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Starch in Plant-Based Meat Replacers: A New Approach to Using Endogenous Starch from Cereals and Legumes

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This review discusses the use of starch in plant-based meat replacing products. Starch is often added to meat and meat replacing products as a functional ingredient. The function of starch in those applications is investigated to be able to describe how it affects the production and structuring process as well as product properties. Often modified starch is used in these products, because of its improved functionality compared to native starch. Next to that, starch can also be present in meat replacing products as part of other ingredients, for example, when using legume or cereal meals or concentrates. It is discussed if this endogenous starch can have similar functionality and if not, whether it can be modified in a similar way as industrially modified starch. A new perspective on endogenous starch is proposed, demonstrating options for in situ modifications and promoting the use of less-refined and therefore more sustainable ingredients in fibrous meat replacing products.

1. Introduction

In the last decades meat replacing products have gained interest on a global scale, with predictions for greater growth in the coming years.[1–5] As a result, more products are appearing in the market that mimic meat or offer protein-rich alternatives to the consumers. One of the biggest challenges in producing these products is creating the adequate texture, flavor, and color.[6] The success of making those structures depends on both the ingredients used and the process to make the products.[7] An often applied combination of ingredients consists of protein (often from wheat and legumes), fat or oil, binding agents, flavors, and color agents.[8] Frequently also starch is used as an ingredient in commercially available meat replacers and real meat products, though mostly applied in low quantities. **Table 1** shows the application of starch in meat replacers and processed meat products currently on the market. In these products, starch is added as a minor ingredient next to other purified ingredients, such as protein isolates or concentrates. Starch often acts as a filler and can increase yield or modify water holding.[9] Such purposely added starch is called exogenous starch and can come from a different source than the protein. Exogenous starch can be modified to improve its functionality even further. The choice of starch is based on the functionality and availability.

Another reason to incorporate starch in meat replacing products, next to functionality, is sustainability. Endogenous starch, as opposed to exogenous starch, is naturally present as a component that remains to a certain extend in an ingredient after purification. The current focus of the industry is on the use of purified protein ingredients, for example, pea protein isolates, in which the endogenous starch is removed completely. As evident from Table 1, this approach sometimes requires even the later addition of exogenous, potentially modified starch for functionality.[35] Consecutive removal and addition of starch seems contradictory and inefficient from a sustainability point of view, as this requires a lot of energy and leads to high material losses. This is especially true for crops that are considered to have an important role in the transition toward a more plant-based diet, and for which no current industrial use exists for the starch. Typical examples are mung beans, cowpea, faba beans, and other pules. Losses in the form of (starch) waste streams could be avoided by using these ingredients in un- or less-refined form. Therefore, the use of such less-refined ingredients is far more sustainable and economic, especially in the situation where the individual components are recombined into a new ingredient mix to achieve the desired composition and functionality.[36] However, the functionality of the components has to be considered as well when evaluating their suitability for food products.[37]

Based on the information above, it becomes clear that the need for more sustainable food (ingredients) poses the following research questions: Is it possible to use endogenous starch as a functional component? If not, is it possible to modify starch that is present in a mildly refined protein-rich fraction in order to

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Table 1. Collection of meat and meat replacing products on the market that contain starch.

Application	Starch	Meat/vegetarian/vegan	Ref.
Albert Heijn Hamburger	Potato	Meat	[10]
Albert Heijn Runderbraadworst	Potato	Meat	[11]
Albert Heijn Shoarmareepjes	Potato	Meat	[12]
Albert Heijn Biologische Rundergehaktballetjes	Potato	Meat	[13]
GoodBite Vers Gehakt	Potato, corn, wheat	Vegetarian	[14]
GoodBite Hamblokjes	Potato, corn, wheat	Vegetarian	[15]
Quorn Meatless Nuggets	Wheat starch	Vegetarian	[16]
Garden Gourmet Schnitzel	Wheat flour, corn	Vegetarian	[17]
Vivera Kaasschnitzel	Wheat	Vegetarian	[18]
Vivera Wokreepjes	Wheat	Vegetarian	[19]
Albert Heijn Stukjes Als Van Kip	Wheat	Vegetarian	[20]
De Vegetarische Slager Visvrije Tonyn	Unknown	Vegetarian	[21]
De Vegetarische Slager MC2 Burger	Wheat	Vegetarian	[22]
Moving Mountains Burger	Wheat	Vegan	[23]
Moving Mountains Sausage	Wheat	Vegan	[24]
Beyond burger	Potato	Vegan	[25]
Vivera Krokante Schnitzel	Wheat	Vegan	[26]
Vivera Balletjes	Potato	Vegan	[27]
Vivera Steak	Wheat	Vegan	[28]
Vegan Zeastar Zalmon Sashimi	Tapioca	Vegan	[29]
Gardein Beefless Strips	Wheat	Vegan	[30]
Gardein Beefless Tips	Potato	Vegan	[31]
Gardein Chicken Strips	Potato	Vegan	[32]
Tofurky Chick'n	Corn	Vegan	[33]
Like Schnitzel	Corn, potato	Vegan	[34]

