

# Report on Minnesota Plant Based Proteins for Food

A REPORT PREPARED FOR THE AGRICULTURAL UTILIZATION  
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## **Forward**

The mission of the Agricultural Utilization Research (AURI) Institute is to foster long-term economic benefit for Minnesota through value-added agricultural products. In pursuing its mission, AURI conducts a biennial stakeholder analysis to determine the needs and challenges faced by various organizations and businesses within the value-added agricultural sector.

A recent stakeholder analysis revealed the food industry desired more information about plant protein ingredients and their potential for utilization. Because this need aligns with AURI's efforts to increase the utilization of Minnesota's commodities, a study exploring potential plant protein ingredient applications was conducted.

The study that follows provides a user-friendly overview of the drivers of increased demand, characteristics of various protein ingredients, extraction processes and an overview of both currently available and potentially viable sources of plant protein ingredients.

For more information on plant protein sources, benefits and uses, contact AURI at 218.281.7600.

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## Introduction

The global demand for protein ingredients is expected to grow from a value of \$25.62 billion in 2016 to \$48.77 billion by 2025 (Grand View Research, 2017). Protein ingredients have gained prominence in the food industry due to the escalating consumer awareness and demand for healthy foods. In general, consumers want more protein in their diet. A 2014 New Product Development (NPD) group study showed that protein content is the number one item consumers look for on a nutrition label, with greater than fifty percent wanting more protein in their diet (NPD, 2014). Consumers are also using protein content as a criteria in choosing new products. This seemingly insatiable consumer demand for protein is driven by negative consumer perception towards fats and sugar while proteins are viewed as a key part of a healthy diet due to well-known benefits such as promoting satiety, building lean muscle mass, and improving glycemic control (Paddon-Jones et al., 2008).

There is a growing interest in plant-based protein ingredients. Several reasons have led to this interest. Among them is the cost of traditional protein ingredients. Plant proteins can offset market share from animal proteins (dairy, egg, and meat) because they are producible at competitive prices. Other reasons for the interest in novel plant proteins include the rising incidence of allergenicity (dairy and soy proteins are among the “big 8” allergens) and several functionality limitations. Increases in vegan and health conscious consumers, in addition to existing number of consumers seeking halal and kosher foods, are another driver of plant proteins’ popularity.

Other opportunistic reasons include utilizing current processing streams to increase value and revenue (valorizing by-products), finding a unique and a competitive place in the market, replacing chemical ingredients with functional proteins (clean label), and utilizing all possible resources to expand the ingredients supply. Additionally, there is a growing interest in sustainable and environmentally friendly sources. Some may also argue that the food industry reached a maturation stage, and there is a pressing need for innovation. Thus, demonstration of equivalent or superior/new functions of novel plant proteins compared to existing alternatives is essential to their market success. However, there is limited consumer and producer knowledge of plant proteins other than soy; nevertheless, novel plant proteins such as proteins from pulses, potato, rice, corn, oats, canola, and ancient grains are gaining traction (Grand View Research, 2016).

Novel plant proteins are emerging, with some utilized in the marketplace; meanwhile food producers and entrepreneurs seek information on the nutritional, physiological and functional characteristics of these proteins. There is a need to understand the potential of these novel proteins to replace traditional protein ingredients (partially or wholly) in various food products to deliver optimal nutrition and functionality. While there has been some research done to characterize novel plant proteins, available information is far from comprehensive.

This report will summarize current knowledge, advantages, barriers, and areas requiring further investigation. Additionally, the intent of this report is to provide basic information that helps Minnesota entrepreneurs explore the potential of utilizing various regional plant protein sources in various food applications to address the growing market demand for such products.

The report that follows contains information on plant protein sources that are currently available, emerging, or potentially viable sources of proteins. For the purpose of this report, the definition of currently available protein sources is “commercially produced”; emerging protein sources are researched and likely to be on the market in the near future; potentially viable protein sources are those with minimal research, but show promise. This report provides information about nutritional quality, currently available ingredient forms, functionality and applications, advantages, and barriers to assess the feasibility of its production and utilization for each protein source. The knowledge summarized will benefit companies and entrepreneurs in Minnesota aiming to determine their best approach for entering the plant protein ingredients and products marketplace.

## Why the Exponential Increase in Demand for High Protein Foods

Due to their physiological, nutritional and functional contributions, many food applications use protein ingredients. Protein ingredients have gained prominence in the food industry due to the escalating consumer awareness and demand for healthy foods. In general, consumers want more protein in their diet. Screening new products, including ready meals, cereals, protein beverages, and snacks, on supermarket shelves reveals an ubiquity of “High in Protein,” “Protein Added,” and “More Protein” claims. Sports nutrition for example had 11-17% increase in protein-based new launches in the past year. A 2014 New Product Development (NPD) group study showed that protein content is the number one item consumers look for on a nutrition label, with greater than fifty percent wanting more protein in their diet (NPD, 2014). Consumers are also using protein content as a criteria in choosing new products: 70% of consumers consider products higher in protein as an influencing factor for trying different food and beverage brands (Innova, 2016). This seemingly insatiable consumer demand for protein is driven by negative consumer perception towards fats and sugar while proteins are viewed as a key part of a healthy diet due to well-known benefits such as promoting satiety, building lean muscle mass, and improving glycemic control (Paddon-Jones et al., 2008). In conjunction with an increasing number of consumers seeking plant based protein options, there is a growing group of “flexitarians.” These are consumers looking for lower calorie, lower fat, less expensive protein sources (NPD, 2014). In 2015, approximately 120 million Americans (38%) ate meatless diets at least one day a week (Innova, 2016). Consequently, this has led consumers to seek out affordable plant protein sources with established health benefits.

Naturally, industry is quick to respond to consumers’ demands; hence, the exponential increase in the need for finding and using novel plant protein in various food and beverage products. With the industry responding to consumers’ demands, the marketing of protein-based ingredients and associated claims is resulting in a further increase in consumers’ demand for a protein rich diet. Science and technology, however, has yet to catch up with this exponential increase in the demand for novel plant protein.

Soy protein is the most researched and widely used protein ingredient. Long strides in soy protein production and ingredient functionalization have been made over the past few decades. A lot is known about the functionality, nutrition, and health benefits of soy protein (Friedman and Brandon 2001; Fukushima 2011; Mojica and others 2015). However, soy protein is one of the U.S. Food and Drug Administration’s (FDA) “big 8” allergens, and the planted

soybean acreage is predominantly genetically modified varieties. Consumers are thus seeking other plant protein sources, and the industry has to adapt.

Furthermore, consumers are calling for clean labels (i.e. replacing functional and synthetic ingredients with natural ones). Given that proteins have multiple functions including, but not limited to, stabilizing properties, structure building, and flavor enhancement, producers are seeking to replace synthetic ingredients with functional proteins in various applications including high value ones such as encapsulation of bioactives and flavors (e.g. fish oil and orange oil).

Sustainability, transparency and food security are additional factors driving increased demand for plant proteins. Several companies, from small to large, have introduced or are exploring alternative protein ingredients, including plant proteins, to meet the consumer's desire to lessen their environmental footprint.

## Key Characteristics of Protein Ingredients

There are four categories by which a protein ingredient gets evaluated for food use: nutritional, functional, organoleptic, and claim/labelling. The nutritional properties of a protein is a function of the protein quality in terms of essential amino acids content and digestibility (bioaccessibility of the amino acids needed for metabolism and growth), and of physiological benefits such as promoting satiety and repairing muscle mass. Protein digestibility corrected amino acid score (PDCAAS) is a measure of the protein quality, and is used to calculate % daily value (PDCAAS x protein content per serving/50 g) reported on the nutrition label. The highest PDCAAS score is 1. Soy protein for example is a complete protein and has a PDCAAS score of .96. But, if a protein has a PDCAAS of zero, regardless of the content of this protein in a serving, it will contribute to 0% Dietary Value (DV). It is worth noting that processing will impact protein digestibility, thus affecting the PDCAAS. In some cases processing results in enhanced digestibility, while in other cases processing results in protein polymerization and thus reduced digestibility. It is important to take this into account when evaluating PDCAAS. Confirmation of physiological benefits of a protein follow a series of *in vivo* studies. For example, in 1999, the Food and Drug Administration (FDA) approved a heart health claim in response to significant scientific evidence that suggests "diets low in saturated fat and cholesterol that include 25 grams of soy protein a day may reduce the risk of heart disease" (21 CFR § 101.82, e-CFR 2017). The 2016 consumer attitude study by the United Soybean Board found 43% of consumers were aware of the FDA Heart Health claim (United Soybean Board, 2016). In conjunction with this increase in consumer awareness, there has been a 36% total growth of heart health claims on packages of soy protein containing products, in the past five years (Innova, 2016).

Often, the use of protein ingredients is for their functional properties, not only their nutritional benefits. For food processors, protein ingredients are value-added ingredients imparting viscosity, gelation, water absorption and binding, emulsification, foaming, and flavor-binding functions to complex food systems (Fukushima, 2011). Protein solubility and powder dispersibility are also key functional properties that impact all other functionalities. Depending on the application, one or more functional properties are desirable over the other. For example, in a beverage application protein solubility and thermal stability are desirable, while protein gelation over storage is undesirable. On the other hand, protein gelation in products

such as tofu, yogurt or plant based sausages, is desirable. Different proteins differ in functionality due to differences in composition and structure. Thus, understanding the structure/function relationship in various proteins is essential for the selection and use of protein ingredients.

Organoleptic properties of proteins also affect their selection for various applications. Plant proteins, specifically, are often associated with off notes, such as beany, hay, cardboard aroma, green flavor, and bitterness. Ways to mitigate off notes include application of various processing approaches (e.g. dehulling, enzyme deactivation, membrane processing and flavor extraction) and/or masking by formulation. Different plant protein sources have unique flavors requiring targeted approaches to eliminate or mask them.

Nutrition labeling rules mandate the reporting of the presence of allergens and processes used to modify the protein (e.g. hydrolysis), while mandatory labeling of biotechnology-derived products is forthcoming with the passage of recent legislation. Often consumers are looking for allergen free, non-genetically-modified (GENETICALLY MODIFIED), organic, non-hydrolyzed ingredients (just because they don't know what this process is) and clean label food products. Accordingly, food processors consider these characteristics in the selection and search for novel plant protein ingredients.

## Common Plant Protein Extraction and Purification Processes

Oil extraction often precedes plant protein extraction and purification, as is the case for oilseeds (e.g. soybean), air classification to separate starch granules from protein bodies, as is the case for beans, or steeping as in the corn milling process, which separates the corn into its four components, germ, fiber, starch and protein. Cleaning and initial concentration steps are crop dependent. Following initial separation, the fraction rich in protein is further processed to produce a protein concentrate (60-80% protein) or isolate (90% protein or greater).

### *Common Purification Practices*

Protein concentration and purification is achievable following membrane filtration, chromatography, pH solubilization/precipitation and salt extraction. For commercially available plant protein ingredients, namely soy and pea protein, the most common practices are pH solubilization/precipitation and salt extraction, as the other two processes, though, may produce a protein ingredient that is more functional, are more involved and costly. The protein in an initial concentrate can be solubilized at a pH where the protein is most soluble, while the starch and/or fiber will precipitate post centrifugation.

