted that changes in the mold temperature (and/or in the holding time at which the blend is exposed) can modulate the final properties of the product [17,95,97,121,160], modifying the water uptake capacity and

rheological and mechanical properties [32,59,97,159]. Different protein sources have been injection-molded, such as soy [18,58,59,62,161–168], sunflower [77], albumen [155,160], porcine plasma [95–97], pea [121,159], whey [144], rice [169], or gluten [32,35,170].

3.3. Extrusion

Extrusion is widely employed for the manufacturing of plastics with constant section [171–173], with the extra benefit of its continuous processing mode. In this technique, temperature is controlled along a cylindrical barrel containing a screw that allows the mixing and the transport of the material from the hopper to the die. During processing, the protein/plasticizer blends combine thermoplastic behavior with heat-induced crosslinking of different nature (e.g., protein aggregation and disulphide bonds). The selection of the temperature profile along the barrel is extremely important [174]. Therefore, a previous thermal characterization of the blend is helpful, using the T_g, which depends on the glycerol content [34,175], as the temperature threshold that must facilitate the flow inside the extruder and through the die [176]. Commonly, lower temperatures are required for protein-based blends, due to their usually lower T_g and to avoid massive crosslinking that would impede the process [171,174]. Furthermore, shear impact, time, and specific mechanical energy have been also pointed out to be key parameters to control the extrusion process [177,178]. At the extruder die, the material is conveniently shaped [178] and successively cooled down [172].

The protein more extensively processed through this method has been wheat gluten [34,36,171,177–183], followed by soy protein [184–189].

3D-Printing

In the most extended 3D technique (fused deposition modeling, FDM) for the automated and additive manufacturing of 3D objects, after layer-by-layer deposition, no excessive equipment investment is required [190], and relatively low energy is demanded per batch [158]. However, its main competitive drawback is referred to the time required for great productions [191]. This strategy has been developed fundamentally for polymeric materials, and it has been used in different fields such as biomedicine [191,192], electronic [193,194], or food [195,196], among others that typically involve small-scale production and high-value-added products. Although this manufacturing strategy has been scarcely exploited using protein-based products, some studies have been developed using pea [197], plasma [198], soy [199], and milk proteins [200,201]. All these studies highlighted the importance that rheology exerts on the 3D printing process. Thus, the main control parameters are those which exert influence over the rheology, such as temperature and shear rate (related to the flow through the nozzle).

3.4. Casting

The processing of protein-based biowastes into bioplastics films by casting is the most reported strategy, with applications as coating or packaging extensively described [127,202,203], despite generally possessing lower mechanical properties than those of synthetic materials [128,204]. It is a wet processing technique in which a prior disruption of linkages and disulphide bonds is carried out through a chemical reagent [205]. Then, the protein source is solubilized in a proper solvent, along with the plasticizer and other components such as crosslinking or antimicrobial agents. To produce the protein-based film, the solution is first spread, and then the solvent evaporation or drying is produced [127,206]. This procedure is mainly controlled by the pH and temperature of the solution, as well as by the selected solvent [206]. Based on its high content in cysteine residues, which are key since they promote covalent bonds [207], gluten is the protein most employed to give rise this kind of biodegradable films [207–225]. Furthermore, several studies were aimed at obtaining films of protein-based materials by casting, such as zein corn [226–229], soy [61,230,231], milk [232–236], sunflower [79], pea [237–240], and fish [205,217,241–244].

3.5. Electrospinning

The electrospinning technique produces nanofibrous polymeric materials using a high-voltage electric field [245–247]. The polymeric solution is confined in a syringe and flowed out through the needle using a syringe pump. When an electric field acts over the polymeric solution, the so-called Taylor's cone is formed at the end of the needle [248]. Thus, the produced flow is boosted toward the collector by applying a direct electric current field (commonly from 5 to 25 kV) connected to the collector and needle, which are usually placed away at a distance of 10–20 cm [249,250]. Electrospun materials are formed by ultrathin fibers with diameters in the nanoscale and commonly possess low density and a high porosity, resulting in materials with large specific surface areas [251]. If a certain degree of alignment is required for the nanofibers (e.g., biomedical functions), a rotating collector should be employed during their production [252–254]. The main parameters to control the morphological characteristics of the fibers formed are the type of polymer and solvent used, the surface tension, the viscosity of the solution, the flowrate, and the voltage applied [255]. Other parameters that may affect the process are the electrical conductivity, the presence of electrostatic interactions, and the distance between the needle and the collector.