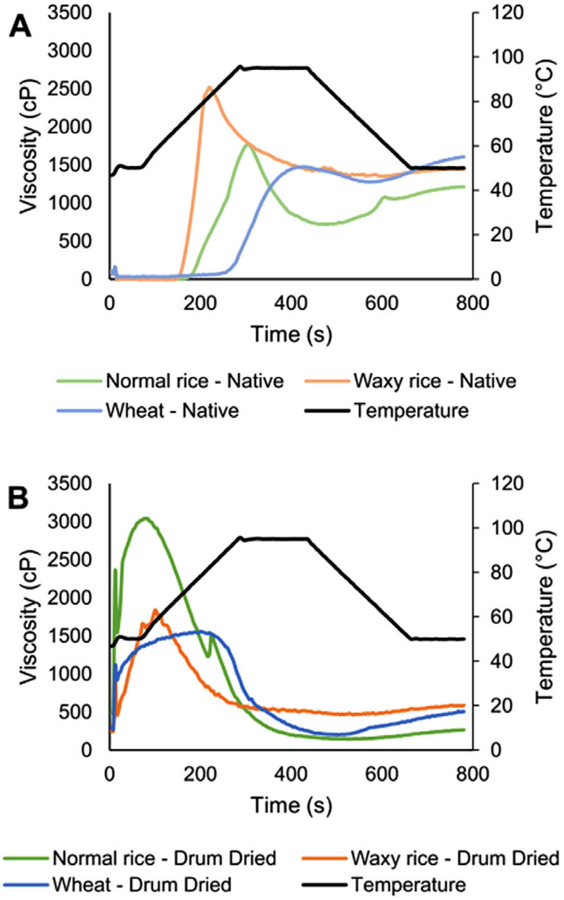

Figure 1. RVA profiles of (A) native and (B) drum dried normal rice, waxy rice and wheat starch. Reproduced with permission.[54] Copyright 2020, Elsevier Ltd.

functionalize it? This review aims to contribute to the answers by summarizing the current research on the role of starch in meat replacing products. The processes to structure plant proteins considered here are extrusion cooking, gel formation processes, as well as the newly emerging shear cell technology.[38] We first describe the general role of starch in foods and modification routes. After that, the interactions of starch and proteins are described. Then we show how starch is used as a functional ingredient when it is purposely added to meat products. Furthermore, we discuss the effect of exogenous starch on protein gelation and extrusion processing for meat replacing products. Next we lay out the known effects of endogenous starch on structure formation of protein gels. Lastly, we propose a new perspective on endogenous starch as a possible functional ingredient and introduce concepts to functionalize it.

2. Exogenous Starch

2.1. Modified Starch

Native starch is used widely in foods as a thickener or stabilizer. However, it also has less favorable properties, which are limited solubility in cold water, loss of viscosity and thickening power after cooking, high tendency to retrogradation, low shear resistance and thermal resistance.[39] Therefore, starch is often modified to tune its functional and physicochemical properties toward the food application.[39,40] A large variety of reviews and books are available on the modification and functionalization of starch.[9,39–44] Starch modifications can be on chemical and physical basis. Physical modifications are, for example, dry heating, agglomeration and granulation, multiple deep freezing and thawing, and pre-gelatinization. The latter can be achieved by drum drying, causing the pasting behavior of the starch to change: The starch swells and pastes at room temperature, while the viscosity at higher temperatures as well as the viscosity after cooling is reduced (**Figure 1**). These thermo-mechanical treatments applied to native starch result in so-called "functional native starches" that are not required to carry an E-number and are potentially considered "clean label."[9,45] Despite being created to increase consumer trust in food labels and manufacturing practices, E-numbers seem to do the opposite.[46–48] Therefore, physical modifications can be a powerful tool to functionalize starch for use in meat replacers and improve the product quality without decreasing consumer trust in the product. The food industry is well aware of these trends, evident by the patents filed in this field[49–51] and products on the market such as Ingredion's NOVATION product line[52] or Cargill's SimPure,[53] to name a few.

Chemical modifications can alter the properties of starch even further. Chemical modifications include hydrolysis, oxidation, esterification, etherification, cross-linking, hydroxypropylation, and acetylation. Hydrolysis can be achieved by enzymatic or acid hydrolysis leading to a breakup of the starch molecules into dextrin, maltose, and glucose. The main effect of (partial) hydrolysis is a strongly reduced viscosity when in solution. Esterification results in increased viscosity, due to the fact that the gelatinization temperature is lowered and the tendency to form a gel is reduced.[39] Esterified starch is used as an emulsion stabilizer and for encapsulation, and can also be used to partially replace fat in emulsion-based food products,[55,56] such as plant-based sausages. As a thickening agent in foods, etherified or cross-linked starch is used frequently. Both modifications lead to decreased solubility of starch, by adding inter- and intramolecular bonds, strengthening and stabilizing the starch polymers. Cross-linked starch is increasingly resistant to high and low temperatures and pH, however also come with smaller swelling volume.[40] Hydroxypropylation is a modification of starch based on etherification with propylene oxide in the presence of an alkaline catalyst. Hydroxypropylated starch improves freeze–thaw stability and decreases gelatinization and pasting temperatures.[57,58] In acetylated starch, hydroxyl groups are replaced with acetyl groups, increasing the viscosity as well as the solubility.[59]