To separate the protein from soluble sugars and oligosaccharides, the protein becomes a precipitate at its isoelectric point. The next steps are washing, neutralizing and spray drying the precipitate. Sometimes there is an introduction of a diafiltration step prior to drying to reduce the amount of salt. The pH of solubilization may affect functionality, color, flavor, and digestibility. On the other hand, one can purify the protein in an initial concentrate

using salt extraction. The protein solubilizes at a specific pH and salt concentration, to separate it from starch and/or fiber.

To separate the protein from soluble sugars and oligosaccharides, the process uses a relatively high salt concentration to salt out the protein. The precipitated protein goes through a washing and diafiltration to remove excess salt. The protein from any given source is a heterogeneous mixture of different types of proteins. Therefore, purifying the protein following different methods will result in different protein profile, quality and functionality.

### *Powder Functionalization*

Often protein powders are subjected to several functionalization processes including agglomeration, lecithin coating, and high-pressure homogenization (Barbosa-Cánovas and others 2005). These processes affect particle size, shape and surface properties. Agglomeration increases particle size by forming bridges using binders, such as starch, gums, or hydrocolloids. This process enhances dispersion as water can diffuse easily within the agglomerate, while lecithin coating enhances wettability and prevents powder caking.

High-pressure homogenization coupled with controlled spray drying conditions impact protein functionality. For example, high-pressure processing results in increased water holding capacity and viscosity, desirable for meat-like applications.

It's possible to manipulate the powder functionalization through processing for targeted functionality enhancement. Different protein sources, however, may require unique processing approaches to enhance their functionality. A lot is known about soy and dairy protein functionalization. However, functionalization is an area that requires investigation for novel plant proteins.

### *Protein Modifications*

The use of proteins in food formulations is subject to processing challenges due to their sensitivity to various processing parameters including pH, temperature, shear stress, and enzymatic activity. Methods to improve protein functionality and stability during processing commonly focus on modifying the protein structure to improve solubility, increase flexibility, alter the hydrophilic/lipophilic balance, or promote protein crosslinking. Commonly reported protein modification techniques include chemical modification, limited enzymatic hydrolysis, and Maillard-induced glycation.

Chemical modification by phosphorylation, acylation, and alkylation were among the initial approaches to simultaneously improving functionality, such as solubility, water holding capacity, and emulsification abilities, while solving processing challenges. Such modification involves chemical changes to the amino acid side chains that may alter the protein's net



charge and/or hydrophobicity through reactant choice. (Sung and others 1983; Damodaran 2008; Kinsella 1979; Matemu and others 2011).

However, these methods face concerns over the use of harmful reactants like sodium cyanoborohydride (acid contact liberates toxic gas, water contact liberates highly flammable gas), decreased nutritional quality due to blockage of essential amino acids, formation of toxic amino acid derivatives (e.g. carcinogen thiourea formed during alkylation with isothiocyanate), and regulatory concerns (Baslé and others 2010; Damodaran 2008). These regulatory concerns have led the United States (USA) and Europe's food regulatory bodies to ban chemical modification of proteins, thus shifting research to non-chemical approaches for improving protein functionality.

Enzymatic hydrolysis is the most researched and most common approach used in the industry for protein modification intended to improve functionality and physiological benefits. Degree of hydrolysis (%DH) and enzyme choice dictate the functional properties of the produced protein hydrolysate by influencing protein structure and peptide profile. A limited extent of hydrolysis (i.e. low % DH) is particularly important for producing functionally enhanced ingredients, because it controls for both the loss in structure and release of bitter peptides associated with more extensive hydrolysis. Excessive hydrolysis (i.e. high % DH) results in a product high in free amino acids and short chain peptides with minimal if any functionality. Limited enzymatic hydrolysis of soy protein (DH = 2 – 15%), for instance, resulted in increased solubility (Jung and others 2005; Meinlschmidt and others 2016; Sun 2011), foaming (Tsumura and others 2005), and emulsifying ability (Meinlschmidt and others 2016; Sun 2011).

There is some research on the impact of limited, controlled Maillard-induced glycation on improving protein functionality but it has not been commercially applied. A 2016 review highlighted 31 studies showing improved functionality for glycated proteins (de Oliveira and others 2016). Maillard-induced glycation may result in improved solubility, thermal stability, emulsification, foaming, and gelation properties due to increased hydrophilicity, viscosity, and protein cross-linking, while lowering the protein's isoelectric point and preventing denaturation (de Oliveira and others 2016; Wang and Ismail, 2012; Wang and others 2013). However, the structural modifications and functional changes of glycated proteins depend on the Maillard reaction conditions, protein conformation, and polysaccharide characteristics (e.g. chain length). Therefore, optimization of Maillard-induced glycation parameters is required to achieve the desired functionality of a particular protein, while minimizing the propagation of the reaction to advanced and undesired stages (leading to browning and off flavors).

## Currently Available and Emerging Plant Protein Ingredients and their Uses

Extensive research on several plant-based proteins lead to their commercial availability in the United States. While soy protein is the most commercially used plant protein, pea

protein is gaining commercial popularity, and canola protein is emerging and will soon be commercially available in the market.

### *Soy Protein Ingredients*

#### *Recent statistics on soybean yields/sales in Minnesota*

Estimates of Minnesota's soybean yields in 2016 were 52.5 BU/acre and total sales were around \$3.6 billion (USDA 2017); Minnesota's average price for soybeans in 2016 was \$9.25/bushel, slightly below the national average of \$9.50/bushel (Crop Values 2017). Apart from production of oil, significant utilization of soybean for the production of soy protein ingredients and products is another option. The lower per bushel rates can benefit Minnesota soy protein ingredient manufacturers by reducing soy protein production costs. Additionally, soybeans are eligible for price loss coverage under the 2014 Farm Bill, offsetting some of the monetary risks for farmers related to control of weeds and disease (2014 Farm Act 2017).

#### *Nutritional quality*

Soybeans are generally comprised of over 40% protein, making them more protein-rich than other legumes. This, in part, makes soy protein a desirable alternative to milk and meat proteins. The PDCAAS score for soy protein ranges from 0.9 – 1.0 (max PDCAAS score is 1.0) depending on processing conditions (Young, 1991). Having this high PDCAAS score means sole consumption of soy protein at a level of 0.6 g protein/kg body weight is sufficient to meet the protein needs of children and adults; hence, classifying soy protein as a complete protein source (National Kidney Foundation, 2002; Young, 1991). For infants, modest supplementation of soy protein with methionine, an essential amino acid, is suggested because the methionine content of soy protein can be limiting for this population (Young, 1991).

As previously mentioned, the 1999 FDA approval of the soy protein heart health claim was the initial force driving the increased commercial use of soy protein. This claim was supported by consistent findings that consumption of soy protein, instead of animal protein, significantly lowered serum levels of total cholesterol, LDL cholesterol, and triglycerides (Anderson and others 1995). Soy protein benefits extend to satiety and weight management. Animal studies showed that soy protein consumption improves insulin resistance, a cornerstone of human obesity, and reduces body fat accumulation (Velasquez and Bhathena 2007). For example, studies with genetically obese rats have consistently shown that the consumption of soy protein, in comparison to casein, leads to decreased body mass and fat, decreased plasma and liver triglycerides, and decreased plasma glucose and glucose resistance (Aoyama and others 2000; Iritani and others 1997; Iritani and others 1996; Velasquez and Bhathena, 2007). Similarly, clinical studies with obese individuals showed a reduction in body mass and improved insulin resistance in addition to the lowered triglyceride levels (Velasquez and Bhathena, 2007).

### *Soy protein components*

Soy protein is mainly comprised of globular proteins (90%), with the remaining being albumin proteins (10%). Globular and albumin proteins differ in their solubilities, with the globulins soluble in dilute salt solutions and the albumins in water (Damodaran 2008). The two globulin proteins, glycinin (~ 40 – 50%) and  $\beta$ -conglycinin (~ 25 - 30%), are the two main soy storage proteins (Liu 1997; Mojica and others 2015), comprising 65 - 80% of the total seed protein. As the two main proteins, they influence the functionality of soy protein ingredients. The individual protein composition of a soy protein ingredient and the fractions derived from it depends, however, on the compatibility of a protein's solubility in the extraction solvent used during the extraction and purification process.

### *Currently available protein ingredient forms*

There are three basic types of soy protein ingredients: soy flour (50 – 60% protein, dry basis), soy protein concentrate (SPC: 65 – 80% protein), and soy protein isolate (SPI: > 90% protein). To achieve desired biological or physicochemical functions, soy protein isolate is commonly subjected to enzymatic hydrolysis to produce soy protein hydrolysates. Extruding, steam injecting, or jet-cooking soy flour, SPC or SPI generates texturized soy protein. For the production of soy flour, cleaning, drying, dehulling, conditioning, and then flaking is necessary. The soy flakes can then be milled to produce soy grits and/or soy flour depending on final particle size (Riaz 2004). Soybeans typically contain about 18 – 22% oil; therefore, it is common to hexane extract the oil from soy flakes before milling to produce a defatted soy flour (Riaz 2004). Defatted soy flour (approx. 52 – 54% protein) is typically further processed into SPC and SPI to increase protein content, reduce the “beany” flavor, and improve digestibility (Riaz 2004; Singh and others 2008). Three methods are commonly used to reduce the content of ash, oligosaccharides, and other minor constituents, found in soy flour, and thus produce SPC (~80% protein). Known for producing the blandest flavor, the first method precipitates the proteins and polysaccharides with a 60 – 80% aqueous alcohol wash (Riaz 2004; Singh and others 2008). The second method acidifies soy flour to a pH of about 4.5 to insolubilize the major globulins and polysaccharides while leaching the sugars (Singh and others 2008). An advantage of this method is an improved protein dispersibility index compared to SPC produced using aqueous alcohol (Riaz 2004). Finally, moist heat can be applied to denature and insolubilize the protein allowing for the removal of sugars and other minor components with a water wash (Singh and others 2008). This process however, results in a concentrate with lower functionality compared to the other two processes. To produce SPI (>90% protein) commercially, the most common method is pH solubilization/precipitation described earlier.

### *Potential functionality and applications*

Soy protein ingredients are elements of several food applications for their viscosity, gelation, water absorption and binding, emulsification, foaming, and flavor-binding

properties. For example, soy protein is added to comminuted meats (e.g. hot dog batter) to improve gelation, and to coffee creamers to assist with emulsification and foaming (Fukushima 2011). When used in comminuted meat products, the emulsifying abilities of soy proteins reduces cook loss, improves texture and juiciness (Jideani 2011). The different functional properties of soy proteins are dependent on its solubility, so processing conditions impact their effectiveness (Jideani 2011), which in turn affects the solubility. If soy proteins are texturized they can be used as meat replacements, extenders, or texturizers. In meat-less products, they can mimic the texture of meat, and in meat products, they can improve texture (Jideani 2011; Thrane and others 2017). Adding texturized soy protein to comminuted meat products can reduce production costs by increasing water-retention and replacing up to 30% of the expensive animal-based protein with less expensive soy protein without affecting eating quality (Jideani 2011; Thrane and others 2017). The nutritional quality and diverse functionality of soy protein makes it useful in a variety of foods, including salad dressings, frozen desserts, pastas, breakfast cereals, baked products, meat and meat-like products, beverages, and protein bars (Thrane and others 2017).