Although it is difficult to carry out the electrospinning of protein solutions, they could be denatured to some extent to induce the process [251,256–258]. For aqueous protein solutions, pH is also a key parameter since it may modulate the charges of protein surfaces and protein solubility. Several studies have been focused on the electrospinning of protein solutions using gelatin [246,259–261], soy [262], egg albumen [263], silk fibroin [264], or whey protein [265].

4. Characterization of Protein-Based Materials

Any material processed for any purpose (e.g., packaging, coating, and agricultural) must reach some specific characteristics to properly provide the functionality required. Thus, the mechanical properties, the thermal behavior, and/or the optical properties of these materials should be controlled to meet the requirements of their final use. Characterization techniques quantify the macroscopic parameters, relating them to the microscopic (even molecular) structure of the materials. The most important characterization techniques are explained in the following subsections.

4.1. Mechanical and Rheological Properties

Mechanical properties help to understand and predict the behavior of materials subjected to different kind of stresses. The majority of polymer-based materials show a variety of viscoelastic responses after an applied strain or stress. When tested, materials are typically submitted either to continuous or oscillatory deformation. It can be noted that only some of the most important tests for material characterization are described in this section.

4.1.1. Tensile Strength Tests

During these tests, the material is subjected to axial deformation at a constant rate until breakdown. The results are plotted in stress–strain curves where three different stages can be typically differentiated in polymer-based materials: (i) Initially, the strain suffered by the material is linearly proportional to the stress applied, due to the elastic deformation of the material. From this initial constant slope, the Young's modulus (E) is defined. (ii) Subsequently, a remarkable decrease in the slope takes place, showing a nonelastic deformation. The maximum value or ultimate stress (σ_{max}) is commonly reached at the end of this section. (iii) Finally, the stress decreases due to the fast reduction of the cross-sectional surface, ending the test when the probe collapses at maximum deformation (ε_{max}). [266]. The mechanical properties of different materials obtained from several biopolymers, such as rice [267], albumen [268], plasma [95,97], soy [160], pea [121], or whey protein [32,120], have been analyzed through this technique. A wide spectrum of

values for the mechanical parameters was obtained, depending not only on the source, the biopolymer/plasticizer ratio, and the presence of additives (e.g., a crosslinker) but also on the processing technique and conditions.

4.1.2. Rheological Tests

Rheology is the science that studies the flow and deformation of matter and applies to a wide range of materials [269]. Moreover, its importance is key since the majority of polymeric materials show complex viscoelastic behavior [270], which is the result of the combination of solid-like elastic properties and fluid-like viscous properties [271]. Rheological tests are normally carried out using oscillatory or continuous deformation.

Dynamic Mechanical Analysis (DMA)

In these tests, an oscillatory deformation (with a small-strain amplitude, γ_0) is applied to the sample, obtaining a sinusoidal stress response with amplitude σ_0, below the limit for the linear viscoelastic region (LVR). The viscoelastic behavior of the sample is obtained relating both stimulus and response, giving rise to the linear viscoelastic functions that remain independent of the applied strain [270]. The most important linear viscoelastic functions are the storage modulus (E′ or G′), which is a measure of the elastic response of the material; the loss modulus (E″ or G″), representing the viscous properties; and the loss tangent (tan δ, where δ is the phase angle). This parameter represents the relative predominance of the viscous over the elastic properties (tan δ = E″/E′ or G″/G′). The viscoelastic characterization of the material consists of obtaining the dependence of these parameters on frequency and temperature.

Common dynamic mechanical tests are: (i) stress (or strain) sweep tests, which allow the determination of the LVR through the identification of a critical strain value; (ii) frequency sweep tests, which provides information about the unperturbed microstructure of the sample [33,176]; and (iii) temperature sweep tests, in which the dependence of the material on temperature is analyzed. These measurements can be carried by applying different deformation modes (e.g., compression, tension, bending, and shear). The nature of the sample tested would determine the type of geometry and mode that better fits the analysis (parallel plates [272], rectangular [59], dual cantilever [169,273], or three-point bending, among others).

The results of these tests could give relevant information that can be related to processing parameters of the materials [97,274] or even predict some correlations with other properties, such as printability [198] or biodegradability [275]. This rheological characterization has been largely performed for different protein-based bioplastics, such as soy [58], plasma [95,198], zein [276], or pea [121,159], among others [277,278].

Continuous Deformation Tests

Stress relaxation and creep tests could be regarded as the most useful long-term assays using continuous deformation. Stress relaxation tests record the evolution of stress until it reaches a plateau while applying a constant strain γ_0. On the other hand, creep tests apply a constant stress σ_0 while measuring the progressive deformation of the sample.