2.2. Interaction of Starch and Protein

Interactions between starch and proteins are widely studied to understand the properties of products containing both components. The interaction of protein and starch can occur on multiple scales: associative interactions of the molecules,[60] steric hindrances,[61] competitive hydration[62] and phase behavior.[63] Heterotypic associative interactions of molecules, that is, interactions of molecules of different types, are thermodynamically less favorable than homotypic interactions, that is, interactions of molecules of the same type. However, it is still possible for heterotypic associative molecular interactions to occur, for example, at the interface of two liquid phases or within the phases themselves as they might contain multiple components. These interactions include the formation of ordered heterotypic junctions (similar to homotypic junction zones in single component polysaccharide gels), electrostatic attraction between negatively charged polysaccharides (polyanions) and proteins below their isoelectric point (polycations), and formation of Maillard reaction complexes between proteins and reducing sugars.[60] Interactions can also be more indirect in multiphase products. Especially when added in higher concentration, starch and protein might form separate phases with little molecular interaction, although interaction can occur through differences in water binding.[64] Therefore, the addition of starch can influence the macro- and microstructure of protein gels and products.

To create food products with starch and protein, powders containing plant-based protein and possibly starch are mixed with water, resulting in a wet biopolymer blend. Almost all biopolymer blends form a two-phase system under certain conditions.[60,63,65] At low concentration the dispersion can split into two co-existing phases after initial full mixing because of thermodynamic effects that drive phase separation. The latter leads to two phases that are enriched in one polymer and depleted in the other. The latter mostly occurs at low concentrations. For example, a carbohydrate concentration of 1% and a protein concentration of 5% are enough to form a two-phase system.[66] The concentrations used for meat replacers usually well exceed these values, with protein concentrations >30% and carbohydrate concentrations above 2%.[7,67,68] Thus, these materials likely form a multiphase blend with limited molecular interaction, but steric hindrance and competitive hydration influence gelling behavior of starch and protein reciprocally. For example, the onset temperature of starch gelatinization becomes higher upon the addition of protein,[62,69] while paste viscosity decreases.[69–71] Eliasson[72] researched the water migration during thermal processing of gluten and starch and calculated the amount of water associated with the gluten based on the enthalpy change of the starch gelatinization. Gluten forms a network already at ambient temperatures and thereby hinders starch gelatinization.[62] Li et al.[73] investigated thermal behavior of soy protein isolate and corn starch blends at 30–70% dm. They concluded that there was no significant chemical interaction between the protein and the starch, since the high concentration of the samples (50% dmc) favors a phase separated matrix. They further deduced that the starch restricted protein denaturation and protein restricted starch gelatinization indirectly.

2.3. Starch in Meat and Comminuted Meat Products

The use of starch in meat products has been extensively researched over the last decades.[74–76] It is used as a filler in comminuted meat products, like sausages or meat patties. Such fillers are non-meat ingredients, which help bind water and are usually good bulking agents. Starch is considered a good bulking agent and is added to bind water,[74] which would otherwise be exuded from the product. If added as pre-gelatinized starch, the ability to absorb water can even be increased further.[76] In its pre-gelatinized form, starch binds water at lower temperature, which is favorable as it prevents water loss upon heating of the meat.[74] The water binding properties of starch further enable the reduction of the caloric content of, for example, sausages.[77] Starch can retain the sensory and textural properties of products, for example. by increasing the firmness and strength of the product when animal fats are replaced by vegetable oils, improving the lipid profiles toward higher contents of unsaturated fats.[76] Furthermore, it was shown that the presence of starch in meat emulsions results in a more compact and stronger heat-induced protein matrix.[75,78] The reported advantages of employing starch in meat products are low cost and good control over functional properties such as cold swelling capacity, water solubility, and rheological properties through physical or chemical modification.[76] The examples in **Table 2** show that the applicability of starch in meat products has already been widely researched and might therefore provide understanding of the potential role of starch in meat replacing products.

2.4. Starch in Meat Replacers

As pointed out in Section 2.3, starch is often used in processed meat products for its water binding ability, its influence on

Table 2. Types of starch investigated for their functionality in application in meat and meat replacers in the literature.