### *Advantages*

Soy protein has several agricultural, nutritional, functional, and technological advantages. A major agricultural advantage is that soybeans require less fertile soil for growth than other crops like corn, making their production possible in inhospitable regions (Coulter and others 2010). Soybeans can also fix nitrogen in the soil, reducing fertilizer needs and benefitting subsequent crops in a crop rotation (Coulter and others 2010; Thrane and others 2017). In terms of nutritional advantages, soy protein is a high quality, complete protein, making it suitable for those on vegan and high protein diets (Thrane and others 2017), and is also associated with several physiological benefits as discussed in a previous section. Advertising these nutritional advantages could attract consumers and increase the popularity and sales of products containing soy protein. Finally, there were technological advances made over the past few decades for the production of functional soy protein ingredients for various applications. Moreover, plant breeders are currently developing soybean varieties with low to zero allergens and reduce beany flavors.

### *Barriers*

In soy protein ingredients, beany/grassy and bitter/astringent off-flavors can be problematic (Murphy 2008; Asgar and others 2010). As some of the off flavors are related to lipid oxidation, a reduction in off flavors has been achieved when using soybeans bred with little or no lipoxygenase, or by following effective extraction methods to remove excess oil (Thrane and others 2017). The two main issues, however, are consumer acceptance of biotech-derived crops and soy protein allergenicity (one of the “big 8” allergens). In 2016, 94% of the total US acreage used for soybean production grew herbicide tolerant soybeans (USDA ERS 2017b). As many consumers have yet to understand and accept genetically modified (GM) crops, companies may be reluctant to

incorporate soybean ingredients into their products. Non-GM soybeans, or identity preserved (IP) soybeans, are available, but the vast majority of soybeans are modified to be herbicide tolerant (Thrane and others 2017). Additionally, the market for IP soybeans requires several stringent contractual processes, such as the use of certified non-GM seeds and to have documentation demonstrating their adherence to IP procedures (Thrane and others 2017). In regards to allergenicity, thermal and enzymatic processing can reduce the allergenicity of soy protein, but there is currently no method completely eliminating it (Thrane and others 2017).

### *Feasibility*

Good yields and high total sales demonstrate that Minnesota is well suited for soybean production. In most areas of Minnesota, the soil contains enough nitrogen, so that rhizobium inoculation is not needed for soy production, reducing farming costs (Coulter and others 2010). Soybean production in Minnesota is clearly feasible despite the low frost tolerance of soybeans (Watson and others 2017). However, Minnesota farmers must determine whether they wish to grow GM or non-GM soybeans. GM soybeans are more herbicide tolerant and/or insect resistant, but may deter consumers; while non-GM soybeans attract less negative attention, contracts often require them to be grown using certified seeds and specific procedures (Thrane and others 2017). Non-GM soybean growers may also have access to a larger market, because they can incorporate their soybeans into GM and non-GM foods.

Processing soybeans to obtain functional soy protein ingredients is also feasible, but requires additional steps to remove the oil, antinutritional factors (e.g. trypsin inhibitor), and off flavors. Producers must consider the impact that specific processes will have on the solubility, functionality, and effectiveness of the soy protein ingredients; the changes that prove problematic will depend on the role the ingredient intends to fulfill in each product. Incorporating these functional ingredients can lower costs and enhance the nutritional value of foods, but can also elicit concerns of allergenicity. To determine the feasibility of reformulating their products with soy protein ingredients, entrepreneurs should consider how important these concerns are to their consumers and if the added benefits of soy protein ingredients outweighs these concerns.

### *Pea Protein Ingredients*

#### *Recent statistics on pea yields/sales in Minnesota*

Estimated total sales in 2016 for Minnesota-grown peas were around \$24 million (USDA 2017c). Yields in 2016 were about 41.2 cwt/acre for peas (2016 State 2017). In 2016, peas were sold for an average of \$10.80/cwt in the US and \$12.60/cwt in Minnesota specifically (USDA 2017b; USDA 2017c). The higher average price in Minnesota for peas could be an incentive for farmers to increase their production, resulting in an increase in supply available for pea protein production. For farmers interested in entering the pea market,

the risk involved is minimal, as the crop is eligible for loss coverage under the 2014 Farm Bill (USDA ERS 2017d). In fact, adding legumes to crop rotations can be economically beneficial for farmers; legumes positively impact soil nutrient status and disease inoculum levels, increasing the yields of subsequent crops (Watson and others 2017). Tulbek and others (2017) have also reported that specifically adding peas to cereal and oilseed rotations can limit disease issues, fix nitrogen, and conserve water. These benefits can reduce production costs and promote sustainable farming by increasing yields and decreasing future fertilizer and natural resource use (Tulbek and others 2017).

### *Nutritional quality*

Yellow field peas are about 20-30% protein (Peng and others 2016). Due to their high lysine levels, pea proteins are complementary to those in cereals (Tulbek and others 2017); however, they are limited in methionine, making them incomplete (Tulbek and others 2017). The PDCAAS score for pea protein ranges from 0.8 – 0.9 (Rutherford and others 2015), depending on processing conditions. While it has lower PDCAAS than soy protein, pea protein is still a good quality protein. Unlike soy protein, research on the physiological benefits of pea protein is limited. However, pea protein is relatively high in branched chain amino acids (Rutherford and others 2015) that are essential for muscle repair. Overduin and others (2015) have shown that pea protein was comparable to dairy protein in promoting satiety. Another human study showed that pea protein may improve glycemic control (Mollard and others, 2014). While the effect of pea protein (compared to soy protein) on reducing the risk of heart disease is not widely researched, there is a general agreement that plant proteins compared to animal protein decrease the risk of heart disease (Kelemen and others, 2005).

### *Pea protein components*

Given that both pea and soy are legumes, pea protein components are similar to those of soy protein. The majority of pea proteins (65-80%) are globulins constituting the storage proteins legumin and vicilin (Barač and others 2010), which are responsible for the functional properties of pea protein. The ratio of legumin to vicilin can vary widely (0.2-1.5) among pea cultivars, which contributes to the differences in functionality (Casey and other 1982). The differences in content, composition and structure between vicilin and legumin result in different nutritional and functional properties (Barač and others 2010; O’Kane and other 2004; O’Kane and others 2005; Cserhalmi and other 1998; Rangel and others 2003; Kimura and others 2008). As is the case for soy protein, the individual protein composition of a pea protein ingredient and the fractions derived from it depend on the compatibility of a protein’s solubility in the extraction solvent used during the extraction and purification process.

### *Currently available protein ingredient forms*

The pea protein ingredients market is the fastest growing segment of the global plant protein market (Grand View Research, 2016). This growth is not only attributable to the protein being from a plant source, but also to the crop being non-GM and non-allergen, and to the advances in extraction techniques. Pea protein ingredients are gradually finding their way into several food and beverage applications. Food manufacturers are exploring successful ways of incorporating pea protein, as the sole protein ingredient contributing to the desired functionality/texture in unique and new applications.

Most pea protein ingredients in the market are produced from dry, whole, yellow peas (Pea Proteins Ingredients 2016). While pea protein ingredients are not the major driver for whole yellow pea production, they are of the highest value compared to pea flour and starch. In 2014, the production of ~ 21 thousand tons of pea protein ingredients corresponded to ~ \$30 million market value (Pea Proteins Ingredients 2016). Globally, ~ 20 manufactures are producing pea protein ingredients, including Cargill, Roquette, Cosucra, and all are competing on production efficiency and functionality. Commercially, pea protein ingredients are obtainable upon further purification post milling (including cleaning, drying, de-hulling, milling, and air-classification). Pea protein ingredients include, air classified pea flour 55 (protein content 55%), pea protein concentrate, pea protein hydrolysate, and extruded pea protein. Pea protein concentrate and isolate are produced mostly via isoelectric precipitation, which is based on alkaline solubilization of the protein to separate it from starch and fiber, followed by precipitation at the protein's isoelectric point (pH 4.5-5.5) to separate it from mono-, di-, and oligosaccharides (Adebiyi and Aluko 2011). As with soy protein, salt extraction produces pea protein isolates. Differences in pH treatment, holding time, salt concentration, and processing/drying temperature and time, contribute differences in the protein profile, structure, and accordingly its functionality.

### *Potential functionality and applications*

Pea protein functionality does not outperform commonly used protein ingredients. Pea protein solubility, for instance, is relatively poor (Adebiyi and Aluko 2011). Pea protein emulsification properties are comparable to soy protein (Toews and Wang 2013), while gelation strength is inferior to that of soy protein (Sun and Arntfield 2010). Differences in functionality, however, are dependent on the source of the protein used for comparison. Commercially, development of many different variants of soy protein ingredients occurred over the years for targeted functionality. While soy protein ingredients have gone through years of research and development to improve functionality, pea protein ingredient development is yet to catch up. Limited enzymatic hydrolysis may result in enhanced functionality of pea protein, depending on the enzyme used, degree of hydrolysis, and environmental conditions (Barać and others 2011). Recent research has shown that enzymatic hydrolysis of pea protein by trypsin results in enhanced emulsification properties (Tamm and others 2016). Choice of enzyme and hydrolysis

conditions to impart a targeted change in molecular characteristics to improve functional properties of pea protein for unique applications is still an open area for research.

Currently, pea protein is used in a variety of products, including pastas, meats (meat extenders/texturizers), extruded snacks, and bakery goods (Tulbek and others, 2017). In these foods, the protein functions as emulsifiers, gelling agents, foaming agents, and meat extenders/texturizers. However, the function of pea protein and its effectiveness in such products is highly dependent of the method of production and the processing conditions. In general, manufacturers of protein isolates must consider the characteristics they desire for their products prior to implementing a processing plan (Kiosseoglou and Paraskevopoulou, 2011).

### *Advantages*

From a sustainable agriculture perspective, adding peas to crop rotations enhances the structure, nutrient content, and levels of organic matter in the soil, increasing the yields of subsequent crops (Watson and others 2017). From a manufacturer's, and subsequently consumer's perspective, pea proteins are not as of yet allergenic, nor genetically modified. Additionally, processing of peas to extract protein is a cleaner process than that of soybeans, which typically utilize hexane for the extraction of the soybean oil.

### *Barriers*

Compared to soy protein, pea protein has lower solubility and consequently lower functionality, as mentioned previously. Accordingly, manufacturers often blend pea protein with other sources of proteins (e.g. whey protein) in different applications (baked goods and beverages) to obtain the desired functionality and mask the beany flavor. On the other hand, while the FDA doesn't require labeling pea protein as an allergen, it may potentially become allergenic with increased exposure, as was the case for soy protein. The composition and structure of pea proteins are similar to soy protein, so it will not be surprising if allergenicity incidences start occurring. The solubility of pea proteins at certain pH can also present problems. Other potential barriers are the antinutritive factors, including lectins, trypsin inhibitors, and phytic acid, which can limit protein digestibility and cause gastrointestinal distress (Tulbek and others 2017; Asgar and others 2010). Several processes, however, remove antinutritive factors, such as dehulling, soaking and thermal treatments (Asgar and others 2010). While these processes can remove or make many antinutritive factors inactive, they can also increase the time and cost of pea protein ingredient manufacturing, as well as negatively affecting protein solubility and functionality (Kiosseoglou and Paraskevopoulou 2011). Entrepreneurs interested in manufacturing products containing pea protein ingredients should consider all these factors.