Within the LVR, a relaxation modulus (G(t)), defined as the ratio between stress (σ(t)) and strain (γ_0), and a compliance modulus (J(t)), defined as the ratio between deformation (γ(t)) and stress (σ_0), are defined for relaxation or creep tests, respectively. Representative relaxation and retardation times may be defined from these tests. Creep tests have been used to identify the crosslinking degree by glutaraldehyde in gelatin-based materials [279]. Furthermore, soy-based [280] and fish-based materials [217] have been rheologically characterized through these kinds of tests.

4.2. Thermal Characterization

The knowledge of the thermal events when samples are subjected to changes in temperature is quite relevant to identify a suitable end-use of the material. Glass-transition

temperature (T_g) should be identified to properly set the processing parameters of techniques involving heating/cooling of the sample [183]. T_g is the temperature above which the mobility of polymeric chains increases prominently as secondary interactions disappear, resulting in biopolymer conformational changes [174,281]. This temperature depends on the nature of the biopolymer (e.g., amino acid sequence) and the amount of plasticizer used in the formulation [282].

The most extensively used tests to measure the changes induced by temperature in materials are differential scanning calorimetry (DSC), thermogravimetric analysis (TGA), and dynamic mechanical thermal analysis (DMTA).

4.2.1. Differential Scanning Calorimetry (DSC)

The main thermal events observed through DSC in protein-based bioplastics are protein denaturation and glass-transition temperature. They could be clearly identified in a thermogram since denaturation is an irreversible event commonly recognized as a minimum in the curve (exo-up type plot). On the other hand, a reversible glass transition is showed as a change in the slope or an inflection point. Additionally, protein aging prior to processing can be also determined by DSC, being identified as a minimum in the curve, but at a much lower temperature than the denaturation temperature [283]. Thus, two scans are typically performed for separating reversible and irreversible thermal events. Although the irreversible events (denaturation) are only shown in the first scan, the reversible events (glass transition) are always observed. Physical aging, being a reversible event, requires longer times and is normally not observed in the second scan neither. This assay is applied to protein-based powder, blends, or materials [17], and it has been used to characterized microalgae protein [284], soy protein [285], wheat gluten [33,149,286], egg yolk [287], pea protein [159], plasma protein [97], canola protein [74], and whey protein [288], among protein systems used for the bioplastic formation.

4.2.2. Thermogravimetric Analysis (TGA)

TGA measurements can be used to determine the thermal stability of protein/plasticizer blends and bioplastics since it measures the weight reduction of a sample as the temperature increases. Several regions can be observed in protein-based bioplastics. Thus, below 150 °C, the diminution in the weight is caused principally by the loss of volatile components and water [283,289]. At higher temperatures (c.a., 180–350 °C), the weight loss has been attributed to protein degradation [290]. Moreover, these thermograms can indicate the fat content of isolated proteins [125], the presence of volatile components after processing [144], or the response of some active ingredients such as citric acid when used in the production of soy-based porous materials [58]. Moreover, this technique has also been used to determine the thermal stability of protein-based materials after the acylation of the protein [163].

4.2.3. Dynamic Mechanical Thermal Analysis (DMTA)

This technique measures the rheological response of a material as a function of temperature, after application of a small-amplitude oscillatory strain (or stress), as previously commented in Section 4.1.2. This technique relates the evolution of the viscoelastic moduli of biopolymers with temperature to their molecular structure, giving complementary information to that obtained from DSC and TGA analysis. T_g can also be determined from DMTA tests, although its value is dependent on the heating rate and the technique employed [183,291].

Several protein-based materials have been tested through DMTA, such as materials obtained from egg albumen [155,268], soy [17,59], microalgae [284], pea [121], wheat gluten [33,149,290], rice [169], bloodmeal [92], plasma [97,292,293], and canola [76], among others.

4.3. Morphological Properties

Microscopy is widely applied to bioplastics to analyze their morphology. Scanning electron microscopy (SEM) is the most widely used for the materials and matrices based on a wide variety of proteins, such as soy [17,18,165], pea [121], wheat gluten [33,149,150], rice [169], or plasma [96,292]. Furthermore, the surface of bioplastics films could be also studied by SEM [294,295]. On the other hand, when transmission electron microscopy (TEM) is used some considerations have to be made as this technique requires electron transparency, which may be acquired directly by thin bioplastic film [296] or by cutting thin slides from bioplastic probes [185,297]. Additionally, atomic force microscopy (AFM) is useful to study the topography of the three-dimensional surface of protein-based materials [155,296]. Finally, confocal laser scanning microscopy (CLSM) can exploit the autofluorescence of certain proteins, being quite useful to characterize the morphology of protein-based films [298].

