2023

Extraction, Characterization, And Utilization of Yellow Pea Starch

Abdulmalik Albu Tuwaybah
South Dakota State University, Abdulmalik.albutuwaybah@jacks.sdstate.edu

EXTRACTION, CHARACTERIZATION, AND UTILIZATION OF YELLOW PEA STARCH

BY

ABDULMALIK ALBU TUWAYBAH

A thesis submitted in partial fulfillment of the requirement for

Major in Biological Sciences

Specialization in Food Science

South Dakota State University

2023

THESIS ACCEPTANCE PAGE
ABDULMALIK ALBU TUWAYBAH

This thesis is approved as a creditable and independent investigation by a candidate for the master's degree and is acceptable for meeting the thesis requirements for this degree. Acceptance of this does not imply that the conclusions reached by the candidate are necessarily the conclusions of the major department.

Clifford Hall
Advisor Date

Sanjeev Anand
Department Head Date

Nicole Lounsbery, PhD
Director, Graduate School Date

This thesis is dedicated to my father's soul (Fahad Albu Tuwaybah). Also, for my mother (Ebtisam Al Hamadan) and my siblings who without their love and support this would not happen.

ACKNOWLEDGEMENTS

First and foremost, I would like to express my sincere thanks to my advisor, Dr. Clifford Hall, for his continuous support and guidance during my M.S. study and research, as well as his inspiration, passion, patience, and valuable knowledge. I am glad to have Dr. Hall as my advisor. With his guidance, motivation, and profession, I had the chance to improve my research, connection, management, and leadership skills. Also, I would like to thank my thesis committee: thank you, Dr. Srinivas Janaswamy, for his support, encouragement, and valuable feedback and conversation. Thank you, Dr. Douglas Raynie, for opening his lab and his guidance and insightful comments with Supercritical Fluid Extraction.

I would like to thank Dr. Serap Vatansever for her support with the beginning of my master journey. I would like also, thank Mahmoud Hamouda for helping with the food application preparation. My thanks also, for my previous and new lab mates, Sushmita Karki, Dana Edleman, Hussain Al Naser, Shirin Kazemzadeh Pournaki, Jessica Christoffer, Jeffery Lehman, Reedlyn Sperlich, Prakriti Dhakal, Mastaneh Shokri and Hojjat Abdollahikhamene for their friendship and help throughout this journey. Your comments, questions and corporation have made this journey more valuable and enjoyable.

My sincere gratitude also goes to the Department of Dairy and Food Science and everyone who works there: thank you for making the department a nice place to work.

I would like to acknowledge SACAM program, the Saudi Ministry of Education and The Ministry of Environment Water & Agriculture for awarding me the scholarship to support my study in The United States.

CONTENTS

ABBREVIATIONS

mm	millimeter
sd	standard deviation
se	standard error
M	molar
conc	concentration
h	hour
min	minute
SC-CO_2	supercritical carbon dioxide
SFE	supercritical fluid extraction
ETS	ethanol treated starch
LSE	lab scale starch extraction
LSE-SFE	lab scale starch extraction-supercritical fluid extraction
ISE	industrial scale starch extraction
ISE-SFE	industrial scale starch extraction- supercritical fluid extraction
WPS	whole pea seed
WPF	whole pea flour fraction (100 mesh size)
DPS	dehulled pea seed
DPF	dehulled pea flour fraction (100 mesh size)

LIST OF FIGURES

LIST OF TABLES

ABSTRACT

MODIFICATION, CHARACTERIZATION, AND UTILIZATION OF YELLOW PEA STARCH

ABDULMALIK ALBU TUWAYBAH

2023

Dry peas, a leguminous crop, are one of the world's oldest crops, accounting for 35–40% of total pulse trade. Pea starch is an inexpensive source of starch since it can be obtained as a by-product of protein extraction. Recovering the starch from protein isolation streams (i.e., before or after) improves the environmental impact of the protein isolation process. The goal of the study was to transform low-value pea starch into high-value starch through starch extraction and food application. Two methods of starch extraction were used (47.5% ethanol treatment and SC CO_2 + EtOH extraction). Starch samples were analyzed for chemical, physical, functional, and pasting properties. All starch treatments were applied to the food product (yogurt and pudding), and syneresis and hardness were determined. The obtained results illustrated that the isolation process from dehulled seed led to significantly higher starch purity (1.42% protein, 0.32% ash, and 93% starch content (d.w.b.)) and starch recovery (79%), compared to whole pea seed (WPS), whole pea flour fraction (WPF), and dehulled pea flour fraction (DPF). After isolation of starch, SC-CO_2 + EtOH extraction was done on dehulled pea seed (DPS) (the selected sample). A significant ($p \leq 0.05$) reduction of moisture, starch, and fat contents and peak and trough viscosity with an increase in setback viscosity in all samples after the extraction with SC CO_2 + EtOH. On the other hand, ethanol treatment significantly increased ($p \leq 0.05$) the total starch from (88.51% to 90%), starch damage (0.52 %) and water holding capacity (0.96 g/g). Physical results of starch-added yogurt and pudding

showed an increase in the syneresis with the storage time. Also, there was a positive correlation between syneresis and hardness value in food products. To conclude, dehulling prior to starch isolation is a suitable method of pea starch extraction. Also, modified pea starch impacted the texture and water holding capacity in food products.

1. INTRODUCTION

1.1. General Introduction

Pulses, which is based on the word puls in Latin, means the thick soup. They are an ancient plant species having health, nutritional and environmental benefits. Pulses, including dry peas, have been around for thousands of years and grown in a wide range of climates and conditions such as extreme hot climates and very wintry weather conditions (FAO, 2016). Dry peas are found in different shapes, sizes, and colors, and grown in a wide range of soil types and are known as smooth or field peas. Garden peas (*Pisum sativum* ssp. hortense) and field peas (*Pisum Sativum* L.) are the two main types of the cultivated peas. These two types have different starch characteristics and granular morphologies. Pulses are one of the few foods that is considered by the United States Department of Agriculture (USDA) as both a vegetable and a protein resource. Among pulses, dry peas, the dehydrated edible seeds of leguminous crops, are used for human food and animal feed (Ratnayake & Warkentin, 2002). They are high in complex carbohydrates (55-72%), protein (14-31%), and dietary fibers (3-20%) (Hall, 2021).

Besides nutrient denseness that dry peas have, they are good for the environment and are economical. Globally, Pea (*Pisum Sativum L.*) is a widely grown pulse accounting to 8 to 14% of the total production of pulse crops around the world (Joshi & Rao, 2017). The harvesting of dry pea is estimated to be 6.7 million hectares worldwide. In the U.S.A, dry peas are mostly grown in the northern states such as Palouse region of Washington, Idaho, Montana, and North and South Dakota. According to the USDA National Agricultural Statistics Service and the U.S. Dry Pea and Lentil Council, pulse harvested

acres and total output predicted for 2021 were 1.73 million and 700,000 MT, respectively (Hall, 2021).

Canada, on the other hand, is the largest producer and accounts for approximately 25% of the total world productions and 40% of total world exports of field peas. The largest use as human food is in Asia and South America, where whole or split seeds are boiled and then eaten. However, Europe and North America are the largest users of field peas for animal feed, where whole seed is ground and mixed with ground cereal seeds to produce feed (Dahl et al., 2012; Ratnayake & Warkentin, 2002).

Pulses provide energy (carbohydrate), dietary fiber, protein and vitamins and minerals that are needed for human health. Regular consumption of pulses may potentially enhance a person's health by lowering cardiovascular diseases, cancer, diabetes, osteoporosis, hypertension, gastrointestinal disorders, adrenal illness, and LDL cholesterol risks (Jacobs & Gallaher, 2004; Mizelman et al., 2020). In addition to their health benefits, pulse proteins have special functional qualities that could increase their application in the creation of a wide range of food products. They may be used as a substitute for top allergies such as soybean, gluten, dairy, eggs, and nuts. Pulses have been applied as ingredients in baby food, imitation milks, meat products, baked goods, extruded products, pastas, and noodles (Boye et al., 2010). In fact, their utilization is not limited only to protein but also starch and dietary fiber sources.