Modification	Source	Variety	Added amount / % w.b.	Added to	Function in application	Meat/ Vegetarian/ Vegan	Ref.
None	Unknown	Resistant	4	Sausage	Replace or reduce fat	Meat	[79]
Extruded with wheat gluten	Corn		10–20	Beef burger	Reduce cooking loss, improve sensory attributes	Meat	[120]
Partially pre-gelatinized	Tapioca		0–3	Beef burger	Replace or reduce fat	Meat	[80]
None	Potato	Native	2.8	Poultry meat batter	Reduce cooking loss	Meat	[81]
Low cross-link and mid to high substitution modification	Potato	Native	2.8	Poultry meat batter	Reduce cooking loss	Meat	[81]
None	Tapioca	Native	2.8	Poultry meat batter	Reduce cooking loss	Meat	[81]
Low cross-link and mid to high substitution modification	Tapioca	Modified	2.8	Poultry meat batter	Reduce cooking loss	Meat	[81]
Stabilized	Waxy corn	Modified	1.5	Chicken rolls	Increase yield	Meat	[9]
Stabilized, cross-linked	Tapioca	Modified	1.5	Chicken rolls	Increase yield	eatM	[9]
None	Rice	Native	1.5	Chicken rolls	Increase yield	Meat	[9]
None	Waxy rice	Native	1.5	Chicken rolls	Increase yield	Meat	[9]
Unknown	Corn	Modified	3–7	Meat replacers	Bind water	Vegetarian	[82]
Pre-gelled	Potato	Modified	5	Meat replacers	Replace or reduce fat	Vegetarian	[82]
None	Wheat	Native	2.8	Extruded meat replacer (based on peanut protein)	Improve texture	Vegan	[92]
Cross-linked	Tapioca	Modified	1–7	Extruded meat replacer (based on soy protein)	Replace gluten	Vegan	[94]
Hydroxypropylated	Unknown	Modified	1–7	Extruded meat replacer (based on soy protein)	Replace gluten	Vegan	[94]

textural properties, or as a bulking agent. Therefore, one would expect potential to use starch in meat replacing products in a similar manner. Literature on the use of starch in meat and meat replacing products suggests that especially modified starch is used. As is evident from Table 1, starch is indeed added to commercial meat replacing products, with Table 2 showing that this is done most likely for similar reasons as it is done in meat products. Starch is successfully being used as an ingredient in meat replacing products to modify the sensory as well as the textural properties. This is due to the rheological properties of starch at different temperatures (Figure 1). When starch gelatinizes, viscosity increases drastically, followed by a drop in viscosity over time and shear. Upon cooling, the viscosity increases again, allowing starch to contribute to the, for example, the hardness of a product. Different starches have different pasting profiles, depending on their amylose and amylopectin content and the origin of the starch,[83] as well as any modification. Pre-gelatinized or "Cold Water Swelling Starches" increase the viscosity of a product already before heating, while simultaneously taking up more water. While the effects on sensory properties such as hardness, chewiness, and springiness can be partially explained by the rheological properties of different starches combined with the polymer blending law,[60,84] the knowledge about the effect of starch on structure formation is merely empirical. This is due to the fact that the exact mechanism of structure formation is not yet fully understood.[85] It seems that the effects depend on the origin of the starch, the moisture content and the other ingredients in the mixture used for processing. The influence of starch on structure formation could be based on indirect interactions,for example, by changing the water content of the protein through different water binding, leading to changes in the rheological properties of the phase responsible for structure formation. As starch also undergoes significant changes during thermal processing (swelling, gelling, degrading, setting), it could also cause a steric hindrance to the formation of a protein matrix that is able to form a fibrous structure. However, all the studies mentioned have an inductive approach, and the assumptions are not tested. Deductive studies on this matter do not exist yet. Furthermore, the rheological behavior of starch itself could influence the formation of fibrous structures. Several studies suggest that the rheological properties of the individual phases are crucial for fiber formation in the shear cell.[86–88] There is little knowledge about the rheological properties at conditions relevant for the shear cell or high moisture extrusion. Starch pasting curves such as in (Figure 1) are measured at lower temperatures, lower pressure, lower shear, and lower concentration. At higher temperatures, shear rates, and pressure, starches tend to disintegrate. In addition, starch generally exhibits shear-thinning behavior[89,90], which makes the prediction of rheological properties of starch even more difficult. Pasting curves can give an indication about the rheological behavior of the starch, but studies designed to elucidate the properties of starches under the relevant conditions are necessary.