4.4. Optical Properties

The optical properties, such as color, transparency, or refractive index, may be sometimes neglected, but they are quite important for several end-uses of these materials. They are highly dependent on the nature of the protein source [17], the composition of the system, the amount of plasticizer [121], and the processing technique or conditions [159,299]. Transparency has been used, for instance, to quantify the presence of some microbial polysaccharides in gluten WPI-based films [300].

5. Applications of Protein-Based Bioplastics

The main agri-food industrial biowastes and co-products for bioplastic applications has been described in the previous section. Food biowastes can be used for the production of biofuels [301], but the present review focuses on their application in the field of greener materials, which has been extensively studied but is less exploited commercially, especially the protein fraction. The selection of a suitable biopolymer source is key in the development of any final product with a particular application, which may be chosen based not only on its processing suitability but also on consumer requirements. For instance, animal proteins are commonly rejected in cosmetics, despite being widely accepted in agricultural applications [302]. Moreover, the design and development of bioplastic materials need to bear in mind the accordance between service conditions and the final mechanical and functional properties of the material developed. Although many researchers focus on the mechanical properties of bio-based bioplastics, many applications (i.e., superabsorbent, drug delivery, controlled release, etc.) do not require excellent mechanical properties for their final usage [18,97]. Some critical requirements are demanded for these bio-based materials when used in food applications. For instance, food quality and safety during storage should not be compromised. Moreover, extended shelf-life and a reduced permeability to volatile compounds (i.e., oxygen and moisture) are also pursued [303]. This section summarizes the main applications for the agri-food industry biowastes whose end-use can be linked to the goals of the bioplastic industry. Moreover, this section also addresses the requirements of these bio-based materials for certain applications.

5.1. Food Packaging and Coating

Apart from the specific safety and security requirements in food applications, the new generation of packaging materials aims at biodegradability, in order to avoid accumulative pollution, together with advanced extra features [304,305]. The requirements in food packaging and coating depends on the nature of the food contained [306]. For instance, to extend the expiration date of vegetables, respiration and transpiration rates must be reduced during storage (i.e., controlling temperature, relative humidity, light, and gas permeation) [307]. However, these requirements should be adapted for every specific food application. Dairy products are mainly degraded by oxidation and microbial growth, leading to nutrient loss, which causes color changes, as well as the appearance

of undesirable flavors [308]. Analogous effects are observed in meat products, where the CO_2 and O_2 levels should be kept in a suitable range [309]. Eventually, a wide variety of food products are maintained frozen during its conservation, regardless of the nature of the food matrix. Although low temperatures prevent microbial growth, a suitable package design for food preservation is still required when freezing food products. Light and oxidation promote the degradation of vitamins and pigments, destabilization of proteins, and oxidation of lipids. Thus, package design should avoid those phenomena. Additionally, packages for frozen food products should avoid moisture loss by water sublimation (i.e., low water permeability), which results in undesirable consequences such as weight loss, the appearance of burns, and morphology changes [310].

These undesirable phenomena in different types of food products have been so far commonly overcome by the use of synthetic polymers. However, the food packaging industry is facing a challenge in providing an adequate solution for correct food conservation and environmental sustainability. In this sense, zein has been used for the preservation of tomatoes, avoiding color changes [311]. This protein has been also used for fruits, showing a reduction in weight loss, which is something expected from every coating [312]. Zein films have been also used for extending the shelf-life of dairy products, reducing protein oxidation [313]. Soy protein has been used in the development of coatings which prevent peanut deterioration [314]. Gelatin has been extensively used for the manufacturing of protein films with coating applications for the conservation of fresh products (both meats and vegetables) [315–318]. Protein biowastes from the dairy processing industry have also been used for the manufacture of films. Thus, sodium caseinate and whey protein concentrate were the most common proteins for a new generation of films for food applications not only in dairy products, such as cheese, but also in other food products, such as meat [319–321]. Keratin has been used for the coating of meat derivatives, exhibiting good properties for the formation of films [322]. Eventually, as protein hydrophobicity is not enough to avoid the water permeability required for some applications (e.g., frozen products), improvements should be addressed to reduce the abovementioned side-effects of water loss [304]. In any case, suitable values were obtained for frozen fishes [323,324].