Extraction of protein as food ingredient has been extensively investigated and used in the protein industry. However, research on the extraction of starch and its functional characteristics and food application is needed. For what, the extraction of starch from

lentil, chickpea, and dry pea had reached 40.5 million tonnes in 2006-2007 (Hoover et al., 2010). Due to the presence of insoluble protein and highly hydrated fine fiber, it is difficult to separate the starch from the seed. Practically, pulse starch can be obtained using dry milling or wet milling approaches. The alkaline steeping method is one of the commonly used isolation techniques by starch industry (Sun et al., 2015). Modification of starch, to increase the viscosity, the resistance to mechanical shearing, acid hydrolysis, high temperatures, and/or enzyme hydrolysis and to reduce the degree and rate of retrogradation, has been done chemically or physically in the food industry (Ratnayake & Warkentin, 2002).

Dry peas have been recognized as a protein source by governmental agencies and by the food industry. Extensive research on the isolation and utilization of pea protein has been completed (Boye et al., 2010); nonetheless, information regarding extraction, modification and food application of pea starch is needed. Moreover, pea starch research regarding the characteristics and functionality is limited compared to cereal and tuber starches. Therefore, this study was conducted to 1) optimize pea starch isolation method, 2) subject starch to different extraction including supercritical carbon dioxide and ethanol treatment, and 3) determine the impact of pea starch in the functionality of food application (yogurt and pudding).

1.2. Research Objectives

The goal of this study was to transform pea starch, a low-value co-product into a high-value product through an examination of the impact of supercritical technology and ethanol treatment on the functional and pasting properties and a demonstration of starch in food system. This information will be necessary to extend its utilization and provide

important information about its functionalities, added nutrition, and customer acceptability of new products.

1.3. Hypothesis

We hypothesis that dehulling of yellow dry pea will increase the starch yield and purity. The different isolation methods and the modification process of the starch will have an impact on some of the chemical property and most of the functional properties of the starch. Modified pea starch will facilitate result in less syneresis in the food application of yogurt and pudding.

2. LITERATURE REVIEW

2.1. Introduction to Pulses and Dry Peas

The consumption of pulses has been increased worldwide due to the ease of storage and handling (dry seed) and their nutrient denseness. Pulses are rich in protein, carbohydrates, dietary fiber, vitamins and minerals, and other bioactive substances such as enzyme inhibitors, lectin, phytates, oligosaccharides, and phenolic compounds (Boye et al., 2010; Campos-Vega et al., 2010; Hall, 2021). Frequent pulse consumption, such as dry peas, chickpea, lentil, beans, can help to reduce the risk of coronary heart disease (CHD) and cardiovascular disease (CVD) (Campos-Vega et al., 2010).

According to the USDA National Agricultural Statistics Service, the predicted total production of pulses for 2021 was 1.73 million acres, or 668,466 MT In the United States concentrated on the Northern Plains region and Pacific Northwest region (Hall, 2021). Dry Pea (*Pisum Sativum* L.), when compared to other pulse crops in terms of overall harvesting, is one of the most widely grown in the world. Dry pea is a cool season

legume crops with a total production of 10.3 million metric tonnes worldwide for human or livestock food (Simsek et al., 2009). Eight to 14.6% of the total world production is grown in Canada, France, and China, followed by Russia, India, and the U.S.A (Joshi & Rao, 2017). More than 13.5 million tons of dried peas were produced globally in 2018, with Canada, Russia, and China serving as the top three producers (FAOSTAT 2020).

Historically, dry peas were grown mostly in the Palouse region of Washington and Idaho in the United States. In 1990, North Dakota and Montana started production of dry peas. In 1991, approximately 647 hectares of dry peas were planted in North Dakota, and 247,000 hectares, or 66% of the US production, were grown there in 2006. Furthermore, the US raised over 1.8 million hectares and more than 70% of the production was exported for food and feed processing to countries including India, China, and Spain (Simsek et al., 2009). Moreover, the total dry pea production of the United States in 2021 was 387,780 metric tons (Hall, 2021).

2.2. Economic Importance of Dry Peas

Pulses grow better in cool, dry seasons; therefore, production in western and northern North Dakota and eastern Montana is ideal. As a result, pulse planting has the potential to deliver significant financial and environmental advantages to these places. Prices for North Dakota's dominant crops, such as soybeans, corn, and wheat, are declining, implying that pulses may provide better profits than traditional crops. In addition to this, a rise in pulse production has an impact on the economy of North Dakota and eastern Montana, from farm-level production through final product processing (Coon et al., 2015). Coon found these economic implications to be extremely large. In 2015, North Dakota received $115.7 million in pulse-related expenditures, including sales and

personal income, with field pea accounting for 67.8% of this total. Profits from the transportation and processing of pulses are included in this value. Furthermore, a growth in the usage of pulse flour as an ingredient may generate chances for the development or establishment of new milling and processing facilities.

2.2.1. Food Industries

Yellow pea flour has been used as a replacement (partial and 100% replacement) for wheat flour in gluten-free goods such as pasta, biscuit (Zhao et al., 2019), bread (Bourré et al., 2019), cookies, cake, crackers, and other gluten free products (Gohl, 2019; Hillen, 2016). Pea flour has a high protein percentage, which may help to address the functional or structural issues seen in gluten-free alternatives. Because gluten-containing components cannot be utilized in bakery product and gluten substitutes are limited, high quality protein content is critical to the quality of gluten-free food items (Patil et al., 2016)

As a fractionated ingredient, pea starch, which is the major component of dry pea, is used to modify food product texture and water holding capacity of the product as well as (Simsek et al., 2009) to produce edible films that are economical, environmentally friendly, non-toxic, biocompatible, and have similar qualities as synthetic polymers. Food texture is important not only for processing but also for consumer acceptance. Extensive research has been done on common starch; however, limited studies exist on pea starch properties (Simsek et al., 2009). Pea starch properties include being odorless, tasteless, transparent, semi-permeable to CO_2, and resistant to O_2 diffusion. Apart from this, pea starch has a higher amylose content than other typical starches, which has been found to make films with better physical and mechanical qualities (Saberi et al., 2016). Pea starch

was used to improve the quality attributes of food products such bread (J. H. Li & Vasanthan, 2003; Z. H. Lu et al., 2018a; N. Wang et al., 2012).

Pea protein fractionation from the seed have been extensively studied (Shand et al., 2007). Research has been carried out to examine the formulation of novel foods and drinks, such as bread (Espinosa-Ramírez et al., 2018), pasta (Linares-García et al., 2019), meat products (Baugreet et al., 2018), baked goods (Gularte et al., 2012), snacks (Morales-Polanco et al., 2017) and beverages, and demonstrate the potential of pea protein to enhance nutritional and functional properties of the products. Additionally, pea components can be used to make gluten-free products.

Due to growing interest in fiber fortification and extending the shelf life of food goods, pea fiber obtained from recovered pea hulls, split pea processing, or protein separation is becoming more popular globally. It can be regarded as the finest option for creating low-carb and low-calorie food products, such as bread, snack foods, biscuits, crackers, pasta, tortillas, and nutritional supplements (Damian and Olteanu, 2014). Yogurt with high viscosity and less syneresis compared to control were discovered when pea fiber was added to the yogurt-making process (Damian and Olteanu, 2014) and used to create fiber-rich instant pea soup powder, which improves the soup's functional qualities while also making excellent use of a byproduct (Hanan et al., 2020).

2.2.2. Feed Industries

When used as animal or cattle feed, dry peas are an energy and protein-rich pulse that is equivalent to other feeds including barley, maize, canola meal, and sunflower meal. Dry peas are reportedly a great supplement and palatable feed item for beef cattle, dairy cattle, poultry, pigs, and sheep (Anderson et al., 2014). There were no deleterious effects

from pea intake in lactating dairy cows fed ground dry peas instead of soybean meal and maize grain. The milk production or composition were unchanged by the ground peas (Vander Pol et al., 2008). Dry peas might also be used as a feed for fish. It has been effectively utilized to replace wheat in seabass feed, and there has been no difference in the fish's ability to develop, the quality of their carcasses, or their organoleptic characteristics (Adamidou et al. 2009). Moreover, studies on the consumption of dried peas in the diet of blue shrimp provided evidence that they are a viable and acceptable element in shrimp feed (Cruz-Suarez et al., 2001).

2.3. Environmental Impact

In addition to financial rewards, pulses could improve soil health in a field. Pulses such as field peas and dry beans are commonly utilized as break crops in North Dakota (Kirkegaard et al., 2008). Break crops have environmental advantages such as improved disease management, increased soil nitrogen levels, and reduced water use. The increased nitrogen supply from legumes eliminates the requirement for fertilizers derived from nonrenewable energy sources (Smith & Chalk, 2020). Moreover, Kirkegaard et al. (2008) observed that including break crops such as pulses into a field resulted in a 20% or greater production boost for wheat sown the following season. Undoubtably, increased use of pulses in food products increases demand for land, benefiting the economics and environment of pulse-growing regions such as North Dakota.