## *Feasibility*

Growing peas is very feasible in Minnesota especially with the recent development of pea varieties for northern regions, and the additional benefits that adding peas to crop rotations provide to the soil, subsequent crops, and farmers via reduced fertilizer and water use (Tulbek and others 2017; Watson and others 2017). The antinutritive compounds, effects of processing and environment on solubility and functionality, and impacts on sensory characteristics, are all factors that limit the potential of peas as protein ingredient sources; however, some of the aforementioned barriers can be overcome with thermal treatment, enzyme inactivation, protein modification via limited hydrolysis, and product reformulation. The feasibility of manufacturing pea protein ingredients is demonstrable by the pea flours, concentrates, and isolates already on the market.

## *Canola as an Emerging Protein Ingredient Source*

### *Recent statistics on yields/sales in Minnesota*

Canola or rapeseed is a member of the genus Brassica, which also includes members such as broccoli and Brussels sprouts. The term canola (Canadian oil low acid) was given to rapeseed oil, which has less than 2% erucic acid, a fatty acid associated with adverse health effects (Knutsen and others 2016) and only 30  $\mu$ moles/g of glucosinolates, a class of potentially toxic secondary metabolites (Sedbrook and others 2014). The land in the region dedicated to growing canola has grown significantly over the past decade. Over the last four years, about 20 million acres of land in Western Canada and 1.5 million acres in the Northern U.S. states are used for growing canola. Across the United States, the average price and yields of canola in 2016 were \$16.40/cwt and 1,824 lbs/acre, respectively (USDA 2017a; USDA 2017b). In Minnesota specifically, the average price was slightly higher at \$17.60, while yields were slightly lower at 1,700 lb/acre (USDA 2017c). Canola grown in this region is mainly for the production of canola oil. It is a very healthy oil, high in the Omega-9 monounsaturated fatty acid oleic acid and low in saturated fatty acids. The meal remaining after extraction of oil is usable as animal feed. However, the meal is high in protein, and could be a good source of protein ingredient for food use.

### *Nutritional quality*

The average protein content of canola is 20% (Wanasundara and others 2017). The nutritional quality of canola protein, in terms of PDCAAS, is comparable to that of soy protein (Wanasundara and others 2016), in fact it could potentially surpass soy protein as the gold standard among vegetable proteins (Wanasundara and others 2017). The essential and sulfur-containing amino acids, methionine and cysteine, in canola protein exceeds the nutritional requirements for both children and adults, while soy protein and casein both fall short of the requirements for infant nutrition and only casein meets the requirements for higher age categories (Wanasundara and others 2017).

### *Protein components*

Canola protein is made up of four different components albumins, globulins, prolamins, and glutelins. The albumin and globulin fractions constitute the majority of the canola proteins. The albumin protein in canola is napin, which makes up 20% of the protein content. The globulin protein in canola is cruciferin, which makes up 60% of the protein content (Hoglund, and others, 1992). Cruciferin is potentially an allergen for people allergic to mustard, a seed belonging also to the genus Brassica (Palomares and others 2005). Mustard allergies are not very common, but this is an important consideration when developing potential food applications for canola proteins.

### *Currently available protein ingredients*

Canola meal, mostly used as animal feed, generally contains 37 - 41% protein (Wanasundara and others 2016). The protein from the meal is extractable and concentrated in a number of ways, mostly alkaline pH extraction and salt extraction (Wanasundara and others 2016). Each extraction method uniquely impacts protein composition, functionality, and digestibility (Wanasundara and others 2017). Unlike canola meal, canola protein flour, concentrates (>70% protein), and isolates (>90% protein) obtained via extraction and separation are intended for human consumption (Wanasundara and others 2016, 2017).

Most notably MCN BioProducts (MCN), BioExx Specialty Proteins (BSP) and Burcon NutraScience Corporation (BNC) have attempted the development of extraction technologies for the concentration of proteins from canola. BNC is the only company that has been successful in developing commercial canola protein ingredients, utilizing a unique, patented aqueous extraction processes. BNC's produces three canola protein products, Supertein<sup>®</sup>, Puratein<sup>®</sup> and Nutratein<sup>®</sup>. Supertein is a napin protein isolate and Puratein is a cruciferin protein isolate, both targeted for food use. Nutratein contains mixture of both napin and cruciferin for feed use. Burcon has obtained Generally Recognized As Safe (GRAS) approval from the FDA for Supertein and Puratein and have made a substantial equivalence application for Nutratein. Another company close to commercializing canola protein is Ancher Daniels Midland (ADM), with an estimated launch date of 2020.

### *Potential functionality and applications*

In general, canola protein offers good functional properties, including water binding capacity, solubility, emulsification, foaming, and gelation, compared to other oilseed proteins (Aider and Barbana 2011; Moure 2006). Due to the variety of protein fractions in canola protein concentrates and isolates, as well as the myriad processes for their production, the functional properties of canola protein ingredients may vary (Wanasundara and others 2016).

Cold-pressed canola flour is useful in sauces, sausages, and baked goods (Wanasundara and others 2017). Canola protein can be an emulsifier for mayonnaise in the form of hydrolyzed canola meal (Wanasundara and others 2017). However, when alkaline extraction and acid precipitation is used to concentrate the protein, isolates demonstrate limited emulsifying properties (Wanasundara and others 2017); yet they are still comparable to those of soy proteins (Wanasundara and others 2017). In contrast, when the proteins are hydrolyzed the emulsifying abilities are enhanced (Wanasundara and others 2017). In their purified form, canola proteins also have potential as gelling agents (Wanasundara and others 2017). The napin fraction of canola protein appears to be better suited as a foaming agent over a wide-range of pH than cruciferin (Wanasundara and others 2017). In its isolated form, napin's foam-forming abilities are comparable to those of whey protein isolate (Wanasundara and others 2017). Entrepreneurs interested in canola protein ingredients might want to consider different fractions and isolates for different applications.

### *Advantages*

Canola crop is better suited for areas with drier and shorter growing seasons than corn and soybean (USDA ERS 2017a). In such areas, adding canola to crop rotations can diversify crops and provide a more effective use of the land. Including canola in three or four year crops rotations can also limit the spread of disease and pests over time (USDA ERS 2017a). For farmers interested in introducing canola production to the crop rotation, the monetary and labor-related risks are minimized due to the recently developed herbicide-resistant varieties (USDA ERS 2017a).

Utilizing canola protein, a by-product of oil extraction, generates another source of income for canola producers. Canola protein, as of yet, is not one of the FDA "big 8" allergens, and has promising functional properties compared to other plant proteins. Using chemical and solvent-free protein extraction methods, canola protein ingredients can be used in clean-label foods (Campbell and others 2016), thus increasing the market value.

### *Barriers*

Canola is a genetically modified crop, thus manufacturers seeking to claim non-GM on their label cannot use canola protein ingredients. Canola also contains 2S albumin proteins, which have some amino acid sequence homology to those responsible for allergenicity in mustard (Wanasundara and others 2017). In the EU and Canada, canola ingredients are labeled as possible allergens (Wanasundara and others 2017). Additionally, the processing of canola protein ingredients may require unique processing steps to remove canola's myrosinase may be required. Myrosinase is an enzyme that hydrolyzes glucosinolates into products responsible for pungency and in some cases goitrogenic activity and hepatotoxicity (Botti and others, 1995). However, glucosinolates are present in low concentration in canola, so the removal of myrosinase may not be necessary.

On the other hand, removal of phenolic compounds is necessary. Phenolic compounds, extracted with the proteins, are problematic as they impart a dark color and bitter, astringent flavor (Wanasundara and others 2017). Additional processing steps would increase the costs of the production of canola protein ingredients. However, other benefits, such as the protein's functionality may increase its value, offsetting the cost of production.

Further processing issues may include the variability of yields, the impact of growing conditions, and the negative impact that methods used to maximize oil extraction may have on protein extractability, functionality, and nutritional quality (Wanasundara and others 2017). Overcoming these barriers will involve future research and testing of canola processing and the protein's nutritional and functional properties.

### *Feasibility*

With its preference towards cooler climates and a tolerance for a variety of soils, growing canola in Minnesota is not only feasible, but a natural fit for the region (Wanasundara and others 2017). The average price of canola in Minnesota for 2016 was higher than the national average because the yields per acre were lower. But, as production increases, the price will decrease (USDA 2017a; USDA 2017b; USDA 2017c). Reductions in canola price would potentially decrease the cost of canola protein production. The feasibility of commercial production, however, is dependent on several factors, including antinutritive content, functionality, nutritional quality, consumer acceptance, and most importantly advancement in processing technologies.

Although canola protein ingredient production is possible using what is known from soybean protein processing, differences in seed and protein characteristics, necessitates further investigations of tailored processing technologies and functionalization of this new ingredient (Wanasundara and others 2017). Additionally, current processes maximizing canola oil extraction negatively impact protein extraction and quality. Thus, a choice to either maximize oil yields or follow processes that maximize protein yield and protect its quality becomes necessary. Alternatively, and as is the case with soybean processing, manufacturing of canola products can be divided into two targets, one targeting maximum oil production and utilization of the resultant meal for animal feed, and the other targeting lower oil yield and utilization of the resultant cake for the production of high quality protein ingredient. The production of a high-value ingredient would compensate the reduced profit for lower oil yield, which may result in higher profit. Overall, the future of canola protein production is promising if advancement in extraction processes and functionalization is pursued.

## Potentially Viable Sources of Plant Protein

For emerging protein sources, such as canola protein, which have been studied somewhat extensively, commercialization and wide utilization are more certain and will happen sooner than for potentially viable sources, in which research is still in the preliminary stages. Since emerging protein sources require less effort, time, and research for their initial manufacturing and commercialization, entrepreneurs are likely to invest in these protein sources, generating a great deal of competition. In contrast, potentially viable sources are less known, and far less researched. However, businesses that begin exploring their potential now will likely face little competition for several years after commercialization. Investment in researching these proteins will aid in reaching the manufacturing stage. The potentially viable protein sources discussed in this paper include pulses (other than pea), hemp, sunflower, corn, oats, potato, camelina, and pennycress.

### *Other pulses*

The term “pulses” pertain to leguminous crops producing seeds within a pod, and harvested only for dry grain, not green for food.

Pulses are receiving quite a bit of buzz as viable sources of protein. The UN General Assembly named 2016 the international year of the pulses. It promoted recognition of the nutritional benefits of pulses while making the public aware of these sustainable crops that could contribute to food security and nutrition. Pulses, which include various types of beans, lentils, chickpeas, and peas, are low-fat sources of protein, fiber, minerals and vitamins. While, there has been major advances in the production of pea protein (globally 20 manufacturers are producing pea protein ingredients), utilization of other pulses as sources of protein ingredients remains limited. This section will focus on other pulses, mainly beans, as viable sources of protein ingredients.