2.4.1. Textural Properties

Textural properties such as hardness, chewiness, and springiness are of importance for meat replacers to mimic the eating experience of meat. There are studies that investigate the influence of starch on these textural properties of protein extrudates.[91–93] Zhang et al.[91] investigated the influence of 20% (d.b.) starch from different sources on the textural properties of extruded soy protein isolate/wheat gluten mixtures (50% moisture, d.b.). They report variations of the degree of fibrousness between the investigated starch sources, connecting the different thermal transition properties (including peak temperature, enthalpy changes) of the starch sources with the extrusion response parameters, such as specific mechanical energy (SME) and die pressure. They conclude that the thermal transition properties of the starch affected the textural properties of the extrudates by influencing the extrusion response parameters. Through fitting the physicochemical properties of extruded casein/wheat starch mixtures based on extrusion trials, it was found that the addition of 50% starch gave a maximum compression force of the extrudate when using 28% moisture and 194 °C in the extruder.[93] For all moisture contents and temperatures studied (126 °C $< T <$ 194 °C, 18% $<$ moisture $<$ 32%), wheat starch addition first increased the firmness of the product, while a further addition lowered the firmness again. The authors attribute this to the formation of bonds between the starch and the hydrophilic groups of the protein, made possible by the structural modification of both biopolymers caused by the high temperatures in the extruder. They claim that these bonds were responsible for the limited hydration capacity of the protein, a higher initial viscosity and the dense and rigid structures they observed after addition of starch. A study on SPI-based meat analogues, designed to find hydrocolloids suitable to replace gluten in the recipe, found that the addition of 1–4% hydroxypropyl starch and cross-linked starch decreased the hardness, gumminess, springiness, and cohesiveness of the extruded meat analogues.[94] The changes in textural properties were found to correlate well with the rheological data they obtained directly from the extruder. The starches were not found to be suitable replacers for gluten in the recipes, unlike blends of other hydrocolloids. Another study[95] also found that the addition of amylopectin to pea protein isolate decreased hardness, springiness, and chewiness of the extrudates, while amylose had the opposite effect, underlining that the effect of starch also depends on the composition and origin of the starch. This means that it is possible to adjust several textural properties of meat replacing products by adding the right amount of the right starch, depending on the desired outcome. Generalizing the effect of starch on textural properties is difficult, especially since the effect is indirect via the extrusion response parameters, as pointed out by Zhang et al.[91] The aforementioned polymer blending law can give indications of the effect of a starch on the textural properties if the rheological properties of the starch and the other ingredients at process conditions are known. Generally speaking, firmness can be increased or decreased by addition of amylose or amylopectin-rich starches, respectively. Modified starches could be able to deliver tailor made textural properties for meat replacing properties (Section 2.1). There are also a number of publications describing extrusion experiments that use starch as an ingredient, but do not specify the purpose or do not report on the influence of starch.[96,97]

2.4.2. Fiber Formation

When it comes to whole cut meat replacers, aimed to resemble whole cuts of meat, fibrousness and a meat-like structure are key factors for consumer acceptance of those products.[98–103] To create plant-based products with such a fibrous texture, some sort of processing has to be done, such as extrusion or shearing in a shear cell. In both processes, plant-based, protein-rich powders are combined with water, heat and shear are applied, and the material is cooled down.[38,104] In extrusion, the fibrousness is sometimes expressed as "degree of texturization," quantified as the ratio of the force needed to cut a sample lengthwise and crosswise of the direction of extrusion.[91,105,106] A similar measurand exists for samples prepared in the shear cell, called the anisotropic index.[38,68] Here, the ratio of the tensile strength parallel and perpendicular to the direction of the shear flow is used to quantify the fibrousness. Generally speaking, there is still a limited number of methods to evaluate the structure of meat replacing structures, especially when it comes to quantifying fibrousness.[107] A lack of standard procedure makes quantitative comparison of studies difficult.

The mechanism behind fiber formation in these materials is still debated in the literature.[68,85–87,96,103,108–116] Next to process parameters such as dry matter content, processing time, temperature, and pressure, the origin and composition of the ingredients are of utmost relevance as well. Many studies focus on protein–protein interactions and polymerization,[114,117,118] but also the influence of carbohydrates is investigated.[7] Dekkers et al.[119], for example, added pectin in a soy protein isolate dough to aid fiber formation. The effect of starch on fiber formation and textural properties of extruded protein has been investigated by ref. [120]. The study revealed that a fibrous texture can be achieved with blends of whey protein and starch at starch contents from 20% to 40%. They report that starch was also responsible for the increase of water holding they observed. Contrary to that, ref. [92] recently showed that the addition of native wheat starch had a negative effect on the fibrousness of peanut protein extrudates. Furthermore, the addition of starch lowered the chewiness and hardness of the extrudates. The outcomes were explained by considering that wheat starch promoted the aggregation of protein, supposedly "(...)breaking the intramolecular disulfide bonds, enhancing the hydrophobic interactions and increasing the apparent viscosity to stabilize the newly formed conformation."[92] Similar results were obtained for other polysaccharides in this study, but the effect of starch was found to be the highest. Other polysaccharides, such as sodium alginate, actually promoted the formation of fibrous structures.[92] A study on extrusion-based meat replacers made of potato protein, oat protein, wheat bran, potato dietary fibers, and potato starch showed that potato starch addition had no effect on the microstructure observed within different sections of the extruder when added at 25% to 50% (d.b.).[121] The shape of the protein aggregates was considered here as a measure for the deformation and therefore as an indicator for fiber formation.

It was found that potato starch did not affect the size of protein aggregates formed but did decrease the size of (dietary) potato fiber aggregates. The aforementioned recent study by ref. [95] investigated the influence of 10% (d.b.) amylose and amylopectin on high moisture extrusion of pea protein isolate. They conclude that amylopectin promotes formation of a fibrous structure (even though the difference in fibrous degree was not statistically significant), while amylose does not.