2.4. Classification Of Pulses and Dry Pea

Pulses are one of the few foods that is considered both a vegetable and a protein resource by the U.S. Department of Agriculture (USDA). There are 11 different types of pulses including dry beans, dry broad beans, dry peas, chickpeas, cow peas, pigeon peas, lentils, Bambara beans, vetches, lupins and puls. They grow in pods and come in different shapes, sizes, and colors. According to the Food and Agriculture Organization (FAO), "pulse" refers to crops that are only harvested for dry grain. Therefore, legumes used to extract oil, such soybeans and peanuts, are not considered pulses (Rebello et al., 2014). Specifically, dry pea also known as the field pea or smooth pea (*Pisum Sativum* L.), makes up 8 to 14.6% of the world's total production of pulses (Joshi & Rao, 2017). However, a fresh pea, which is normally marketed as a fresh or canned vegetable for human consumption, is different from a field pea with ideal growth temperatures between 13 °C (55 °F) to 18 °C (65 °F). Field pea is often well suited to cool semi-arid areas. The main market classes are the green and yellow cotyledon types (NDSU, 2021).

2.5. Nutritional Composition of Dry Peas

Many people struggle to maintain an ideal diet for their daily lives. Pulses, including dry pea, provide the key solution for a healthy diet due to their low-fat, high protein, complex carbohydrates, fibers, vitamin, and mineral contents, and are an excellent source of antioxidants (FAO, 2016). Dry peas contain (14-31%) protein, (55-72%) total carbohydrates, which includes starch (30-49%) and total dietary fibers (3-20%) (Hall, 2021).

2.5.1. Carbohydrate

Starch, the major component of field peas, ranges from 36.9 to 49.0% of the total composition and found to be significantly impacted by the environmental and cultivar factors (Dahl et al., 2012; N. Wang & Daun, 2006). Starch is a major source of energy and an important raw material for industrial applications (Boye et al., 2010; Hall, 2020). Starch is made up of two different types of glucan polymers (i.e., amylose and amylopectin). Amylopectin, which is more complicated than amylose, is a highly branched molecule of α-(1-4)-linked glucopyranosyl units in a chain that are joined by α-1,6 glycosidic bonds. Unlike amylopectin, amylose is a straightforward linear glucan molecule with α-(1-4)-linked D-glucose units (Vanier et al., 2017).

Pea starch content differs depend on the cultivar, growing region, and year. For example, the starch content showed a significant interaction relationship between cultivar-by-year and cultivar-by-location. The Cooper and Cutlass varieties were found to have the greatest starch content, while the CDC striker had the lowest (Wang et al., 2010). This content of starch consists of 35–65% amylose and with remaining of amylopectin (Zhou et al., 2019). When talking about the market class peas, green and yellow peas have mean starch content of 44.1% 43.4%, respectively. Additionally, among the green peas, Ariel had the highest starch content at 46.3%, while DL Apollo had the highest starch content at 46.4% among the yellow peas (Hall, 2019).

Resistant starch is a part of the overall amount of starch. Resistant starch can be utilized as a functional fiber since it cannot be broken down by mammalian enzymes (Sajilata, Singhal, & Kulkarni, 2006). Pea starch contain around 2.45% resistant starch.

However, harsh treatment, including cooking, of the starch reduces the resistant starch content compared to raw starch (De Almeida Costa et al., 2006).

Dietary fiber is defined by the Food and Drug Administration (FDA) as the "intrinsic and intact non-digestible soluble and insoluble carbohydrates and lignin in plants" (FDA, 2018). Dry peas contain 0.6-3.7% soluble fiber, which slow the absorption of lipids in the human body, and lower blood cholesterol, and 8.7–12.9% insoluble fiber, which aid in maintaining regularity and help prevent gastrointestinal problems (Stoughton-Ens et al., 2010). Dietary fiber, in form of seed coat such as hull or cotyledon, is composed of cellulose, hemicellulose, pectins, hydrocolloids, and lignin. Dry pea dietary fiber content varies from 14 to 26% (Brummer et al., 2015; Coon et al., 2015).

2.5.2. Protein

Pea protein, the second most prevalent component of the seed, is a high-quality plant-based protein source. The protein content ranges from 24 to 31% depending on the cultivar (Boukid et al., 2021). Protein is a macronutrient composed of amino acids that are bonded together in long chains. The protein bulk in legumes is composed of globulins, albumins and prolamins. Globulins, which is soluble in salt solution and the storage protein during seed development, accounts for 55 to 65% of the total protein. Albumins make up most of the remaining proteins ranging from 18-25% of the dried pea seed. Albumins provide functions within the seed, such as lectins and lipoxygenases. Prolamin and glutelin are small proteins found in lesser concentrations, i.e., 4-5% and 3-4%, respectively (Z. X. Lu et al., 2020; Vatansever, Tulbek, et al., 2020). Dry pea protein provides a high proportion of lysine, and amino acid that is lacking in cereal grains.

Globulin is rich in arginine, phenylalanine, leucine, and isoleucine whereas albumin consists of a high amount of tryptophan, lysine, and threonine. However, dry peas are poor in sulfur-containing amino acids such as methionine and cysteine (Vatansever, Tulbek, et al., 2020). When cereal grains and pulses are mixed, they can produce a complimentary amino acid profile or a complete protein (Awika, Rose, & Simsek, 2018). As a result, pea proteins are gaining popularity across the world due to those nutritional and health advantages, as well as their affordability, sustainability, and availability. Pea proteins are less prone to promoting allergic reactions and have a high digestibility when compared to other plant proteins (Z. X. Lu et al., 2020). Furthermore, proteins serve to construct and repair cells and bodily tissue, as well as to supply energy to the human body (Rennie, 2005). In addition to this, proteins provide functional properties relating to gelation, emulsifying, and foaming behavior (Wang & Arntfield, 2016).

2.5.3. Lipid

Lipid content of dry pea is low, ranging from 0.6% to 3.9% (Vatansever, Tulbek, et al., 2020). There is limited research on the pea lipid content since the focus on dry pea has been placed on their major components (carbohydrate, protein, and fiber). Phospholipids (52.2 to 61.3%) and triacylglycerides (31.2 to 40.3%) are the major components of dry pea lipid. Other minor compounds including diacylglycerols (2-4%), free fatty acids (1.3-2.7%), steryl esters (0.8-2.4%), and hydrocarbons (0.5-0.9%) (Yoshida et al., 2007). The fatty acids of dry pea are divided as saturated (15 to 20%), monounsaturated (27 to 37%), and polyunsaturated (42 to 57%). The most saturated lipids are palmitic and steric, with oleic and linoleic being the predominant monounsaturated and polyunsaturated lipids, respectively (Villalobos, Patel, Orstat, Singh, & Lefsrud, 2013). The off flavors found in

pulses are hypothesized to be produced by the breakdown of polyunsaturated lipids (Roland et al., 2017). Nonetheless, these polyunsaturated fats are beneficial to human health. Linoleic and oleic lipids both enhance cardiovascular health by raising HDL levels and reducing LDL levels (Akoh & Min, 2008).

2.5.4. Other Minor Components

Vitamins, minerals, and bioactive substances are essential minor components in dried peas. Ash, which is the indicator of minerals, ranged from 2.3 to 3.0, with a mean of 2.6% in dry peas (Hall, 2021). Dahl et al. (2012) discovered that dry pea seeds have a high concentration of potassium (1.04% on a dry weight basis). Other elements found in dried peas include phosphorus (0.39%), magnesium (0.10%), and calcium (0.08%). Furthermore, field peas produced in the United States are a good source of iron (46-54 mg kg-1), zinc (39-63 mg kg-1) and magnesium (1350-1427 mg kg-1) (Amarakoon et al., 2012). Dry peas also include bioactive substances such as phenolic compounds, oligosaccharides, saponins, phytate, enzyme inhibitors, and lectins.

Pulses contain essential vitamins and minerals in addition to macronutrients. Vitamins found in dry peas include vitamin B- Folic acid, often known as folate, is an important dietary component necessary for the formation of red and white blood cells as well as digestive epithelial cells. The folate content in yellow peas ranges from 23.7 to 55.6 µg/100 g and from 24.9 to 64.8 µg/100 g in green peas (Han and Tyler 2003). In addition, field pea is a good source of Fe, Zn, Mg, and Se. Dueñas et al., (2004) discovered phenolic chemicals in the field pea seed coat and cotyledon. Phenolic chemicals are bioactive plant molecules that function as natural antioxidants.