### *Recent statistics on yields/sales in Minnesota*

In 2016, dry beans grown in Minnesota yielded an average of 2,230 lbs/acre and generated just over \$97 million in sales (USDA 2017c). For the same year, the national average price of dry beans was \$30.20/cwt (USDA 2017b); \$0.60 higher than the average price in Minnesota (USDA 2017b). Compared to peas, dry beans are about \$17.00/cwt more expensive (USDA 2017b; USDA 2017c). While lower bean prices would offer Minnesota-based businesses using bean proteins an advantage over out-of-state competitors, they may be too expensive to compete well with manufacturers using other, less expensive pulses, namely peas.

### *Nutritional quality*

Certain varieties of dry beans, such as, kidney contain around 18 to 24% protein (Wani and others 2015). Similar to other legumes, kidney beans have a low concentration of sulfur-

containing amino acids, namely methionine (Wani and others 2015), as well as a low concentration of tryptophan, both are essential amino acids.

Most legumes appear to have a relatively low PDCAAS with chickpea, sword bean, kidney bean, and velvet bean having PDCAAS values of 0.72, 0.58, 0.68, and 0.61, respectively (Bridge2Food; Wani and others 2015), mostly attributed to low digestibility. Soybean is an exception with an extremely high PDCAAS of 0.96, making many other legumes (pulses) nutritionally inadequate replacements (Wani and others 2015). However, several techniques, including dehulling, soaking, and autoclaving can improve the digestibility of some pulse proteins (Boye and others 2010). Also, beans contain some antinutritional factors, such as lectins, saponin, trypsin inhibitor, and phytic acid (Rui and others, 2016). Different varieties have varying concentrations of these components, but processing and thermal treatment may reduce their content.

### *Protein components*

The majority of pulse proteins are albumins and globulins, which are water and salt-soluble, respectively (Boye and others 2010). As in soybean, the most notable globulins in beans are legumin (11S) and vicilin (7S) (Boye and others 2010; Wani and others 2015); both are complex storage proteins with multiple subunits (Boye and others 2010). Globulins in pulses are present in higher abundance than albumins and thus are the main contributors to the protein's functionality.

### *Currently available protein ingredient forms*

Flour, concentrates, and isolates produced from pulses include chickpea, black-eyed bean/pea and common bean (Boye and others 2010). For example Top Health Ingredients Inc., supplies AdvantaFAVA 85 protein concentrate (85% protein).

### *Potential function and applications*

Research is limited on pulse proteins other than pea. However, bean proteins studied as meat extenders showed potential for improving water retention, preventing fat loss, and increasing protein content (Asgar and others 2010; Boye and others 2010). Additionally, proteins, such as kidney bean proteins, showed they could be acceptable emulsifiers and foaming agents at neutral pH (Wani and others 2015). However, as is the case for pea protein, the solubility of bean proteins is far from being adequate at pH levels relevant to food applications, impacting other functionalities (Wani and others 2015). As most pulses have similar properties, pulse protein manufacturers must consider the limited functionality of these proteins, and consider ways to improve their performance following protein modification and unique processing.

### *Advantages*

Adding legumes to crop rotations results in improvement to the nutrient levels in the soil (Watson and others 2017). Nutrient improvement can reduce synthetic fertilizer needs and thus encourage farmers to enter into pulse production. There are also no pulses that have GM varieties. Another advantage is the low occurrence of allergenicity to pulse proteins, with only few incidences reported (Boye and others 2010).

### *Barriers*

Legume production may present environmental problems, such as nitrate leaching into the water supply and nitrous oxide production (Watson and others 2017). Addressing these problems may increase the cost of pulse production and reduce profit margins. Another barrier pertains to the limited functionality of pulse proteins discussed earlier.

### *Feasibility*

The production of pulses, such as dry beans, is possible in Minnesota, as demonstrated by the high yields and sales for 2016; in fact, dry beans may be more profitable than the production of other pulses like peas, since bean prices in 2016 were much higher per cwt than pea prices. Additionally, the average price for Minnesota beans was lower in 2016 than the national average, which may encourage manufacturers to buy Minnesota beans, benefiting local farmers. Pulse protein functionality is inferior to other protein ingredients, namely soy and milk proteins. Creative measures need to be employed during manufacturing of the ingredients and during formulation with such ingredients. However, considering consumer desire for non-GM characteristic and low allergenicity incidences, focusing efforts on functionality improvement is worthwhile.

### *Hemp Protein*

#### *Recent statistics on yields/sales in Minnesota*

The production of hemp (*Cannabis sativa L*), which is also known as industrial hemp, became legal in Minnesota after the 2014 Farm Bill shifted the responsibility of regulating hemp farming from the federal government to the states (Meersman, StarTribune, 2017). However, its production is still limited and heavily controlled. Regulation stems from both hemp and marijuana coming from the same cannabis species. Yet, industrial hemp contains negligible levels, if any of the narcotic substance, tetrahydrocannabinol.

In 2016, the Minnesota Department of Agriculture (MDA) began its first hemp pilot program with 37 acres being planted the first year and 2,100 acres the second (Meersman, StarTribune, 2017). While Minnesota farmers can participate in the program, they must apply and pay program fees each year, which could increase production costs and deter involvement (Meersman, StarTribune, 2017). Hemp production, however, limits the need

for herbicides, fungicides, or pesticides, which may negate the additional cost of entering the program (Meersman, StarTribune, 2017; Aluko 2017).

Hemp seed is available through the MDA to special permit holders. While MDA pilot program participants cannot sell their seeds for propagation or save them for the next growing season, they are able to sell the seeds to processors (Minnesota Department of Agriculture, 2017). Currently, there are few Minnesota processors of industrial hemp and only processed (non-viable) seed can cross state borders. For reference, hemp seed sells for about \$2.50/lb in Canada, which results in a seed cost per acre of about \$125 (Meersman, StarTribune, 2017); slightly higher than the \$100 to \$125 for corn seed (Meersman, StarTribune, 2017).

### *Nutritional quality*

In addition to high oil content (30%), hemp contains about 25% protein, which is similar to the protein concentration in yellow field pea, but lower than that of soybean (Yin and others 2008). Hemp protein is low in lysine and tryptophan. Although the protein has high digestibility (~95%), it has a relatively low, PDCAAS, 0.66, attributed mainly to lysine deficiency (Aluko 2017). Nevertheless, hemp meets the United Nation's Food and Agriculture Organization's essential amino acid recommendations (except for lysine) for infants and children (Yin and others 2008). An absence of protease inhibitor, compared to beans, further improves its nutritional value (Aluko 2017).

### *Protein components*

Hemp protein is composed mainly of the globulin edestin and albumin. Edestin, a hexameric legumin, is the major component of hemp protein (60-80% of the total protein). Additionally, hemp protein contains a methionine- and cystine-rich protein component (Tang and others 2006).

### *Currently available protein ingredient forms*

Hemp use in the United States is limited, but a number of hemp protein related products are available on the Canadian and European markets, including hempseed cakes, defatted hemp seed flour, hemp protein concentrate, and hemp protein isolate (Cherney and Small 2016). As the byproducts of oil extraction, hempseed cake is high in protein and mostly used as animal feed (Cherney and Smalls 2016). Hemp protein concentrates produced from defatted cake have 50-75% protein, while isolates often have greater than 85% protein (Aluko 2017). Processing greatly affects the characteristics of hemp protein ingredients, as seen with the superior solubility, foaming capacity, water-holding capacity, oil-holding capacity, and digestibility of hemp protein concentrates obtained through membrane filtration as compared to those produced following pH extraction (Aluko 2017).



### *Potential function and applications*

Hemp and its products, namely fiber, are used for industrial applications such as paper and durable fabric production (Tang and others 2006). Historically, the utilization of hempseed was for food sources, and is currently legal for human consumption in both Canada and the United States. In addition to raw, cooked, or roasted consumption, hemp produce edible oil and protein ingredients. Accordingly, there is research on the functionality of hemp protein ingredients for various applications. In general, the most effective use of hemp protein is as emulsifiers, foaming agents, and moisture retainers (Yin and others 2008; Aluko 2017). Compared to defatted hemp seed flours, hemp protein isolates tend to have superior oil-holding capacity, foaming capacity, gel-forming ability, and foaming ability (Aluko 2017). However, compared to soy protein isolate, hemp protein isolates, produced using pH extraction, had inferior functionality (Tang and others 2006).

### *Advantages*

Hemp can be a rotation crop. It prevents soil erosion and enriches the soil with valuable nutrients from after harvest residue. Moreover, the need for pesticides, and fungicides is limited for hemp production, thus growing hemp is potentially less input intensive and inexpensive (Aluko 2017). Hemp is also advertised as non-allergenic, which is an added advantage compared to other protein sources, such as soy (Aluko 2017).

### *Barriers*

Farmers must obtain State approval before they can grow hemp, which presents a barrier to farmers who are not familiar with governmental procedures (Cherney and Small 2016). The production and sale of hemp is further complicated by the differing allowable tetrahydrocannabinol levels among countries, which could potentially hinder the international trade for Minnesota grown hemp (Cherney and Small 2016). In addition to legal barriers, hemp also requires large amounts of water, nitrogen, phosphorus, and potassium (Cherney and Small 2016). Barriers to the use of hemp as a source of protein ingredients include supply chain and inferior protein functionality. Hemp protein isolates have lower solubility than soy protein isolate, contributing to reduced functionality (Yin and others 2008). Protein hydrolysis, however, did result in improved solubility (Aluko 2017; Yin and others 2008). This area, however, requires further investigation.

### *Feasibility*

Based on the MDA's pilot program, hemp production in Minnesota is possible (Cherney and Small 2016). For optimal growth, conditions similar to those for corn are required, including a moderate climate, lots of water, and well-aerated, fertile soil containing large amounts of organic matter (Cherney and Small 2016). Best results stemmed from the addition of hemp to a four-year crop rotation (Cherney and Small 2016). However, commercial production in Minnesota is still illegal, which will have to change for large-scale

hemp protein production to be feasible. There is limited potential for hemp as a protein ingredient because of its inferior functionality compared to soy protein. Additional processing steps, such as hydrolysis, may improve the functionality of most proteins, but may increase costs. Additionally, processors will have to account for expensive protein production options such as membrane filtration, which result in the production of hemp protein ingredients with good solubility, foaming capacity, water and oil holding capacity, and protein digestibility (Aluko 2017).

### *Sunflower Protein*

#### *Recent statistics on yields/sales in Minnesota*

Sunflower's main utilization is for the production of oil. The meal remaining after extraction of oil is mostly for animal feed. However, the meal is high in protein, and could be a good source of protein ingredient for food use.

Minnesota sunflower yields in 2016 were approximately 1,465 lbs/acre, with sales totaling over \$23 million (USDA 2017c). Compared to the 2016 US average price for sunflower of \$17.40/cwt, Minnesota sunflower prices were \$2/cwt higher (USDA 2017b). The higher selling price may incentivize Minnesota farmers to enter into or expand sunflower production. Inclusion of sunflower in the crops eligible for price loss coverage under the 2014 Farm Bill may also act as an incentive to increase sunflower production among Minnesota farmers by limiting their monetary risk (USDA ERS 2017d). Increasing sunflower production would make this crop more readily available for protein ingredient production. However, there is still some financial risk as sunflower yield and protein content is dependent on soil nitrogen, phosphorus, and potassium levels, thus some farmers will need to increase their production budgets to include the costs of soil fortification (González-Pérez 2015).