Most functionalities of biodegradable protein-based materials find applications in food active-packaging, including the controlled release and immobilization of substances for specific purposes (i.e., antioxidant release, enzyme activity, gas selective permeability, etc.) [325]. Active packages are able to protect and interact with the food they contain by "deliberately incorporating components that would release or absorb substances into or from the packaged food or the environment surrounding the food" [326]. Thus, these materials can be considered as active packaging since there is an interaction between the material and the food product contained including activities such as controlled release, which improve its preservation [327,328]. Although some alternatives from synthetic polymers have been proposed, bio-based active packaging also offers the advantage of avoiding phthalate leaching typically produced in synthetic polymers [329]. Thus, the evaluation of active-agent release from biopolymers has received outstanding attention from some years ago, with promising applications in the food packaging industry [152]. More specifically, the efficiency of active packaging for antimicrobial activity is based on the match between the releasing rate of the active agent and the decrease in the growth kinetics of the target microorganism [330]. These innovative materials can extend the shelf-life of food products, providing an increase in microbial safety. This increase in food security can be achieved by the incorporation of antimicrobial agents into the bulk of the biopolymer matrix or onto the biopolymer surface [331,332]. Several authors have used co-products from the dairy industry with antimicrobial constituents (i.e., oregano, rosemary, and garlic essential oils) for the manufacturing of edible WPI-based antimicrobial films [332]. This same application was found for other protein-based materials such as zein [333] and soy protein isolate [333] by adding a mixture of lysozyme and nisin. However, other active agents can be also used. To this end, Redl et al. [334] added ascorbic acid to confer antimicrobial properties to gluten-based bioplastics. Gelatin was also mixed with

chitosan to confer antimicrobial properties [335]. Another biofunctional property, which is typically required in food packaging, is the antioxidant activity. More fresh products are demanded nowadays by consumers [336]. However, as abovementioned, the lipid oxidation reactions bring undesirable off-odors and off-flavors, as well as changes in texture and color [337]. Moreover, apart from physical changes, lipid oxidation can also generate toxic compounds such as aldehydes [338]. However, the use of synthetic antioxidants such as butylated hydroxytoluene (BHT) and butylated hydroxyanisole (BHA) should be avoided since they have been related to undesired health side-effects [339]. Gelatin films have been developed together with citronella oil to manufacture films with antioxidant properties [340]. The release of carotenoids from seaweeds with proved antioxidant activity in synthetic packaging [341] suggests that the manufacture of algae-based bioplastics might be considered to replace the synthetic polymer if the processing method selected do no alter them. Moreover, it can be also indicated that whey and soy protein isolates have been used in propylene-based materials to confer antioxidant properties [342] to them, including even antifungal ones [343].

5.2. Absorbent and Superabsorbent Materials (SAMs)

Some absorbent materials exhibit an exceptional water absorption capacity. They are named superabsorbent materials (SAMs), provided that they can absorb an amount of water greater than 1000% their own weight [344]. The capacity of SAMs to hold a huge amount of water is based on their ability to convert their structure into a hydrogel which is not dissolved when the solvent (i.e., water in most cases) is trapped onto the 3D network [345]. The synthesis of the first fossil-based superabsorbent polymer dates back to the late 1930s, while the first commercial SAM was marketed in 1978 [346]. However, these synthetic polymers show very low biodegradability, and hence, their replacement would lead to a positive reduction in the environmental impact of SAMs [347]. Carbohydrates are key biopolymers in the manufacture of this type of materials [348–350], but many researchers have also focused their investigations on protein biowastes and co-products from the agri-food industry to generate biodegradable absorbent and superabsorbent materials [97,123,351,352]. Different processing techniques have been proposed for the manufacture of protein-based absorbent and superabsorbent materials, covering from injection molding [58,59] to casting [353]. The most important applications for these materials are related to personal care and agriculture [354,355]. However, these materials are also applied in the food industry, especially in the control of moisture during food storage and preservation [328]. The interest in SAMs from the food industry is reflected by the publication of several patents aimed to use absorbent materials for food packaging [356–358]. In this sense, zein has been used for the synthesis of superabsorbent hydrogels, showing applications as metal ion chelators, which in turn can avoid food oxidation [359]. Moreover, co-products from the cattle industry have also been used as raw materials for the generation of superabsorbent materials for food applications, such as whey and casein [360–362].

Due to a lower added value, the agricultural applications reported in the literature are lower. Proteins with low-value applications such as keratin and potato protein concentrates were proposed for the development of superabsorbent materials to be used in agriculture [113,123]. More details about this end-use are shown in the following Section 5.4.