Antioxidants have been offered as a means of preventing illnesses caused by free radicals. The composition of field pea, ranging from macromolecules to micronutrients, further enhances the benefits of incorporating pea into new food items.

2.6. Health Benefit of Dry Peas

2.6.1. General Health Benefits

Pulses' protein, dietary fiber, and mineral content are beneficial to human health. The USDA recommends a 12-cup dose of beans and peas per week for a 2,000-calorie diet (USDA, 2015). This recommendation is greater for vegetarians and vegans to guarantee appropriate protein consumption. Legume proteins are deficient in the important amino acid methionine (Galili & Amir, 2013). Combining them with a secondary incomplete protein, such as a cereal grain, can give full protein combinations. Owning to the contribution that pulse can make in human diet, 2016 has been declared the international year of pulses (Joshi & Rao, 2017).

When consumed appropriately, legumes are high in dietary fiber and low in fat content, giving cardiovascular benefits (Dahl et al., 2012). Furthermore, dietary fiber of pulse can promote gut health. Many people suffer from constipation, which causes mild to severe pain. Dahl, Whiting, Healey, Zello, and Hildebrandt (2003) offered 3 to 4 items containing 1-3 g of pea hull fiber to patients in an elderly home each day, resulting in substantial improvements in bowel movement and a reduction in the quantity of prune-based laxatives required for each patient. Flogan and Dahl (2010) discovered that snack items supplemented with pea hull fiber in conjunction with inulin fiber supplements

enhanced bowel movement frequency in young children suffering from constipation in a similar trial.

Consuming dietary fiber has been associated with a reduction in the risk of cancer and heart disease (Mckee & Latner, 2000). The daily fiber recommended by health experts is 20 - 30 grams per day for most people according to The Food and Drug Administration (FDA). One-half cup of peas provides 40%, or 10 grams, of the daily recommended 25 grams of dietary fiber (based on a 2000-calories diet). According to American Dietetic Association (ADA), the ordinary American eats only about 11 grams of fiber a day. The most consumed grains, fruits, and vegetables, contain 1 to 3 grams of dietary fiber (USA Pulses, 2018).

2.6.2. Glycemic Index

The low glycemic index of pulses is a significant advantage. The glycemic index (GI) measures the effect of a carbohydrate on blood sugar or glucose levels (Singh et al., 2021). The index is based on a scale of 0 to 100. Foods with a high GI digest swiftly and elevate glucose levels quickly while low GI meals are digested over a longer period of time, resulting in smaller variations in blood glucose and insulin levels. The starch and fiber composition of the pea is suggested to contribute to the field pea's low glycemic index (Trinidad, Mallilin, Loyola, Sagum, & Encabo, 2009).

The rate of diabetes is rising alarmingly in the US. According to the CDC's National Diabetes Statistics Report for 2020, there are expected to be 34.2 million cases of diabetes worldwide (CDC, 2020). According to American Diabetes Association, type 2 diabetes is characterized by insulin resistance, which occurs when the body is unable to generate enough insulin to manage blood glucose levels (Johnson et al., 2019) (American

Diabetes Association, 2019). Unlike Type 1 diabetes, which develops at a young age due to an immune system malfunction that kills insulin-producing cells, Type 2 diabetes is avoidable. Type 2 diabetes is frequently managed with a mix of lifestyle modifications, oral medicines, insulin, or a combination of the three. Low-glycemic meals are frequently recommended as a Type 2 diabetes prevention or therapy (CDC, 2020).

The GI of whole yellow pea flour used as a functional component in the production of pasta, banana bread, and biscotti was compared to whole wheat flour. The findings support the use of dried peas in the production of low GI goods (Barber et al. 2017). The glycemic response of yellow pea flour, pea starch, and maize starch was evaluated directly on these constituents, without their inclusion into food products. Yellow pea flour and pea starch were discovered to have a lower glycemic response than maize starch (Seewi et al., 1999). The content of dietary fibers may have induced differences in GI, with high levels of dietary fiber causing lower GI and vice versa (Trinidad et al. 2010).

2.6.3. Cardiovascular Disease

Fiber-rich diets have been shown to help decrease blood pressure, enhance serum cholesterol levels, and reduce inflammation. Epidemiological research suggests that eating beans four times or more per week reduces the incidence of coronary heart disease and cardiovascular disease (Bazzano et al. 2001). The presence of antioxidant components in the pulses may have aided in lowering cardiometabolic risk. Furthermore, the presence of folic acid in pulses lowers homocysteine levels, which helps to lower the risk of stroke (Rebello et al., 2014).

2.6.4. Obesity

Obesity is responsible for many illnesses like heart disease, cancer, and diabetes (Durstine et al., 2013). According to Rebello et al. (2014), pulse consumption may alter satiety, which can help consumers overcome environmental cues to eat or comply with calorie restriction. Peas are high in protein, micronutrients and low in fat making it excellent whole food for combating obesity-related non-communicable diseases. For example, a meal containing lentils and yellow peas lowered hunger and energy consumption. Consumption of bread with added pea fiber increased the duration of satiety compared to conventional bread (Lunde et al., 2011). A consistent inverse connection between pulse intake and BMI or obesity risk has observed (Shahwar et al., 2018). In addition, prebiotic-rich foods, as peas, modify the microbial colonies in the human gut, improving satiety, regulating intestinal motility, producing short-chain fatty acids, preventing diarrhea and constipation, and reducing pathogen colonization (Mollard et al., 2012).

2.7. Physical Properties of Dry Peas

2.7.1. Swelling Power and Solubility

When starch is heated in the presence of excess water, the hydrogen bonds within the crystalline area are broken, and new hydrogen bonds are created between water and the hydroxyl groups of amylose or amylopectin, resulting in granule expansion. When temperatures rise over 60°C, most starches start to swell, and swelling increases noticeably after they reach 70°C and above. Swelling power and solubility represent the degree of connections between chains in the crystalline and amorphous lamella, which may be influenced by a variety of parameters such as amylose concentration, amylopectin

structural arrangement, and amylopectin chain distribution (Sasaki & Matsuki, 1998). In contrast to tuber starches, legume starches display a single, constrained swelling tendency and minimal amyloses leaching (Schoch & Maywald, 1968). Because of their dense packing inside the amorphous domains of the pulse starches and high amylose content, amylopectin has a limited ability to expand due to strong hydrogen bonding with adjacent amylose and amylopectin chains. Amylose-lipid complexes have also been shown to reduce swelling power and solubility (Hoover & Hadziyev, 1981). Dry peas contain 35-65% amylose of the total starch in dried peas with the rest being amylopectin (Zhou et al., 2019). Therefore, the combination of high amylose concentrations, hydrogen bonding tendencies, and the potential lipid-amylose complex formation support lower swelling capacities compared to other starches such as corn.

2.7.2. Starch Damage

Starch damage refers to small particles of starch that break away from the major starch granules (Arya S et al., 2015). These tiny particles hydrate more easily during dough making. The degree of starch degradation therefore influences the water absorption and dough mixing characteristics of flour and is technologically significant. Starch damage happens often during the milling process as a result of a combination of heat production and physical stress. The formation of minor starch degradation within the pea is thought to have occurred during the flour re-grinding process after extraction. Starch damage play important role in granules swelling and impact the starch pasting and functional properties causing an impact on the final product characteristics. The reduction of setback was associated to starch damage and reduced particle size in treated pea flour

(Elliot, Dang, & Bason, 2019; Song, 2007). In addition, starch damage can cause the releasing of volatiles (Gohl, 2019).

Damaged starch is known as being responsible for gas formation during fermentation and proofing, as well as its contribution to flour water absorption. These two factors could determine the quality of the final product. Starch damage had a major impact on bakery, cookies, biscuit, noodles and vermicelli products. It causes changes in color, time to bake, and cook loss of noodles (Wang, & Yang, 2005). The damaged starch portion also increases endogenous amylose hydrolysis, which results in the production of maltose. Therefore, maltose is utilized by yeast to ferment carbon dioxide in conventional baking methods (and cause loaves to rise). Excessive starch damage, on the other hand, might over-hydrate the dough, increase enzymatic action, and result in poor baking performance (Antoine et al., 2004; Morrison et al., 1994; Nagao, 1995).