3. Endogenous Starch

Nowadays, interest in the use of ingredients derived from pulses and legumes in meat replacing products is increasing. Those pulses and legumes contain starch, part of which, depending on the method of extraction, remains in the fractionated protein ingredients. Generally speaking, the first steps in making protein-enriched ingredients from pulses and legumes are dehulling and subsequent milling. Since no starch and/or protein is lost during the milling step and no heating is applied, flours still contain high amounts of endogenous starch with native functionality. Further purification can be done either by more conventional methods (e.g., acid precipitation) to obtain highly purified protein isolates, or by applying novel fractionation processes such as air classification, which result in protein-rich fractions that only contain a limited amount of starch (e.g., less than 2%[122]). Berghout et al.[123] illustrate the trade-off between purity and yield and therefore resource efficiency for traditional purification methods. Applying novel fractionation processes, especially dry fractionation such as air classification, is less energy intensive due to the lack of drying steps and therefore also more resource efficient in regard to water use. Schutyser et al.[35] suggest the use of a combination of dry and aqueous fractionation to increase purity while maintaining some benefits of the dry fractionation. Tailoring fractionation routes to produce fractions with the desired functional properties (so-called functional fractions) for the intended application could be a key to increase the sustainability of meat replacers.[124] In order to determine the desired functional fractions for meat replacers, it is crucial to understand the effect of endogenous starch on structuring applications such as extrusion and shear cell structuring.

The use of endogenous starch in structuring processes for meat replacers has not been investigated, to the best of our knowledge. In case of protein isolates and concentrates, endogenous starch is often seen as a contaminant or unwanted component. Aguilera et al.,[125] for example, report that starch in peanut flour interfered with texture formation in low moisture extrusion. There are, however, multiple studies on endogenous starch in low moisture extrusion, usually applied for snacks and cereals, but also for production of texturized vegetable protein (TVP). Though they have a much lower moisture content (a very significant parameter in food structuring), these findings could still give indications for the use of endogenous starch in high moisture extrusion or the shear cell. Most studies on the role of endogenous starch focus on flours instead of protein concentrates or isolates.[125–132]

It is noteworthy that the use of endogenous starch and starch in general can also have negative aspects. On the one hand it poses process-related challenges, such as possibly increased Maillard reaction due to increased sugar contents in less-refined fractions or less consistency in ingredient quality and specification. On the other hand, it can have an effect on nutritional quality of the product, since less-refined fractions of, for example, faba bean that include endogenous starch also include the flatulence causing oligosaccharides raffinose and stachyose,[133] as well as other anti-nutritional factors (ANFs) that can limit protein and starch digestions.[134] Furthermore, when the two are present in their native form as starch granules and protein bodies, organized in a tightly packed matrix, protein can compromise the digestibility of starch[135] and vice versa.[136]

3.1. Endogenous Starch in Protein Gels

Pelgrom et al.[122] studied the gelatinization behavior of mildly refined fractions of yellow pea. They found that gel strength increased with higher starch concentration. Similar results were found for gels prepared of purified soy protein and wheat starch as well as from purified lentil protein and lentil starch.[69,137] Furthermore, Pelgrom et al.[122] show that gels prepared from purified starch had higher gel strength compared to those prepared from dry processed fractions for a given starch concentration. This was explained by the presence of protein and fiber that weaken the starch network by forming dispersed domains.

3.2. Endogenous Starch in Extrusion

Several studies report on extrusion of pulse and legume flours, with most of them focusing on low moisture extrusion.[125–132] In general, presence of starch in the raw material contributes to a higher pasting viscosity as a result of its amorphous gel phase.[127] However, extrusion barrel temperatures and SME levels highly influence the behavior of starch in a dense, protein-rich blend. The effect is difficult to predict. On the one hand, high temperatures or SME levels can increase starch degradation, thus reducing the melt viscosity in the extruder barrel. On the other hand, it was found that increased extrusion temperature led to increased cooking of starch and therefore a better expansion of whole pinto bean meal extrudate.[126]

Jebalia et al.[128] compared the morphology and mechanical behavior of extruded pea flour to those of extruded mixtures of pea starch and pea protein isolates. Both products consisted mostly of amorphous starch after extrusion. They found that for samples processed with low SME, starch domains were surrounded by a protein matrix. At higher SME levels, starch formed a continuous matrix around protein aggregates, which is likely related to the higher percentage of starch in comparison to protein. Furthermore, they report a difference in size of the protein aggregates, with the pea flour exhibiting smaller and more dispersed protein domains than the pea protein isolate—starch mixtures with less dispersed but larger protein domains. Additionally, the protein domains of the pea protein isolate—starch mixtures were elongated in the direction of the extrusion flow. Similar morphologies were observed by Kristiawan et al.[129] for low moisture extrusion of pea flour at low SME levels. It was further shown that at increased SME levels the extruded material formed a bi-continuous network of protein aggregates and starch molecules.[129,138] They suggest that different states, sizes, and continuities of protein aggregates can lead to various protein–starch morphologies.