Moreover, the final end-use application is not indicated in many cases, as authors developed SAMs with any final end-use, which includes food packaging, agriculture, personal care, and even scaffolds for cell growth, among others. In this sense, soy protein isolate has been widely used for the manufacture of these materials [58,59,353]. Apart from soy protein, other proteins co-products (i.e., canola and rapeseed protein concentrates) were also used for absorbent materials [76,113,363]. Additionally, plasma protein, a hydrophilic co-product from the meat processing industry, has proven to be useful in the fabrication of absorbent plastic films [91,364–366] and superabsorbent materials obtained by injection molding [96] and even 3D printing [198]. Also from the meat industry, gelatin showed suitable application as SAMs [367]. Eventually, the underutilized bio-resource marine

algal seaweeds can be also converted into added-value superabsorbent materials. Thus, apart from the formation of hydrogels with κ-carrageenan and alginate [368,369], the protein fraction from micro- and macro-algae is suitable for the generation of absorbent materials [119,284].

Furthermore, soy proteins have also been chemically modified, improving the superabsorbent properties of the materials produced by the acylation with ethylenediaminetetraacetic dianhydride (EDTAD) or succinic anhydride [59,62,363]. Other authors have also used EDTAD to functionalize other cereal proteins, like wheat gluten, improving the initial water absorption properties of this co-product [370].

5.3. Agriculture

Polymers have found some interesting applications in agriculture, such as controlled release of specific micronutrients or pesticides, as porous and water-holding matrices that avoid the critical soil drought. Moreover, some of these applications have been linked to the superabsorbent ability of the polymers employed. In the 1980s, several companies manufactured and commercialized composites materials based on plant biopolymers and polyethylene aiming at simultaneously providing biodegradability and fulfilling the requirements of the consumers. However, this strategy resulted in the release of polymer chains and microplastics to the medium, which caused environmental damage due to the toxic polyethylene residues [371]. In this way, microplastics disposed on land end up in rivers that flow into seas, eventually contaminating aquatic environments [372]. Therefore, the reduction of sea microplastics must go through the reduction of inputs onto the inland [373]. Microplastics are particles lower than 5 mm built from larger particles pieces. These particles cause damage to marine species, which eventually may affect the full food chain [374]. The removal of these small bodies is a real challenge; therefore, the best option is to avoid its generation [373,375]. Therefore, if the aim is to increase polymer biodegradability for agricultural applications, the path can go through the use of biopolymers whose degradation does not release undesirable macro- and micro-plastics [371]. To this end, superabsorbent biodegradable polymers have been proposed for their use in water-saving applications as well as for the controlled release of essential nutrients for plants [376,377]. These superabsorbent materials increase the overall porosity of clay-based soils, being recommended for their use in dry agricultural areas to reduce the drought stress during plant growth [378]. Moreover, plant nutrients may also be entrapped within superabsorbent matrices for their controlled release, hindering the water losses due to evaporation, and reducing the irrigation [378]. Although there are polysaccharide-based materials for these applications [379–381], Capezza et al. [370] recently reported the use of gluten co-product as raw material for the manufacture of protein-based superabsorbent materials with agricultural applications. These authors used a crosslinking agent (i.e., genipin) together with EDTAD to improve probe swelling, which in turn increased the water absorption capacity of these materials. Moreover, SAMs with agricultural applications have been generated from other proteins, such as soy [59], canola [363], zein [382], blood plasma [95], keratin and gelatin [383], milk proteins [384,385], and seaweed [347,386]. Moreover, bioplastics can also be used for the controlled release of both micronutrients and pesticides. This is the case of gluten-based bioplastics (releasing pesticides) [387], soy-based bioplastics (releasing zinc and mineral nutrients) [388,389], or even zein to prevent salt-leaching [390]. These results evidenced the potential of co- and by-products from the agri-food industry to produce materials that can be reused in agricultural applications with greater environmental performance. Accordingly, the life cycle of some crops may be enhanced since they can be used to obtain co- and by-products that in turn may lead to bioplastics that could be reused again in the first stages of their own life cycle (e.g., as water suppliers or for nutrient delivery to enhance crop growth).

5.4. Scaffolds

Scaffolds are temporary supporting structures which are used to generate the final structure. In material science, scaffolds are typically used for tissue engineering. Thus, although the properties typically found in scaffolds materials (e.g., high porosity and water absorption) can confer them interesting features, they are only named scaffolds when serving as supporting structure to facilitate cell growth for a certain period of time in tissue engineering [391]. Consequently, scaffolds need to play four fundamental functions: (i) form a complex structure which allows cells to rebuild the original 3D structure; (ii) be temporal support of functional demands (e.g., mechanical support); (iii) enhance tissue regeneration (i.e., the release of bioactive compounds which allow cell fixation and growth, favoring their transport); and (iv) be able to be attached to the surrounding tissue [392,393]. The exposed surface is key for these materials since the larger the surface available, the more cell interactions take place. Thus, the pore size and morphology can significantly influence the performance of these materials, where optimum values should be found for different surrounding microenvironments (where cell dimension also must be considered) [261,392].