2.8. Functional Properties of Dry Peas

Dry pea is one of the best materials to create novel item like pasta, noodles, snacks, plant-bask meat alterative and baked goods due to its functional qualities (Ren et al., 2021). The functional properties define how the ingredient reacts and work throughout the preparation, processing, and storage of food. In other word, it specifies how the component acts throughout preparation and cooking as well as how these processes impact the look, structure, texture, and flavor of the finished goods included among the functional features are the water solubility index, the water absorption index, the water holding capacity, the oil absorption capacity, the emulsion activity and stability, and the foaming capacity and stability. Functional qualities of flours can be affected by

ingredients including fibers, ash, moisture, proteins, lipids, and oils, as well as by the physical makeup of these ingredients (Godswill Awuchi et al., 2019).

2.8.1. Water and Oil Holding Capacity

Water holding capacity (WHC), also known as water hydration capacity, water absorption capacity, and water binding capacity, refers to the amount of water taken up by flour or food per gram of protein or the water retention ability of proteins against gravity separation to achieve the desired consistency. When water is added to flour/starch, the hydration process begins with hydrophilic interactions between starch and protein molecules with water molecules via hydrogen bonds (Godswill Awuchi et al., 2019; Lam et al., 2018). Water binding is caused through the interaction of ion-dipole, dipole-dipole, and dipole-induced dipole interactions (Vatansever, Tulbek, et al., 2020). Furthermore, the amino acid content influences WHC. WHC is a critical functional attribute for a component since it impacts the quality of the completed product in terms of mouthfeel, texture, and taste retention. WHC levels that are either low or too high might have a detrimental influence on food compositions and the textural quality of the food product. Furthermore, WHC is crucial in the creation of baked goods since it may alter numerous characteristics such as proofing, loaf volume, bread yield, bread crumb, shelf-life, and machinability during the bread-making process (Godswill Awuchi et al., 2019).

Oil absorption capacity (OAC) or oil holding capacity (OHC) is one of the most essential functional qualities of flour. Lam et al. (2018) define OAC as the amount of oil that flour could absorb per gram of protein. Protein shape, amino acid content, and surface polarity or hydrophobicity all contribute to OAC in the food system (Godswill

Awuchi et al., 2019). Flour protein content is very consistent with the level of OAC. In other word, flour with high protein content absorbs more oil. The interaction between lipid and protein in flour is generated by the binding of the aliphatic chains of lipid to the nonpolar side chains of amino acids (Lam et al., 2018). Flour OAC influences the mouthfeel, taste, texture, and yield of the finished product. The OAC is a vital functional characteristic of flour that is needed for generating doughnuts, pancakes, baked products, sweets, confectioneries, drinks, salad dressings, meat extenders, and meat mimics, and improving the sensory characteristics of the completed product (Vatansever, Tulbek, et al., 2020; Wang et al., 2020). Furthermore, greater OAC flour results in better palatability, shelf-life extension, and flavor retention when utilized in the production of meat or bread goods that need fat absorption (Chandra et al., 2015). Depending on the technique used to compute OAC, oil absorption capacity is either as a percentage or as a g/g value (Ferreira et al., 2018; N. Wang et al., 2020).

2.8.2. Pasting Properties

Gelatinization of the starch is an important characteristic in many of food processing and application. Gelatinization is the process of starch granule swelling when subjected to thermal and shearing conditions in exist water system. Gelatinization temperatures for pea flour include an onset temperature of 61°C, a midpoint temperature of 67°C, and an end gelatinization temperature of 76°C (Ratnayake & Warkentin, 2002) causes the swelling of the starch granules and molecules, such as amylose and amylopectin, which also leach from the granule causing viscosity or pasting viscosity (Debet & Gidley, 2006). Pasting is a crucial functional property because the sensory attributes such as final

texture and appearance, digestibility, and end use can be affected by changes in starch that occurs during pasting (Ocheme et al., 2018).

To measure the pasting properties of starch, several instrumental methods, such as amylograph (e.g., Brabender Amylograph), dynamic rheometer fitted with a starch pasting cell, Ottawa starch viscometer, Rapid-Visco Analyzer (RVA), and consistometer have been used (Balet et al., 2019a). The RVA viscometer is a heating and cooling system that measure the change in viscosity of a flour-water suspension over a certain time with continuous shearing. A starch pasting profile is created by analyzing variations in the viscosity of the starch solution as a function of temperature and time (Mariotti et al., 2005). According to Balet et al. (2019), the RVA test consists of five stages: addition of water to the sample, heating, holding at the maximum temperature, cooling, and the last stage as a holding stage. The standard RVA pasting profile includes an initial temperature of 50 °C, a holding time of 1 minute at 50 °C, a heating time of 3 minutes 42 seconds to 95 °C, a holding time of 2 minutes 30 seconds at 95 °C, a cooling time of 3 minutes 48 seconds to 50 °C, and a final holding time of 2 minutes at 50 °C.

Peak viscosity, trough (or hot paste) viscosity, breakdown viscosity, setback viscosity, final viscosity, peak time, and pasting temperature are RVA test parameters that are plotted as viscosity changes or resistance encountered during the test (Ohizua et al., 2017). According to Balet et al. (2019), peak viscosity is the greatest viscosity observed during heating or it is the maximum amount of water a granule can uptake. The trough viscosity is defined as the ability of the heated paste to resist breakdown, whereas breakdown viscosity is the ability of the flour to withstand heating and shear stress when cooking. Furthermore, final viscosity is defined as starch's ability to create viscous paste

after cooling down while setback viscosity is a measure of starch's retrogradation propensity. The peak time and the pasting temperature are when the peak viscosity occurs.

Pea starch has a lower peak viscosity than rice starch and other pulse starches (de Souza Gomes et al., 2018). Pasting characteristics of pea starch vary depending on cultivar. At 95 °C, starches from Carneval, Carrera, Grande, and Keoma pea had equal pasting temperatures but considerable variances in viscosity (Ratnayake et al., 2001), amylose leaching, swelling degrees, and distribution of amylopectin branch chain length have all been reported to influence pea starch pasting qualities (Hoover & Senanayake, 1996). Low setback and viscosity of pea starch were ascribed to a shorter amylose chain and a reduced fraction of chains at DP 6-12 (Ratnayake et al., 2001). Other components, such as lipids, also influence the pasting properties of pea starch (Simsek et al., 2009).

2.8.3. Retrogradation

Gelatinization is the process by which starch crystallites are changed from an ordered to an amorphous phase when heated in the presence of sufficient water (Ottenhof & Farhat, 2004). When the viscous solution cools, the amylose and amylopectin chains rearrange to produce a more ordered structure, a process known as retrogradation. Retrogradation can be divided into two types: short-term and long-term procedures. During retrogradation, crystallinity increases, syneresis occurs, the diffraction pattern changes to B-type, and gel formation occurs. In the prophase, the amylose matrix gel is generated, and then the short, exterior chains of amylopectin form double helices, which crystallize to B-type polymorphs, and the crystallinity is enhanced (Zobel, 1988). As a result, retrogradation affects starch structural, mechanical, and sensory properties that are

desired in food applications (Karim et al., 2000). Starch retrogradation can be determined using methods such as turbidity, differential scanning calorimetry (DSC), wide angle X-ray scattering (WAXS) and nuclear magnetic resonance (NMR). Syneresis parameters, which reflect the extent of retrogradation, revealed that the structure and functionalities of legume starch were susceptible to change during retrogradation due to a higher amylose content than cereal and tuber starch (Ratnayake & Warkentin, 2002).

2.9. Isolation and Modification of Pea Starch

2.9.1. Isolation of Starch

Kawamura et al. (1955) first published the starch separation process from pulses, which comprises treatment with 0.2% NaOH solution, washing with water, and dehydration with ethanol and water. Schoch and Maywald later proposed three ways for separating starch from pulses (1968). The first method proposed for separating starch from mung beans, garbanzo beans, and dehulled split yellow peas was steeping in warm water containing toluene to inhibit fermentation following by wet grinding and screening. These authors offered a second way for extracting starch from difficult-to-process legumes like lentil, lima beans, and white navy beans. This procedure involved steeping in warm water with toluene, followed by resuspension in 0.2% NaOH solution (to solubilize dissolve most of the protein). The alkaline solution was filtered through a 220-mesh nylon screen to remove some of the fine fiber, and it then flowed gradually down an inclined "table" that was a shallow, flat, trough made of heavy-gauge stainless steel with a total slope of 0.5 inches (1.27 cm). Multiple methods were evaluated because pulses are high in soluble protein, which is easily isolated from starch. However, fine

fiber and some of the flocculent proteins found in pulses co-settle with starch during the separation process, making starch isolation difficult (Schoch & Maywald, 1968).