#### *Nutritional quality*

On a dry basis, sunflower seeds contain between 20-40% (hulled or de-hulled) protein (González-Pérez 2015). In comparison, defatted sunflower meal is often around 30% protein, but can reach levels as high as 53-66% when extracted using an organic solvent (Ren and others 2017; González-Pérez 2015). Besides having a low lysine content, sunflower protein meets all other amino acid requirements and is considered a source of good quality protein (Sunflowerseed 2017; González-Pérez 2015). Sunflower protein contains about 20% branched chain amino acids, essential for muscle repair. The PDCAAS value for sunflower protein is relatively low, however, having a score of 0.6, given the relatively low digestibility of this protein. On the other hand, there are no antinutritional factors in sunflower (González-Pérez 2015). Sunflower seeds do contain phenolic compounds that can interact with proteins and negatively affect their quality, but minimization of these compounds' concentration is achievable through processing or breeding (González-Pérez 2015).

### *Protein components*

Storage proteins in sunflower seeds represent around 85% of the total seed protein. The main constituents of the sunflower storage proteins are the 11S globulin helianthinin and the 2S albumins (González-Pérez 2015); high levels of globulins and albumins are largely responsible for the high solubility of sunflower seed protein at pH level below 3 (González-Pérez 2015). In general, the helianthinin globulin-type proteins are present in higher concentrations than the albumins, but studies vary greatly in the reported percentages (González-Pérez 2015). Changes in their environment, such as pH and temperature, extraction methods, and processing conditions affect the functional and nutritional properties of these proteins.

### *Currently available protein ingredient forms*

Sunflower protein powders for human consumption are available, e.g. the Natures Plus organic sunflower protein powder, which has about 50% protein. However, inexpensive, large-scale processing methods to isolate sunflower proteins are currently lacking (González-Pérez 2015).

### *Potential functionality and applications*

Sunflower protein has relatively good solubility, thus may potentially be a functional protein (González-Pérez 2015). However, sunflower protein is often a byproduct of oil extraction and usually is denatured during processing, reducing its solubility and functionality (González-Pérez 2015). If sunflower protein fractions are isolated, without being denatured, sunflower proteins may become soluble over a range of ionic strength and pH.

Sunflower seed protein has been incorporated into beef and dairy cattle feed, but its use in food is scarce, apart from the protein powder available in the market from Natures Plus, advertised as organic, allergen free, vegan, and high in branched chain amino acids. Limited research is available on sunflower protein functionality for food applications.

### *Advantages*

Sunflower seeds are widely available, are high in protein content, and have no antinutritional factors (González-Pérez 2015). When extracting the protein under mild conditions, the solubility and overall functionality are preserved. As opposed to several common protein sources, such as soy, dairy, and egg, allergenicity to sunflower protein is rare. Sunflower seeds are also non-GM, which further expands their potential market and eliminates barriers related to governmental deregulation and consumer acceptance (Kleingartner 2015).

## *Barriers*

Not enough research of sunflower protein, extraction methods, processing technologies, and functionalization currently exists. The oil extraction process applied currently is detrimental to the protein quality. Additionally, removal of chlorogenic acid (CGA) is necessary, if the protein is to be isolated for human consumption (González-Pérez 2015). Presence of CGA results in unfavorable changes to the color, and cross-linking of the protein, thus resulting in lower functionality, and bioavailability of the final product (González-Pérez 2015). Removal of CGA will add to the cost of sunflower protein production.

For farmers, among the largest barriers to sunflower production is a lack of varieties resistant to production challenges. Unlike corn and soybean varieties, GM sunflowers do not exist. However, disease and insect resistance is obtainable only through time-consuming methods, such as traditional backcrossing (Kleingartner 2015). GM sunflower seeds may become available in the future, but the market is currently too limited to negate the costs of their development (Kleingartner 2015).

## *Feasibility*

Yields and sales of sunflower in Minnesota demonstrate that production in the state is possible. Sunflower is susceptible to common Minnesota plant diseases, such as, sclerotinia, downy mildew, and phomopsis, which can all reduce yield (Kleingartner 2015). Although these issues have a negative effect on sunflower yield, they counterbalance one another by the advantages of sunflower crops, such as, the drought-resistance of some genotypes and the benefits of adding sunflower to wheat and barley crop rotations (González-Pérez 2015; Kleingartner 2015). Despite the feasibility of sunflower production in Minnesota, currently available processing methods present barriers to commercial production of sunflower protein ingredients. Before wide-scale production becomes economically feasible, less expensive, non-denaturing methods to eliminate the CGA and phenolic compounds in sunflower proteins, while still preserving the protein nutritional and functional properties, will have to be developed.

## *Corn Protein*

### *Recent statistics on yields/sales in Minnesota*

In 2016, sales for corn in Minnesota surpassed \$5 billion and yields topped 190 bushels/acre (USDA 2017c). In the same year, the average price in Minnesota per bushel of corn was \$3.30 (USDA 2017b); ten cents lower than the US average. Minnesota-based protein ingredient producers (or potential producers), specifically would benefit from the lower price of corn in the region (USDA 2017b), as this will offset transportation costs.

### *Nutritional quality*

Depending on the variety, protein content in corn ranges between 10-15%, which is lower than many of the other plant sources discussed in this paper (Hojilla-Evangelista 2014). Corn gluten meal, a byproduct of cornstarch production, has a greater protein content comprising of 60%-71% (Jin and others 2014). Corn protein, however, is limited in lysine and tryptophan, and has a low PDCAAS of 0.42, making it an incomplete protein (Jin and others 2014). The PDCAAS, specifically protein digestibility, may be enhanced through enzymatic hydrolysis during processing. Corn gluten meal may also contain antinutritive compounds, such as tannins (Nassan and others 2016).

### *Protein components*

Corn protein is mainly composed of zein and glutelin-type protein (Jin and others 2014). Zein, which is the major protein in corn, is essentially insoluble in water due to its high percentage of hydrophobic amino acids. The poor solubility of corn protein limits greatly its use and benefits as a food ingredient (Jin and others 2014). When considering corn germ, which contains 18-22% protein on dry basis, the albumin and globulin-like proteins are more functional (Hojilla-Evangelista 2014).

### *Currently available protein ingredient forms*

The use of corn protein in food systems is quite limited. On a large-scale, common applications of corn proteins are their use as corn gluten feed or corn germ meal for animal feed (Jin and others 2014; Hojilla-Evangelista 2014). There were smaller scale attempts to incorporate corn germ protein isolates, germ flour, and germ protein flour in various food systems as extenders and stabilizers (Hojilla-Evangelista 2014).

### *Potential functionality and applications*

Corn proteins have essentially very poor solubility (Nassan and others 2016). This low solubility is detrimental to all other functional properties. If the protein is insoluble, it will not form an adequate gel, will not emulsify, nor stabilize a foam (Hojilla-Evangelista 2014). While there were attempts to improve the nutritional value of foods, such as cookies, muffins, and beef patties, through the incorporation of corn germ protein, they were on a small-scale (Hojilla-Evangelista 2014). The absence of larger scale applications suggests the limited success of corn protein thus far. That said, subjecting corn protein to modification may result in targeted improvement in nutritional value and functional properties. However, this requires further investigations.

### *Advantages*

Corn protein advantages are limited. The main advantage over other plant proteins, such as soy, is not being a common allergen. Corn protein also shows a minimal solubility loss

after heat treatment, which is beneficial when trying to eliminate antinutritive factors (Hojilla-Evangelista 2014). However, an extremely low initial solubility makes this advantage almost inconsequential.

### *Barriers*

Corn protein solubility and functionality are so low that other non-allergenic protein sources, such as sunflower and hemp, may be more desirable. Additional barriers to corn protein is its low nutritional value, antinutritive factor content, and the fact that most supplies are genetically modified (Jin and others 2014; Nassan and others 2016; USDA ERS 2017b). About 92% of the total acreage of US corn production in 2016 grew genetically modified corn. Accordingly, the supplies for utilizing corn protein in organic products is relatively low (USDA ERS 2017b).

### *Feasibility*

Corn production in Minnesota is extremely prevalent; however, the amount, bioavailability, nutritional benefits, solubility, and functionality of the protein in corn is limited. Additionally, the demand for corn has increased with a recent push towards ethanol production. Therefore, while corn production is possible, large-scale manufacturing and commercialization of corn-based protein ingredients will likely not occur until the industry addresses these barriers. Particularly important to improving the feasibility of the production, and utilization of corn protein ingredients, is the development of processing methods to improve nutritional quality, solubility and functionality. Based on current corn sources, technology, and availability of other protein sources, the potential of corn becoming a key source for plant-based protein ingredients is low in the near future.

## *Oat Protein*

### *Recent statistics on yields/sales in Minnesota*

Minnesota oat yields and sales in 2016 were around 68 BU/acre and \$16.7 million, respectively (USDA 2017c). The price per bushel of oats was around \$2.05, which was lower than that of corn and soybean (USDA 2017c). Oats are relatively resistant to disease and compete well with weeds, thus they can be grown with minimal amounts of fertilizers and herbicides, keeping production costs low (Mäkinen and others 2017). Compared to other crops, the low price and production cost of oats could make it a cost effective source for the manufacture of a plant-based protein ingredient. However, manufacturers must also consider the cost of oat protein isolation (Mäkinen and others 2017).

### *Nutritional quality*

After dehulling, the oat kernel (oat groat) contains between 11-24% protein (Mäkinen and others 2017). The protein content in oats depends on growing conditions; particularly, it

relates universally to water availability and directly related to fertilizer nitrogen levels (Mäkinen and others 2017). Higher protein content also negatively correlates with the yield (Mäkinen and others 2017). Oat protein ingredients, such as PrOatein, have a PDCAAS of only 0.46 (Tate and Lyle ppt). In general, the PDCAAS for most isolated oat proteins is between 0.45-0.51 (Mäkinen and others 2017). PrOatein and oat proteins, in general, have inadequate content of lysine (Mäkinen and others 2017; Tate and Lyle ppt). Oat proteins meet all other FAO amino acid requirements for adults and are particularly high in glutamine (Tate and Lyle ppt; Mäkinen and others 2017).

### *Protein components*

Globulin like proteins represent the major protein fraction in oats (Jiang and others 2015). Oats also contain prolamin like proteins, but in substantially lower content than globulins, 4-14% compared to 70-80%, respectively (Mäkinen and others 2017). The globulins in oats are mostly insoluble in solutions with neutral or mildly acidic pH, which negatively affects their functionality and potential applications in various foods (Jiang and others 2015). However, oat globulins have a high denaturation temperature that preserves their structure and characteristics during thermal processing (Jiang and others 2015).