The research and further development of protein-based scaffolds have been focused on films, plastics, foams, gels, and even composites materials [394]. Although these materials are developed for tissue engineering, other biomedical applications have been found, such as drug delivery systems and biosensors [395].

Collagen/gelatin has been widely selected as raw material for the manufacture of scaffolds, since it not only performs a supporting function but it is also involved in a wide range of tissue functions [396]. Therefore, collagen has shown excellent properties in scaffolds, being manufactured in different ways such as phase separation [397,398] or electrospinning [399]. Moreover, other proteins have also been used for the development of scaffolds. Fibrous membranes produced by electrospinning were obtained using PLLA/keratin as raw material [400]. In this case, the presence of keratin facilitated the interactions between osteoblasts and the membrane, favoring cell growth. Nanocomposites from keratin/hydroxyapatite have been generated following a co-precipitation method, showing good biocompatibility tested by in vitro tests [401]. Keratin has also been used in combination with natural polymers (i.e., gelatin and chitosan) for tissue engineering applications [402]. In this context, plant proteins (e.g., zein, soy protein, and wheat gluten) can also be used as raw material for scaffolds. They provide suitable mechanical properties, while at the same time being biocompatible. Moreover, their typically low solubility confers them enough stability in aqueous media to be considered appropriate for tissue engineering applications [403]. Eventually, some authors have also tested the stability of seaweeds for the manufacture of scaffolds, being used in most cases in combination with other polymers such as PLA and cellulose [404,405]. All these results evidence that protein biowastes and co-products from the agri-food industry can even be employed on specific applications such as scaffolds for cell growth.

5.5. Other Applications

Although the most important applications for bioplastics from co- and by-products from the agri-food industry have been mentioned above, other applications can be proposed. Nanoparticles from collagen-serum albumin composite have been used for drug delivery [406]. In the biomedical field, keratin has been used as nanosuspension to analyze cell proliferation in tissue engineering applications (as an alternative to fibronectin and collagen, typically used for this purpose) [407]. Although some authors have investigated the use of protein-based bioplastics in the textile industry, proteins typically lead to bioplastics exhibiting a fairly low elongation and are therefore brittle, which do not convert them into suitable raw material for textile applications [408]. This is not the case of the automotive industry, where Mohanty et al. [162] proposed the use of soy-based composite materials (reinforced with fibers). This same approach was found by Guilbert et al. [206] when soy-based bioplastics were hardened with formaldehyde. Saenghirunwattana et al. [145] proposed the use of a zein protein concentrate for the manufacture of composite materials

with applications in construction. Even electrical properties (dielectric constant) have been analyzed and modulated in soy-based bioplastics for electrical applications [409].

Main protein sources from biowastes together with the processing technique used and application pursued are summarized in Table 1.

Table 1. Main protein sources from biowastes used in the development of plastic materials commented in the present review.

Source	Protein	Processing Technique	Plasticizer/Solvent or Carrier	Application	References
Starch	Wheat gluten	Acylation	glycerol	Superabsorbent materials	[34]
		Compression moulding	glycerol	Horticulture (release of pesticides)	[276]
		Compression moulding	glycerol	Packaging	[29,154]
		Compression moulding	Water/glycerol	Edible films	[153]
		Compression moulding	glycerol	Biodegradable films	[275]
		Compression moulding/Injection moulding	glycerol	Disposable articles	[171]
		Casting	glycerol	Food packaging films	[162]
		Casting	glycerol	Disposable articles	[210]
		Casting	glycerol/ethanol	Edible films	[223]
		Extrusion/Injection moulding	glycerol	Superabsorbent materials	[32]
		Extrusion	glycerol	Disposable articles	[34]
		Extrusion	glycerol	Superabsorbent materials	[36]
	Potato	Acylation	glycerol	Superabsorbent materials	[120]
		Casting	Ethylene glycol, propylene glycol, glycerol, sorbitol and polyethylene glicol	Food packaging films	[46]
Bioetanol	Zein	Compression and casting	glycerol (compression) and glycerol/ethanol (casting)	Antimicrobial packaging films	[333]
		Casting	glycerol/ethanol	Packaging of tomatoes, reduction of color loss	[311]
		Casting	glycerol/ethanol	Apples and pears, reduction of water loss	[312]
		Casting	glycerol/ethanol	Reduction of oxidation in dairy products	[313]
		Casting	Ethanol/Polyols (sorbitol, glycerol and mannitol)	Food packaging films	[227]
		Extrusion	water/ethanol	Food packaging films	[51,52]