Starch from pulses can be isolated using dry milling or wet milling (Gwirtz & Garcia-Casal, 2014; Haros et al., 2003). Dry milling involves extensive particle size reduction of grain, which is usually performed using pin mills, followed by air classification. To minimize the protein content of starch, repeated milling and air classification are used. However, to generate high purity starch with a protein level of less than 0.2%, washing with water and diluted alkali is essential. Wet milling produces starch with a higher purity than dry milling. A diluted alkali with a pH range of 8.5 to 10.0 is typically used (Davydova et al., 1995).Wet milling employs multiple passing through screens and alkaline washing (0.2% NaOH), which significantly lowers the protein content of pea starch. Repeated washing of the starch could produce starch with less than 0.8% protein content. Crucial factors that must be considered during starch separation include avoiding amylolytic or mechanical damage to the starch granules during the initial isolation processes, effective deproteinization of the starch, and reducing the loss of small granules (Singh, 2020). The alkaline steeping method considered one of the commonly used methods in the starch industry (Alavitalab, 2016).

Splitting and dehulling of seeds are the process of detach the hulls and the cotyledons (Saldanha do Carmo et al., 2020). Basically, by using a mill with 2 rollers and air classification, the hull can be separated from the seed. Hull can impact the purification processes; thus, the hull could act as a barrier in the starch isolation during the soaking stage that solubilize the protein. Therefore, dehulling of seeds prior to the extraction and the impact on the physical properties, chemical composition and techno-functional

properties was investigated (Saldanha do Carmo et al., 2020). Prior dehulling, increased starch enrichment of pea and faba bean starch in the course. Also, dehulled green pea flour produced the purest starch separation from particles that passed through a filter of 0.08 mm (Saldanha do Carmo et al., 2020). Dehulling can enable the use of seed coats (hulls) as source of phytochemicals that are then added to nutraceuticals, while cotyledons can be used as a source of plant protein (B. Singh et al., 2017). The dehulling has direct impact on the particle size of the starch, which helps to separate the protein from starch and increase the enrichment of starch during the isolation process. Prior dehulling of pea results in pea starch with high purity (Saldanha do Carmo et al., 2020). Furthermore, reduction of the particle size provides more surface area for the alkaline solution to extract more protein.

Yellow pea starch isolation has gained significant attention lately for its high amylose content compared to other legume starches and as potential food ingredient due to is ability to resist high temperature and shearing (Sun et al., 2015). Pea starch is considered a cheap source of starch compared to corn, wheat, and potato starch since it is a by-product of protein extraction (Ratnayake & Warkentin, 2002). The use of sodium hydroxide solution for extraction of pea starch has been widely adopted. Sodium hydroxide impact on starch structure and functionality and physicochemical has been evaluated (Palacios-Fonseca et al., 2013; Thys et al., 2008; Wang & Copeland, 2012). Sodium hydroxide is widely used chemical in the starch industry for increasing starch purity since it can solubilize protein during starch extraction, which tends to lower the starch gelatinization temperature and increase the peak viscosity (Saldanha do Carmo et

al., 2020). High pH above 8 could enhance the protein solubility and extraction in the first phase of the process (soaking and decanting) (Puchongkavarin et al., 2005).

2.9.2. Modification of Starch

It is generally known that certain starches have a low utilization value due to their poor functionality. As a result, starch modification can open new avenues for its use in the food sector. Physical, chemical, and enzymatic approaches can all be used to modify starches.

2.9.2.1. Physical Modification

Physical modification is often regarded as a non-chemical, safe, and cost-effective strategy. Both annealing (ANN) and heat-moisture treatment (HMT) are hydrothermal processes that do not result in granule component loss. The physicochemical behaviors of pea starch were reported to change when handled at elevated temperatures (100-120°C) and limited moisture content (22-27%) for 16 hours, including a decrease in swelling power, amylose leaching, and peak viscosity (Chung et al., 2010). Pre-gelatinization is the drying of starch after gelatinization. Deep freezing and thawing, osmotic-pressure, instantaneous controlled pressure drops, and thermal inhabited treatment are examples of new methods for physical starch modification (Zia-ud-Din et al., 2017).

2.9.2.2. Chemical Modification

Chemical modification, which includes cationization, crosslinking, and hydroxypropylation, are other treatments that involve the addition of functional groups to starches by derivatization. Cationic starches are formed when the dissociative hydroxyl groups of starch interact with cationic monomers. Cationization of pea starch has been

found to result in a decrease in gelatinization and pasting temperature but an increase in peak viscosity, permitting pea starch utilization in the paper industry due to improved paper strength (Zia-ud-Din et al., 2017). Sodium tripolyphosphate, sodium trimetaphosphate, adipic acid-acetic anhydride combination, phosphoryl chloride, and phosphoryl oxychloride are the most regularly used food grade cross-linking agents. Pea starches are vulnerable to retrogradation, which is undesirable in culinary applications. However, cross-linking hydroxypropyl ester groups to pea starches can considerably enhance pasting viscosity and reduce the amount of syneresis (Zia-ud-Din et al., 2017).

2.9.2.2.1. Supercritical Fluids Extraction

The use of solvent as separating methods go back to Paleolithic age. Throughout the years, the science has been developed to understand more about the solvation and liquid mixtures properties in the extraction processes (Herrero et al., 2010). The first observations of the supercritical fluid (SCF) media were by Hannay and Hogarth's in 1879. Nonetheless, the first use of the technology in commercial process application stared in the 1960 (Herrero et al., 2010). Organic solvent, such as chlorofluorocarbons (CFCs), usage is common in wide range of global industries, which presents a significant danger to the environment and damaging the ozone layer (Knez et al., 2014). In response, the Montreal Protocol objective in 1987 was to restrict or eliminate the use of these hazardous substances (Herrero et al., 2010).

Supercritical fluid (SCF) presents a novel chemical phenomena unseen in the traditional phases (gas, liquid and solid) with environmental, economic, health and safety benefits to replace the environmentally damaging conventional solvents (Capuzzo et al., 2013; Noyori, 1999). The term “supercritical” is used to describe the state of solvent

when it passes its critical temperature (Tc) and critical pressure (Pc). Beyond this point, the substance exhibits certain (Figure 1) typical physicochemical characteristics of liquid-like (solvent power, negligible surface tension) and gas-like (transport) properties (Amaral et al., 2017; Capuzzo et al., 2013). The most significant SCFs, like SC CO_2 and SC H_2O, are thermodynamically stable, nontoxic, nonflammable, noncarcinogenic, and non-mutagenic. Health and safety are two additional benefits of SCFs (Hrncic et al., 2018; Knez et al., 2010).

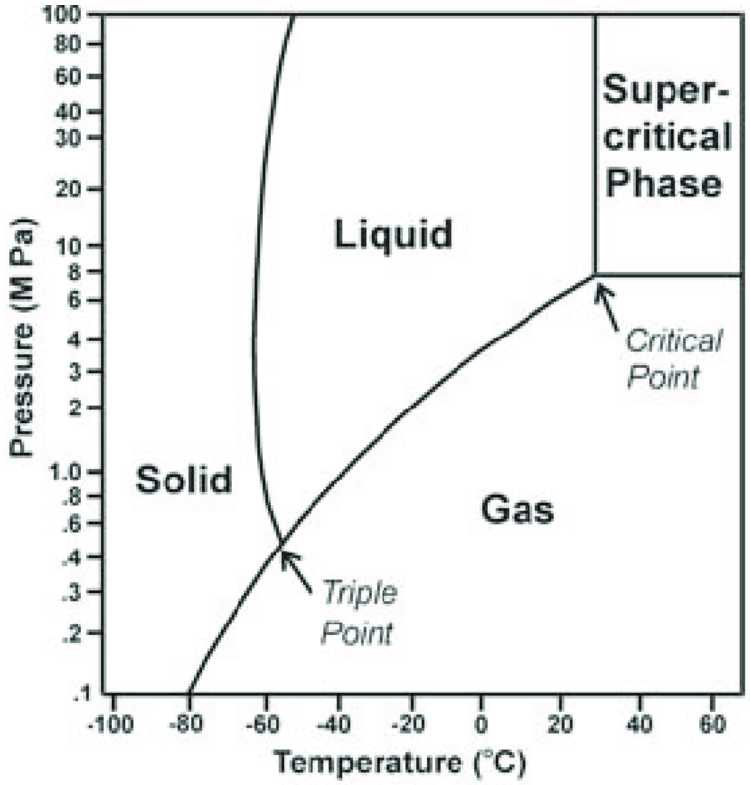

Figure 1: Phase diagram of carbon dioxide (Voormeij et al., 2003).