### *Currently available protein ingredient forms*

Recently, Tate & Lyle released to the market PrOatein (54% protein), an oat protein-based ingredient (Tate and Lyle ppt). They commercialized PrOatein for boosting the protein content of products such as breakfast cereals and bars, breads, nutritional shakes, and healthy snacks (Tate and Lyle ppt). Beyond PrOatein, the market for oat protein ingredients is limited. Oat manufacturers have focused more on utilizing oats for their  $\beta$ -glucans and oil content rather than their protein.

### *Potential functionality and applications*

In addition to utilization for achieving protein content claims, oat proteins may have other potential applications. For instance, emulsions formed with oat protein isolates (OPI) obtained from defatted oat flour demonstrated a high level of stability, (Mäkinen and others 2017). The gelling ability of OPI was acceptable upon enzymatic hydrolysis (Mäkinen and others 2017). Using trypsin hydrolyzed OPI, gels produced at pH 5-7 were comparable to or better than those made with egg-whites (Mäkinen and others 2017), possessing comparable water-holding capacity and mechanical properties (Mäkinen and others 2017). Conversely, OPI did not perform well as a foaming agent compared to soy protein (Mäkinen and others 2017). Functionalization and production of oat protein ingredient require further investigation and development.

### *Advantages*

For farmers, one of the major advantages of oats is that they are relatively disease resistant and compete well with weeds, reducing fertilizer and herbicide usage as

mentioned earlier. These traits make oats well suited for organic farming and usage in organic and clean label products (Mäkinen and others 2017). Oats are gluten free, and reports of allergenicity are low, and found exclusively in infants. With advancement in processing technologies and protein modification approaches, oat proteins may have great potential for various food applications, including those requiring high temperature processing.

### *Barriers*

The main barriers to the utilization of oat proteins in food include their low nutritional value, digestibility, and solubility in comparison to other protein sources. Being low in lysine, oats are an incomplete source of protein, often requiring the addition of legume proteins (Mäkinen and others 2017). The low nutritional quality of oat protein may not impact its potential use as an emulsifier, or gelling agent, but it limits usefulness for protein fortification.

### *Feasibility*

While farmers currently grow oats in Minnesota, production levels are much lower than for soybean and corn. The low production rate, along with a relatively low protein content may hinder the utilization of this crop for protein production on a large scale. However, oats do grow well in northern regions, as well as areas with wet weather and slightly acidic soil, making many locations in Minnesota suitable for its production. Therefore, if demand increases and land is available, oat production may increase. Additionally, oat's inherent disease resistance keeps the economic risk for oat production low, which could encourage farmers to plant it if demand and prices increase. The relatively low price of oats may motivate manufacturers to work with oats instead of the more expensive crops, such as soybeans. However, the additional costs of protein functionalization may negate the monetary benefits of low oat prices. The presence of oat protein ingredient in the market, such as ProOatein, demonstrates that oats can be a viable protein source. However to increase the availability of oat protein in the market, economic oat protein isolation and processing technologies will have to be developed.

## *Potato Protein*

### *Recent statistics on yields/sales in Minnesota*

Yield and sales of potato in Minnesota for 2016 averaged around 400 cwt/acre and \$152 million, respectively (USDA 2017c). Compared to the average U.S. price in 2016 of \$8.90/cwt, Minnesota potatoes were priced slightly higher at \$9.05/cwt (USDA 2017b; USDA 2017c).



### *Nutritional quality*

Potato fruit juice, a byproduct of potato starch manufacturing, contains about 1.5% (w/v) protein (Lomolino and others 2015). Other plant protein sources, such as legumes have over 20% protein, making potato inferior in terms of protein quantity. On the other hand, many find potato protein to be superior to many other vegetable and cereal proteins, having a PDCAAS of 0.93 and high levels of lysine and branched chain amino acids (Waglay and others 2013). Potato protein, however, has a low content of sulfur containing amino acids (Peška and others 2013). Threonine may also be limiting depending on cultivation, storage conditions, and potato variety (Peška and others 2013). Protein quality is also influenced by nitrogen fertilizer use (Peška and others 2013). Increases in nitrogen fertilizer use, results in higher overall nitrogen content, yet lower concentration of essential amino acids, and consequently quality (Peška and others 2013). Potato variety, however, is the main determinate of quality (Peška and others 2013).

### *Protein components*

Patatin is the major potato tuber protein, representing 30-40% of the total protein. Potato proteins are comprised also of protease inhibitors (50% of the total protein) and other minor components (10-20% of the total protein), mostly oxidative enzymes (Schmidt and others 2017). Patatin consists of glycoproteins, is highly soluble, and possesses antioxidative properties (Waglay and others 2013).

### *Currently available protein ingredient forms*

While some companies do manufacture potato protein products, the small amount of protein present in potato juice limits its commercial use. However, AVEBE, a large potato starch manufacturing company in The Netherlands, has introduced a number of potato protein ingredients as part of their line of Solanic® products. Similar to other potato starch manufacturers, AVEBE aimed at valorizing potato fruit juice, which is the by-product of starch production. Solanic® potato proteins, isolated from potato fruit juice, are commercialized as protein isolates with high purity (>90% protein).

### *Potential function and applications*

Published research on potato protein solubility and other functional properties is limited (Waglay and others 2013; van Koningsveld and others, 2001). Some findings suggest the protein fractionation process impacts functionality. Specific protein fractions obtained with hydrophobic interaction chromatography enhance potato protein functionality, namely emulsification, instead of whole protein powder (Schmidt and others 2017). Others have demonstrated that potato protein concentrates have adequate solubility, and foaming properties, and are superior to soy protein products in terms of emulsifying capacity (Løkra and others 2008). Proper extraction methods also generate potato proteins with Solanic® potato proteins, specifically, that demonstrate high solubility, good foam overrun, and a

foam firmness equal to or better than egg albumin (Solanic powerpoint). Similar to other protein isolates, pH changes can negatively impact the functionality and solubility of Solanic® products, restricting their possible applications (Solanic powerpoint). Solanic® potato proteins have been found suitable for meat analogues, gluten and/or dairy-free products, pasta, protein beverages, and a variety of other foods (Solanic powerpoint).

### *Advantages*

Potato protein is an added-value ingredient produced from a by-product of starch production. It has superior nutritional quality and functionality compared to other plant protein sources. Additionally, allergenicity to potato protein is rare (Løkra and others 2008).

### *Barriers*

Compared to other potential plant protein sources, potatoes have an extremely low protein concentration. With a protein concentration of 1.5%, using potato fruit juice for protein ingredient production would require an extremely large quantity of potatoes for sizable commercial production of a potato protein ingredient. Establishing a steady potato fruit juice supply stream of this magnitude may present barriers to potato protein ingredient manufacturers. Additionally, potato fruit juice contains polyphenolic compounds that can react with proteins and reduce solubility and digestibility (Waglay and others 2013). Processing methods to remove these compounds without affecting protein solubility and functionality will have to be developed, and will likely increase production costs. Protein isolation processes will also have to be further researched as the acid and thermal treatments that are often used cause protein denaturation (potato protein denature at low temperatures 60-75°C) and reduced functionality (Waglay and others 2013). Commercial potato protein manufacturing currently is limited, and will likely not expand much outside of Europe.

### *Feasibility*

Potato production in Minnesota is feasible, as demonstrated by the high yields and sales for 2016. With the development of effective methods of protein isolation that limit protein denaturation and functionality loss, the commercial production of potato protein may potentially increase. Relatively recent research showed that using processes such as precipitation with  $(\text{NH}_4)_2\text{SO}_4$ ,  $\text{FeCl}_3$ , or ethanol, instead of acid and thermal treatments, results in higher yields and purity, and reduced sensitivity to pH and temperature changes (Wagley and others 2013).

However, despite advances in processing, potatoes have limited potential as a popular and profitable plant protein source. Having very low protein concentrations, the quantity of potatoes needed for commercial manufacturing could be a prohibiting factor. Obtaining the amount of potato fruit juice needed for large-scale potato protein production will likely

deter entrepreneurs and established large-scale manufacturers. Additionally, limited supply is associated with higher ingredient cost compared to other sources.

### *Camelina Protein*

#### *Recent statistics on yields/sales in Minnesota*

Camelina (*Camelina sativa*, a Crucifer seed and a member of the Brassicaceae family) is a sustainable, short-season oilseed cover crop that is high in both oil/fat (30-38%) and protein (25-30%) (Berti and others 2016; Gesch and others 2012). Thus, it is an attractive choice for the production of both oil and protein ingredients. Minnesota camelina production is in its infancy and currently researched by the University of Minnesota and USDA-ARS as part of the Forever Green initiative. The annual yield for camelina grown in Rosemount, Minnesota as part of the initiative is between 980-1070 lbs/acre) when averaged over several years (DuByne 2017); this equated to approximately 20-34 bushels/acre (University of Minnesota 2017b). Camelina yield is influenced by rainfall and soil nitrogen levels, with poor yield resulting from limited rainfall (Smith 2017a; Ibrahim and Habbasha 2015). Although higher soil nitrogen levels positively impact camelina yields, very little fertilizer is actually needed for production (Ibrahim and Habbasha 2015); this fact, along with camelina's natural pathogen-resistance, insect-resistance, and potential to be farmed using equipment for canola and mustard, minimizes camelina production costs (Ibrahim and Habbasha 2015). With low production costs and eligibility under the 2014 Farm Bill's price loss coverage in a number of states (not yet in Minnesota), camelina farming could be a relatively low risk venture (USDA ERS 2017d).

#### *Nutritional quality*

The amino acid composition and hence the nutritional quality of camelina protein is similar to that of canola protein (Li and others 2015), which in turn is comparable to that of soy protein, the gold standard among plant proteins (Sarwar and others 1984). Specifically, camelina protein meets all of the World Health Organization's essential amino acids requirements for those over the age of one. Some antinutritive factors are present in camelina, such as glucosinolates, phytic acid, and tannins, which can negatively affect protein digestibility (Qi and others 2016; Hixson and others 2016). Heat treatment, however, can reduce or eliminate these antinutritive factors (Hixson and others 2016); thereby improving camelina's nutritional value. Breeding may also result in varieties low in antinutritive factors, an effort pursued by the Forever Green Initiative researchers at the University of Minnesota. Specifically, new winter varieties of camelina possess particularly low concentrations of glucosinolates, below those of canola (Murphy 2016).

#### *Protein components*

Camelina proteins constitute mainly albumins (10.5%, water-soluble fraction), globulins (17.7%, salt soluble), and glutelins (64.6%, alkaline soluble) (Ochiai-Yanagi and others

1978). Legumin-type globulins, cruciferin (11S), and napin-type albumins, napin (2S), are the major storage proteins (~80-85% of total seed proteins) of Brassicaceae seeds, including the Crucifer seed, camelina (Wanasundara 2011). Research on the isolation and characterization of camelina protein fractions is limited (Li and others 2014; Li and others 2015).

#### *Currently available protein ingredient forms*

There is no commercial production of camelina protein today. Given that it is a relatively new crop considered for food use, there are few reports on its oil and protein composition (Li and others 2014), structure and functionality. Camelina meal would be a by-product from oil production. The meal is rich in protein (40-45%), thus it is necessary to explore extraction techniques to produce a functional protein ingredient for food applications. A recent study demonstrated that camelina protein isolated following salt extraction resulted in higher protein yields compared to isolates from alkaline extraction (Boyle and other, 2017). The camelina protein concentrate (70-80% protein) produced following salt extraction had better overall functionality.