Orientation of dispersed phase domains in the direction of shear flow was previously found to be an important parameter for fiber formation in shear cell structuring.[68,139] As mentioned earlier, protein domains in extruded starch–protein blends were found to be oriented in the flow direction, while protein domains in extruded pea flour composites showed no orientation.[128] If we assume that elongation of the dispersed phase is a prerequisite for the formation of fibers with the shear cell technology or extrusion, it is expected that it is not possible to transform untreated pea flour into fibrous materials successfully. The authors further describe that extrudates from pea flour were more brittle than extrudates from starch–protein blends. This is explained by the fact that pea flour extrudates had smaller protein domains and therefore a larger interfacial area. As protein and starch domains have limited interaction, an increased interfacial area leads to easier breaking. Kristiawan et al.[129] suggest that a higher interfacial area favors bubble nucleation and therewith expansion of the extrudates when producing snacks from pulses. This would imply that pea flour extrudates would have a higher expansion rate than starch–protein blend extrudates. In low moisture extrusion of faba bean flour, bubble nucleation was found to be promoted by the formation of linkages between starch molecules, resulting in a larger expansion than reference products.[127] From previous research we know that air bubbles can enhance fibrousness in plant protein blends structured by shear cell technology.[87,139] Addition of starch to these blends might lead to increased air bubble nucleation and thus a more defined fibrous structure. However, one must keep in mind that air is introduced through expansion in extrusion, whereas in shear cell technology it is introduced as a foam. As described above, the structuring properties of a flour is often not identical to that of a blend of protein isolate and starch with similar overall composition. Here, the processing needed to make pure starch and protein isolate probably explains the differences in structuring properties. Protein isolate and starch have undergone wet purification steps, acid precipitation and subsequent spray drying, which alters the functional properties of the components.[35,140,141] The difference in functionality is expected to influence the fiber formation potential. This finding also indicates that we need alternative routes to translate results from exogenous starch systems to endogenous starch-containing materials.

3.3. Wheat Gluten as Endogenous Starch Source

Wheat gluten is often used to make fibrous, meat-like structures. Especially in shear cell technology, functional properties of wheat gluten have been shown to be very important.[38,85,87,113] Starch is often overlooked as a remaining component in protein ingredients. One of these protein ingredients is wheat gluten. According to suppliers there is ≈10% starch still present in the wheat gluten isolate.[142] However, studies on structuring of wheat gluten usually do not comment on the presence or effect of starch. As we know from exogenous starch, the addition of a small amount can affect textural and viscoelastic properties. Therefore, it is highly likely that the starch present in wheat gluten also affects textural and viscoelastic properties of the full system.

4. New Approaches

As described in the previous sections, starch is added to meat and meat replacing products to increase product yield, bind water, and modify texture and structure. At the same time, meat replacing products are often based on highly refined plant proteins, which have been depleted in all other components, including starch. The pure components are eventually modified individually and recombined to make a product with the desired properties. To lower the environmental impact of meat replacers, it would be better to use less-refined plant protein ingredients, while maintaining quality of the end product.[35,36,124] A consequence of less refinement is higher inclusion levels of other components, including starch. This might pose problems, but also creates opportunities as well, due to the richer composition. As pointed out, starch is used in meat replacing products already. The main issue is that the presence of native starch does not generally improve the structuring properties. It explains why mostly modified starch is used in meat and meat replacing products.

Up to now, modifications of starch are mainly performed for purified starch. To maintain the product quality when moving from products made from mixtures consisting of highly purified ingredients to those made from less-refined ingredients, new pathways to functionalize the components inside the ingredients have to be developed. These new pathways could utilize similar treatments as are currently used for purified starch or protein, only applied to more complex, multi-component ingredients. When applying these pathways to mild-fractionated starch or protein-enriched products, these modifications will not only influence the functional properties and molecular structure of the starch, but also those of the other components present. This might especially be influential for proteins present in a starch or protein concentrate. This again could be utilized, as proteins retain their native functionality in mildly refined fractions.[122] The isolates applied in industry exhibit vastly different functional properties due to the conditions during the isolation process, and are thus less prone to fine-tuning to a certain application,[35,140,141] as explained in Section 3.2.

The physical treatments used to functionalize starch isolates, such as freezing and thawing, drum drying, annealing, and agglomeration are controlled by temperature, moisture content and pressure. The same parameters can potentially be used to control pre-treatments of less-refined ingredients and even target single components within one ingredient. For this, the behavior and interaction of the components needs to be understood, which can be achieved when studying those components in the multi-component ingredient. **Figure 2** illustrates this by showing a thermogram of an air classified faba bean fraction containing mainly starch and protein. Depending on the moisture content of the material, the temperature at which the changes occur, such as starch gelatinization or protein denaturation, can be controlled and even separated. The latter is possible, because the denaturation temperature of proteins depends on the moisture content over a wide range,[143,144] while the temperature of the initial starch gelatinization is less dependent on moisture content; only the degree of the initial gelatinization changes with moisture content.[145–147] The presence of other components can also influence the available moisture of the individual components in