Carbon dioxide (CO_2) becomes a supercritical fluid at a critical point (31.2 °C and 7.3 MPa), which reduces product thermal damage specially in food and natural product (Capuzzo et al., 2013). Its low critical temperature, near to room temperature, prevents

the degradation of volatile and thermosensitive chemicals and limiting changes to the food's physicochemical, sensory, and nutritional properties, resulting in high-quality goods. Additionally, the ability to recirculate CO_2 into the system easily is an important factor for the economy, environment, and consumer health (solvent free product). Also, the lower critical pressure compared to other supercritical substances, such as supercritical water (373, 95 °C and 22.064 MPa), offers little energy and investment costs (Amaral et al., 2017). Other unique physiochemical properties that supercritical carbon dioxide (SC CO_2) has, such as high diffusivity, absence of surface tension, low viscosity, tunable density and controllable solvent (by adjusting the temperature and pressure) made it a desirable alternative processing for components separation and starch modification (Herrero et al., 2010; Ivanovic et al., 2016; Muljana et al., 2009; Vatansever & Hall, 2020). For these reasons, 90% of the supercritical fluid extraction has been performed with supercritical carbon dioxide (SC CO_2) (Capuzzo et al., 2013).

The mass transfer of the solute in the supercritical fluid (SCF) is a crucial factor that needs to be taken in account (Hrncic et al., 2018). The solubility of the solute in the selected solvent impacts mass transfer. Under various operating conditions, substances have varied degrees of solubility which are determined by temperature and pressure of SCFs. However, due to the low polarity of SC CO_2, polar compounds solubility is limited. To overcome this problem, polar cosolvent usually uses, such as methanol or ethanol, to increase the power solubility of the solvent. SC CO_2 has low polarity index and to overcome this issue polar cosolvent or modifier usually are used, such as methanol or ethanol (Capuzzo et al., 2013). In addition to this, the moisture content of the solid

material can impact the extraction processing by acting as co-solvent. Therefore, the raw material should have 4-14% moisture (Capuzzo et al., 2013).

Supercritical fluid extraction (SFE) has been applied in the extraction of components and bioactive substances, particle engineering procedures, chromatography, tissue engineering and regenerative medicine, enzymatic reactions, drying, pasteurization and sterilization (Amaral et al., 2017; Herrero et al., 2010; Knez et al., 2010). Using SCFs in a variety of processes may result in the creation of entirely new products with properties that have a very low impact on the environment, such as low energy consumption throughout the process, as well as advantages for health and safety (Hrncic et al., 2018; Knez et al., 2010). In case of extraction, there are two main purposes that include 1) the extracted substances are the main products or 2) the extractions are the unwanted substances (Knez et al., 2010). Examples of the first group of extraction is to achieve high yield of hop constituents, essential oil, wax, aroma and flavor compounds, and other organic components (Capuzzo et al., 2013; Hrncic et al., 2018; Knez et al., 2010). The other group of materials include the decaffeination of coffee and tea (the largest scale process), defatting of some seed plants, deflavoring of flour material and decolorizing of corn zein (Knez et al., 2010; RincÓn et al., 2000; Sessa et al., 2003; Vatansever & Hall, 2020). For example, SC CO_2 + EtoH was employed to improve physical properties of pea starch through deflavoring and removing its unpleasant taste. The removing of undesirable flavor is crucial for quality, marketability and acceptability of products made with pea flour. It also enhanced the digestibility of the pea starch (Vatansever et al., 2021; Vatansever & Hall, 2020).

Supercritical fluid extraction system consists of the gas tank, pump, extractor, and separator. In some cases, a co-solvent (usually alcohol) will be included with an additional pump to facilitate the extraction of the alcoholic soluble (i.e., polar) compounds from the raw material. SFE can be used for solid or liquid mixture separation and the solvent, and the parameters setting is chosen to regard the type of material and the compounds to extract. In large scale processes, regeneration stage for the recycling of the CO_2 gas is added (Capuzzo et al., 2013; Jodhner & Meireles, 2016; Knez et al., 2010).

To ensure highest yields or removal possible of the extract, the process parameters of the raw material had to be studies (Herrero et al., 2010). Alternatively, optimization of extraction parameters using response surface methodology to remove the target material should be done. For example, volatile and non-volatile compounds were targeted to remove the undesirable flavor from the flour (Vatansever & Hall 2020). However, the impact of the same parameter in the extraction of the undesirable flavor and the chemical, functional and pasting properties of pea starch have not been studied (Herrero et al., 2010).

2.9.2.2.2. Ethanol Treatment

Solvent extraction and modification of starch is a common method in the industry to enhance the starch characteristics. Hillen (2016) and Roland et al. (2017) concluded that ethanol extraction was one of the most suitable methods for flavor reduction in yellow pea flour and lentil, respectively. Ethanol and water combinations allow for the extraction of both water and ethanol soluble molecules (Do et al., 2014). As a result, the reduction of water and ethanol soluble volatiles may be the cause of the decrease in pea flavor.

Oftentimes, the use of high-pressure extraction is combined with ethanol solvent (Hillen, 2016). However, the risk of impacting the raw material by pressure is greater. Thus, Gohl (2019) studied the desirable condition of ethanol extraction for maximum flavor removal and the impact on the raw material regarding the composition and physiochemical properties of pea flour. Gohl (2019) reported a reduction in moisture, ash with no loss of protein, starch or resistant starch level using ethanol extraction at ambient conditions. In addition, increases of water absorption, setback and pea time were observed after the extraction. In contrast, the impact of the extraction in pure pulse starch characteristics and functionality is lacking in the literature. Therefore, the goal of this study was to examine the impact of the extraction on the composition, functional and pasting properties of pea starch, as compared to pea flour, solvent extraction and SFE extraction.

2.10. Food Application

The consumption of pulses as vegetable after soaking and cooking, or as canned, frozen, fried, roasted, salted, or soup is more common in developing countries. Therefore, most of the production of peas, for example in the US, is exported globally to those regions (Simsek et al., 2009). However, due to the contribution pulses can make in the human diet and health, economy, and the environment, developed countries currently have been interested in pulses as ingredients in food system (Asif et al., 2013; Boye et al., 2010a). Despite the high-quality ingredients of pulses, their utilization and characteristics in food application have not been extensively studied.

The most utilization of dry peas is as flour. Pea flour has been applied to novel functional food such as banana bread, biscotti, and pasta (Marinangeli et al., 2009), bread

(Bourré et al., 2019), biscuits (Zhao et al., 2019), cookies, crackers, and gluten-free products (Gohl, 2019; Hillen, 2016). On the other hand, as a fractionated ingredient, pea protein is frequently employed as a replacement for soybean or animal proteins in a variety of functional applications due to its availability, affordability, nutritional content, and health advantages (Lu et al., 2020). The market for pea protein as an added value ingredient has been increasing with the increasing (Boukid et al., 2021).

As a result, for every 1 pound of protein we expect 2 pounds of starch as a byproduct of the extraction. Therefore, pea starch is considered as cheap source of starch compared to corn, wheat, and potato starches (Ratnayake & Warkentin, 2002). Food ingredients have different roles in food regarding the functionality and the impact on the product such as appearance, texture, structure, and taste. Starch, in general, is used mainly for thicken food mixtures, ~~and~~ form bulk volume of foods, and is responsible for the gelatinization, browning, dextrinization, and gelation (Godswill Awuchi et al., 2019; Pietrasik et al., 2020). The high content of amylose in pea starch and extended retrogradation are two factor that drive the utilization of pea starch in food application (Ratnayake & Warkentin, 2002). Functional properties of the food are very critical factors that determine the product's end quality. For example, pea starch, when compared to rice starch, has low rates of digestion. This might be a crucial nutritional quality that permits the use of pea starch in diabetic diets (Ratnayake & Warkentin, 2002). Pea starch has been applied to low fat bologna (Pietrasik & Soladoye, 2021), gel (Rong et al., 2022), to enhance the quality of noodle, vermicelli, pea starch gel, beef burgers (Sun et al., 2015b; Pietrasik et al., 2020), noodles (Wang et al., 2012), bread (Lu et al., 2018b), and other vegan food (Rasskazova & Kirse-Ozolina, 2020). Other type of starch has been

adding to yogurt to enhance the texture and reduce syneresis (Mwizerwa et al., 2017; Saleh et al., 2020).