#### *Potential functionality and applications*

Compared to alkaline pH extraction, salt extraction produced less denatured and more functional camelina protein concentrate (CPC), composed mainly of cruciferin and napin proteins (Boyle and others, 2017). The functionality of the salt extracted CPC was comparable and sometimes better than that of soy protein isolate (SPI). Specifically, the solubility (~70%) of the salt extracted CPC at pH 3.4 was significantly higher than that (~50%) of SPI. Additionally, salt extracted CPC had significantly higher emulsification capacity and foaming capacity than SPI. On the other hand, the gelation property of CPC was inferior to that of SPI, an observation attributed to the molecular size of camelina protein compared to soy protein. This study demonstrated the potential of camelina as a novel source of functional plant protein that might gain a position in the protein market place, and possibly compete with soy protein for several applications targeting the use of plant proteins.

#### *Advantages*

Camelina is highly adaptable and production is possible under a variety of soil and climate conditions (Murphy 2016). Therefore, camelina can be produced in less fertile areas and in a variety of locations and requires little water or fertilizer. Being such a low maintenance crop, camelina is a suitable crop for almost any farmer (Murphy 2016). The environmental benefits of camelina are numerous, including reduced soil and water erosion, reduced soil nitrate leaching, increased carbon sequestration, and reduced inputs of energy and pesticide (Berti and others 2016; Gesch and Archer 2012). These environmental benefits make camelina attractive not only to farmers but to consumers seeking sustainable crops

in their food products. Accordingly, food manufacturers will seek commercializing products formulated with protein ingredients derived from sustainable crops.

### *Barriers*

Being a new crop under development, many areas require further investigation before considering it as a viable choice for protein production. Crop yield, and hence availability for industrial use is a considerable barrier. Additionally, research on the properties of camelina proteins is extremely sparse, making it difficult for manufacturers to optimize isolation and processing methods. Therefore, researching protein isolation, functionalization, and modification is necessary.

### *Feasibility*

Camelina has a great potential for production at a low cost and in a variety of soil and environmental conditions, the only restrictions being heavy clay or organic soil (Murphy 2016). Camelina can also withstand drought conditions better than canola and has a natural resistance to insects, frost, and freeze-thaw cycles (Hixson and others 2016; Ibrahim and Habbasha 2015); this limits the risk of crop loss and makes ventures into camelina production more feasible for farmers. As an added benefit to oilseed farmers, camelina production is possible with the same equipment used for canola and mustard production (DuByne 2017). However, farmers require economic incentive to plant this crop. It is important to find a market value for this crop, such as its utilization as a protein ingredient source. For camelina to be a feasible source for plant protein, more research is necessary on extraction methods, processing technology, functionality, and nutritional quality.

### *Pennycress Protein*

#### *Recent statistics on yields/sales in Minnesota*

Pennycress (*Thlaspi arvense*) is a winter cover crop that has numerous environmental benefits including soil stabilization, nutrient sequestration, and reduced nitrate leaching. Being an oilseed crop, pennycress has high oil/fat (30-40%) and protein content (25-35%). Accordingly it presents an attractive choice for both oil and protein ingredients production. Pennycress yields are variable, with research trials yielding 1,500 to 2,000 lbs/acre while farm production averages around 700 to 900 lbs/acre (Smith 2017b). In Minnesota, pennycress production is limited as farmers are unfamiliar with the crop and currently there is no commercial application for pennycress in the U.S. (Smith 2017b). However, the University of Minnesota and USDA-ARS are currently researching pennycress as part of their Forever Green Initiative. The research efforts may lead to increased production and utilization (University of Minnesota 2017a).

### *Nutritional quality*

Pennycress seeds contain around 20-27% protein, while defatted pennycress meal contains about 40% protein (Hojilla-Evangelista and others 2014). The defatted meal, a potential by-product of oil pressing, can thus be an excellent starting material for the production of protein isolates. Research on pennycress protein digestibility is lacking. The amino acid composition and content of essential amino acids are similar to those of soy protein isolate (Hojilla-Evangelista and others 2014). However, wild type pennycress seed is naturally high in glucosinolates (Warwick and others, 2002), a class of potentially toxic secondary metabolites (Sedbrook and others, 2014), and is high in erucic acid (Evangelista and others, 2012), a fatty acid associated with adverse health effects (Knutsen and others, 2016). Recent advances guided by sequencing the pennycress genome (Dorn and others, 2013) and assembling its transcriptome (Dorn and others, 2015) have aided in the identification of new lines of pennycress lacking erucic acid. Additional screening has identified candidate lines of pennycress that reduced seed glucosinolates.

### *Protein components*

Similar to canola and camelina, pennycress proteins are mainly composed of albumins and globulins. The protein components are mostly of low molecular weight (Hojilla-Evangelista and others, 2015) compared to those of soy protein. Research on the pennycress protein components and their functionality is scarce.

### *Currently available protein ingredient forms*

Mainly processed for its oil, pennycress has potential for biodiesel applications, while the protein-rich meal is a by-product (Hojilla-Evangelista and others 2014). Interest in pennycress as a potential source for protein ingredients is relatively recent and under development. Accordingly, commercially available products are non-existent at the present time.

### *Potential functionality and applications*

Research on the functional properties of pennycress proteins is limited. Hojilla-Evangelista and others (2014) found that pennycress proteins might have potential as foaming and emulsifying agents. Pennycress protein had a superior foaming capacity and stability compared to soy protein, suggesting that pennycress could replace soy in some foods (Hojilla-Evangelista and others 2013). Hojilla-Evangelista and others (2015) determined that protein isolated following salt extraction vs. alkaline extraction methods had superior solubility and emulsification properties. Pennycress protein functionality and potential applications in comparison to other protein ingredients requires further investigations.

### *Advantages*

Pennycress is well suited as a cover crop; as it limits soil erosion, weed growth, and nutrient loss, benefitting farmers long-term through a reduced need for herbicides and soil fortification (University of Minnesota 2017a). Pennycress may integrate well with conventional corn/soybean and summer cropping systems. Additionally, pennycress is of nascent interest, thus market competition is low. The demand to utilize pennycress as a novel source of plant protein has the potential to rise. Those that enter the market soon could obtain an economic edge before pennycress garners the attention of others.

### *Barriers*

Pennycress is relatively unknown to the public, which could delay its use and profitability. This lack of familiarity is associated with limited research on the properties of pennycress proteins, its functionality and potential uses. Other barriers include the perception that pennycress is high in glucosinolates, which may remain with the protein extract. However, through selective breeding, new pennycress lines with much reduced glucosinolates are developing, and thus may serve as an ideal source for protein isolates. On the other hand, there isn't an established processing method, of functionalization, for pennycress protein isolates.

### *Feasibility*

Based on the research conducted as part of the University of Minnesota's Forever Green Initiative, growing pennycress in Minnesota is not only feasible, but also offers benefits, including limiting weed growth, which reduces herbicide needs (University of Minnesota 2017a). Low herbicide, as well as, low energy input and labor requirements make pennycress economically feasible for many farmers (Smith 2017b). Factors that keep the monetary risk of growing pennycress low, such as its eligibility for price loss coverage under the 2014 Farm Bill and minimal input and labor requirements, may also increase production (USDA ERS 2017d; Smith 2017b). Research that proves the benefits of pennycress as a source of oil and protein ingredients will provide economic incentives to farmers to increase production, allowing for sufficient supply needed by food processors.

## **Areas Requiring Further Investigation**

Consumers are seeking a diet high in plant proteins that are non-allergenic and non-GM, and producers are intensifying efforts to respond to this need. However, the interest in and demand for a variety of plant proteins are increasing at a much faster pace than the research on the nutritional, physiological, and functional contributions of these proteins in various food applications. While there has been some research done to characterize plant proteins, the information is far from being comprehensive. In order to understand how plant proteins can deliver optimal nutrition and functionality in various food products, a plethora of questions need answers. There is still a need to fully characterize the effect on

structure/function, relationship and quality of each protein by various extraction and processing technologies. At the same time, the industry must determine feasible means of protein extraction and valorization of waste streams. Additionally, given the inferior functionality of plant proteins in general, it is necessary to research functionalization and protein modification methods for targeted applications. Characterizing the impact of processing and functionalization on the protein’s nutritional and physiological quality is equally important. Adopting a new protein source will also need regulatory approval, thus requiring several analytical data to be collected. Last but not least, understanding interactions of plant proteins with flavor compounds in different systems and finding ways to mask plant protein off flavor or reduce undesirable reactions to enhance products’ acceptability are also crucial.

## Summary

This report provided a summary of, and when possible, a comparison among various plant proteins that are at different stages of development (Table 1). While some proteins could potentially have better nutritional and functional properties than others could, each protein may have unique applications, and upon blending with other plant and/or animal proteins may provide complete nutrition. Further research and development may transform emerging and potentially viable protein sources into marketable protein ingredients.

Table 1. Comparison of different plant proteins

Protein Source	Nutritional Quality	PDCAAS Level	Available Ingredients	Functionality	Applications	Development Stage	Potential
Soy	Excellent	0.9-1	Flour, concentrates, isolates, hydrolysates	Very good	Meat analogues, baked goods, beverages, frozen desserts, salad dressing cereal, meat products	Developed	Widely used ingredient; limitations: GM, allergenic
Pea	Good	0.8-0.9	Flour, concentrates, isolates, hydrolysates	Acceptable, but inferior to soy protein	Extruded snacks, baked goods, meat extenders, beverages, confectionary	In development, require functionalization	Good, low allergy incidences, non-GM
Canola	Comparable to soy	0.9-1	Limited availability; isolates produced by Burcon	Good	Meat products, beverages, baked goods	Early stages of development	Very good
Other Pulses	Moderate	0.5-0.7	Flour, concentrates, and isolates	Similar to pea protein	Limited applications; used as meat extenders	Very early stages of development; require functionalization	Moderate
Hemp	Moderate	0.6-0.7	Limited availability of defatted flour, concentrates, isolates	Moderate	Limited applications;	Early stages of development	Moderate
Sunflower	Moderate	0.6	Sunflower protein powders (50% protein)	Potentially good, more research is needed	Limited applications	Very early stages of development	Good
Corn	Low	0.4	Corn gluten meal for animal feed	Poor	None	Very early stages of development; require functionalization	Poor
Oats	Low	0.4-0.5	Protein concentrate; e.g. ProDatein (54% protein)	Moderate	breakfast cereals and bars, breads, nutritional shakes, and healthy snacks	Early stages of development	Moderate



Potato	Excellent	0.93	Limited; Isolates; e.g Solanic	Good solubility and overall functionality	meat analogues, gluten and/or dairy-free products, pasta, protein beverages	Early stages of development	Moderate: due to low protein concentration in potatoes
Camelina	Needs investigation	Not available	Not available	Moderate	None	Not developed	Good, because of environmental benefits and high protein content
Pennycress	Needs investigation	Not available	Not available	Moderate	None	Not developed	Good, because of environmental benefits and high protein content

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