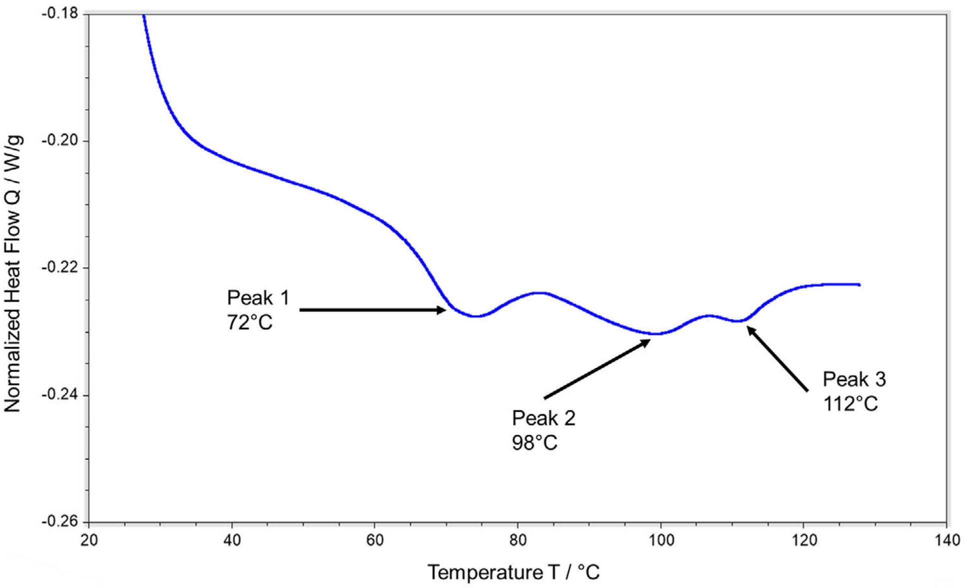

Figure 2. DSC curve of starch-rich faba bean fraction (air classified), 45% dm, 5 °C min^{-1} heating rate. Peak 1 represents the initial gelatinization of starch, Peak 2 represents the denaturation of vicilin, and Peak 3 represents the denaturation of legumin.

such a multi-component ingredient. Therefore, the distribution of moisture among all components also has to be understood.[148]

So far, only a limited number of studies on functionalizing less-refined ingredients exist, such as moisture heating of aqueous fractionated soy[149,150] to increase viscosity and lower solubility of the protein, or dry heat treatment of air classified faba bean concentrate to increase water holding.[151] Geerts et al.[149] show that thermal treatment is a route to improve structuring properties of a soy concentrate. Results with faba bean concentrate suggest similar improvement. Apart from physical modifications, also chemical or enzymatic modifications could be applied to less-refined ingredients. Hydrolysis for example, which is often applied to modify starch, leads to cleavage of proteins into smaller peptides and thereby gradually alters their functionality in terms of, that is, water holding capacity and gelling properties. Nieto-Nieto et al.[152] investigated the gelling properties of partially hydrolyzed oat protein and show that the formed gels exhibit similar mechanical strength and water holding capacity as gels produced from egg white. However, peptic hydrolysis of soy protein isolates resulted in poor gelling properties.[153]

We therefore conclude that mild-fractionated ingredients have great potential in terms of sustainability and, in combination with tailored pre-treatments, interesting functionality. The challenges along the way, such as the interaction of components, the distribution of water or the lack of functionality can be solved by investigating the ingredients as the multi-component mixtures that they are.

5. Conclusion

Starch takes different roles in meat and meat replacing products. It acts as a functional ingredient or filler that is added to the food product to improve textural properties or to bind water. Simultaneously, it alters the gelling properties of proteins and often decrease fiber formation in extrusion or the shear cell. The effect of starch depends on the properties of the different types of starch, ranging from particle size, water holding capacity, and solubility to thermal properties such as pasting profile and viscosity. Furthermore, starch can interact with protein on a molecular level and on a macroscopic level through phase interaction. All those properties can differ depending on the source of protein and starch, the concentration used, and the processing history of those components.

The literature on the effect of endogenous starch on protein gels and meat replacing products is rather limited. Besides, the findings from studies of exogenous starch are not directly transferable, as the functional properties of starch and protein are altered in the separation and other processing steps. It implies that studies on model systems cannot directly be translated to the properties of multi-component ingredients obtained via mild fractionation. It is therefore necessary to further explore the structuring potential of materials that contain endogenous starch, such as concentrates and flours from legumes. To increase the functionality of endogenous starch, modification treatments commonly used to improve properties of starch isolates such as pre-gelatinization or potentially chemical modifications could be applied to the less-refined raw materials as well. Thermal pre-treatments could be a promising way to functionalize less-refined raw materials, as there are potential synergies with the functionalization of protein in the same materials. Great progress can be made in terms of quality, sustainability, and cost-efficiency of meat replacers by designing fractionation processes for protein- and starch-rich crops specifically for meat replacers and combining them with the aforementioned treatments to tune the functional properties of the fractions.

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Conflict of Interest

The authors declare no conflict of interest.

Data Availability Statement

Data available on request from the authors.

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extrusion, food structuring, meat analogue, meat replacer, mild fractionation, shear cell, starch modification

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