Table 1. *Cont.*

Source	Protein	Processing Technique	Plasticizer/Solvent or Carrier	Application	References
Oil	Soy	Casting	water	Edible films	[230]
		Casting	water and glycerol	Edible films	[227]
		Extrusion	glycerol	Disposable articles	[185,189]
		acylation-Injection moulding	glycerol	Superabsorbent materials	[60,62,163]
		Injection moulding	glycerol	Superabsorbent materials	[18,58,59]
		Injection moulding	glycerol	Horticulture (Zn incorporated)	[63]
		3D printing	water/gelatine and sodium alginate	Food matrix	[199]
	Canola/ Rapeseed	Casting	glycerol	Edible films	[72,73]
		compression moulding	polyvinyl alcohol and glycerol	Disposable articles	[75]
		injection moulding	glycerol	Packaging	[76]
		casting	glycerol, 1,3-propanediol, D-sorbitol, triethylene glycol, tetraethylene glycol	Films	[79]
	Sunflower	compression moulding	glycerol	Edible films or packaging	[156]
		extrusion/injection moulding	water	planting containers	[77]
		extrusion	water and glycerol	Edible films	[80]
Animal farming	Blood	Extrusion	Water	-	[92]
		Injection-moulding	glycerol	-	[93]
	Plasma	Casting	glycerol	Food wrap or coating	[91,365,366]
		Casting	glycerol	Food packaging	[364]
		Injection-moulding	glycerol	Superabsorbent materials	[95–97,292]
		3D printing	glycerol	-	[198]
	Keratine	Casting	glycerol, water, SDS	Food packaging, coating, medicine	[109]
		Casting	glycerol, polyethilene	-	[112]
	Gelatine	Casting	Water	Packaging and coating	[279]
		Electrospining	acetic acid and dimethylsulfoxide	Regenerative medicine	[259]
		Electrospining	2,2,2-trifluorothanol	Biomaterials	[288]
		Electrospining	Trifluoroacetic acid	Biomaterials	[260]
		Electrospining	Acetic acid	Tissue engineering	[261]
	Milk protein	Casting	Clycerol, Propylene glycol, sorbitor, sucrose and polyethylene glycol	Coating, food packaging	[233]
		3D printing	Water and sodium caseinate	Costumized food design	[200,201]

Table 1. *Cont.*

Source	Protein	Processing Technique	Plasticizer/Solvent or Carrier	Application	References
Animal farming	Casein	Casting	glycerol	Packaging	[82]
		Hydrogel by solubilization	Transglutaminase	Contolled release	[361,362]
	Whey	Casting	-	Food coating	[319]
		Casting	sorbitol	Active packaging	[321]
		Casting	glycerol	Food packaging	[232,233,235,332]
		Casting	water	Coating	[234]
		Casting	glycerol	Coating, edible films	[376,380,386]
		Freezing	glycerol and sorbitol	Coating	[324]
		Compression	Water	Food packaging	[145]
		Hydrogel by solubilization	glycerol	Coating, food packaging	[297]
		Electrospining	acetic acid	Coating	[265]
	Fish	Casting	glycerol	Active packaging	[340]
		Casting	glycerol	Edible packaging	[209]
		Casting	glycerol	Food packaging	[241,244]
		Compression moulding	glycerol	Active packaging	[141]
Sewage	Microalgae	Compression moulding	glycerol	Disposable articles	[116]
		Injection moulding	glycerol	Packaging	[118,284]

6. Future Trends

Findings gathered in the present review put into focus the wide versatility of bioplastics manufactured from agri-food industrial biowastes or co-products, although the limits for their applicability are still far from being fully explored by the scientific community. In the relatively near future, conventional plastics will disappear in single-use applications, following the European strategy for plastics in a circular economy, which aims to transform the way plastic products are designed, used, produced, and recycled in the European Union. Most current applications are focused on the use of lignocellulose, starch, or fats from food biowaste, and the protein fraction is mostly relegated to low-value applications (e.g., animal food). However, as highlighted in the several applications described above, there is a solid scientific ground to industrially exploit those protein-rich biowastes and co-products. Techniques like electrospinning or 3D-printing have yet to further develop their potential to do so, and proteins which are noncompetitive with the agri-food industry, such as rapeseed or keratin, may find a privileged position. However, the excess of co-products that are only minimally used by the agri-food industry despite being edible, such as blood from the meat industry, should be better employed in applications like those herein presented, in agreement with a circular economy. When bioplastics generated from biowastes and co-products such as those herein indicated are competitive, the laws of supply and demand will help to modulate their use.

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