3. MATERIAL AND METHODS

3.1. Materials

AGT food and ingredient company (Minot, ND) was the source of whole yellow pea flour for the lab scale isolation. Pea seed (Orion cultivar) were used for the industrial scale isolation. Chemical agents (sodium hydroxide, ethanol, acetone, hexane) were provided for the extraction and chemical analysis were obtained from Fisher Scientific (Pittsburg, PA). The samples were stored in ziplock bags at -40 between analyses and warmed to ambient temperature (21 °C) before each analysis.

3.2. Methodology

3.2.1. Starch Isolation

3.2.1.1. Optimization of Industrial Starch Isolation Process

Pea starch isolation from whole yellow pea seed was adopted based on alkaline steeping (Simsek et al. 2009). Four different raw pea materials were used in the optimization of the isolation method including whole pea seed (WPS), dehulled pea seed (DPS), whole pea flour fractionation (100 mesh size) (WPF), and dehulled pea flour fractionation (100 mesh size) (DPF). The starting material were prepared as follows (Figure 2). First, the whole yellow pea seed (WPS) was taken as it is with no pre-preparation. The dehulled pea seed (DPS) sample was obtained by running samples through 2 rollers (DY-168 Grain Mill; Sichuan Shuheng Wenjing e-commerce Co., Ltd., Chengdu, China) followed by air classification (Carter Day, Minneapolis, MN) to

separate the hull from the seed. Air classification was repeated with manual separation to insure high hull removal. The flour fraction samples were obtained from the whole and dehulled peas. These samples (200 g) were milled through a 0.5 mm screen using a UDY cyclone mill and dry sieved through a series ((60 (250 µm), 80 (177 µm), and 100 (149 µm) mesh) of sieves (W.S. Tyler, Mentor, OH) and the particle passing 100 mesh size was collected (WPF and DPF from the whole and dehulled flours, respectively).

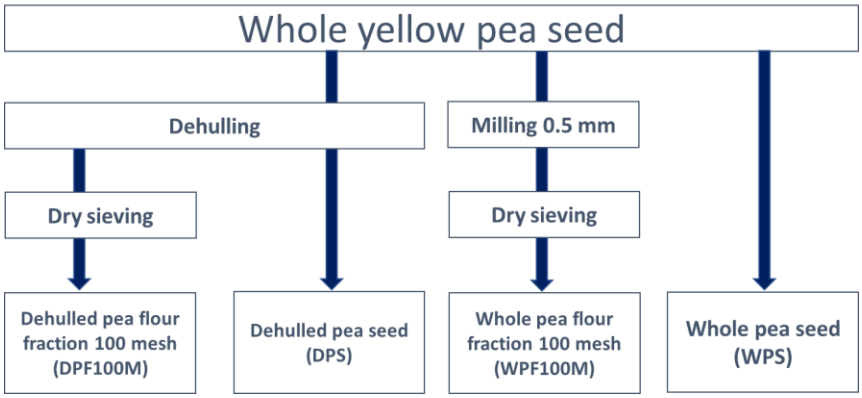

Figure 2. Preparation of starting materials for starch process optimization.

Briefly, 600 g of the WPS or DPS or 100 g of WPF or DPF was steeped in 0.02% sodium hydroxide solution (ratio of 1:3, sample: solution) with pH of 9.5 at room temperature for 18 hours (Figure 3). The NaOH solution was discarded, and the starch cake was blended in food processor (Cuisinart, Wast Windsor, NJ) for 3 mins with distilled water (ratio of 1:1). The resulting starch was passed through 35 (500 µm), 60 (250 µm), 80 (177 µm), and 100 (149 µm) mesh sieves. The collected starch cake in the bottom pan was transferred to a beaker and allowed to settle for 1h or until the appearance of starch separation. The residual pulp was blended again with Waring blender (Waring, New Hartford, USA) for 1 min with distilled water (ratio of 1:1). This

step was repeated two to three times to ensure complete separation of starch and the collected starch solution was left to settle in a beaker again. After washing the separated starch with distilled water, the supernatant was discarded by carefully decanting, and the starch cake transferred to aluminum foil pans for drying. The samples were dried at 45 °C for 18 h at air oven. The samples were milled with a 0.5 mm screen using a UDY cyclone mill to reduce starch particle size. The samples were stored in Ziplock bags at - 40 °C until analyses were completed.

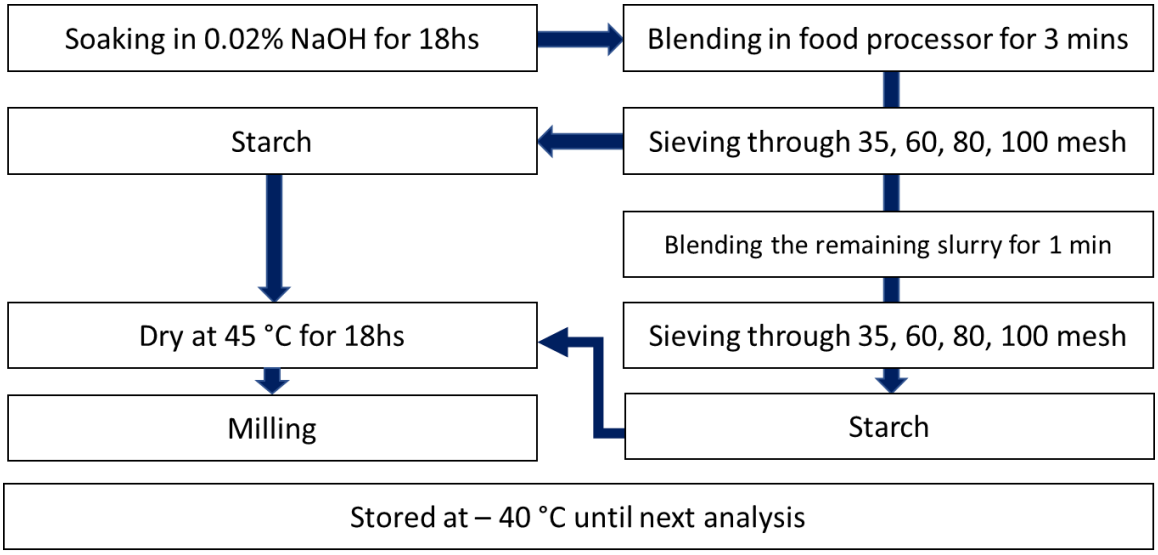

Figure 3. Starch isolation process using alkaline steeping.

3.2.1.2. Laboratory Scale Starch Isolation

The isolation of starch from yellow pea flour was obtained following the method of Vatansever et al. (2021) with some modifications. Whole yellow pea flour (40 g) was homogenized with 100 ml distilled water using a Waring blender (Waring, New Hartford, USA) at high speed for 70 s with stopping after 10 s. This stopping period was to ensure

a proper mixing of the flour with distilled water before continuing. The slurry was passed through 60 (250 µm), 70 (212 µm) and 230 (63 µm) mesh. Final slurry was centrifuged at 3000 rpm for 5 min and the precipitated starch cake was dispersed in 50 ml of 0.02% NaOH and then stored 1 h at 4 °C. After the adjusting of pH to 6.5 using 1 M HCl, the starch slurry was centrifuged at 4750 rpm for 6 min. The supernatant was discarded, and the starch was washed with 100 ml distilled water, 100 ml 95% EtOH, and 100 ml acetone, respectively. After each washing agent, the mixture was centrifuged at 3000 rpm for 5 min, and the supernatant was discarded. After acetone washing, the starch was dried at 45 °C for 18 h (overnight). The isolated pea starch samples were stored in Ziplock bags at – 40 °C until analyses were completed.

3.2.2. Post-Treatment

3.2.2.1. Supercritical Fluid Extraction

The supercritical fluid extraction (Figure 4) with carbon dioxide protocol was preformed according to previous study (Vatansever & Hall, 2020) with some modifications. Briefly, an Applied Separations Spe-ed SFE-NP (Allentown, PA) extractor was used with CO_2 as main solvent and ethanol as modifier or co-solvent. A 15 g of the starch was mixed with glass beads in a 25 ml stainless steel vessel. Due to the unavailability of an additional pump for the co-solvent addition, 4 ml of ethanol was poured directly into the vessel from the bottom. The samples were subjected to 85 °C temperature, 427 bar pressure, and CO_2 gas rate flow of 1 L/min for 40 min. Afterward, the extracted samples were dried at 70 °C for 1 h in convection oven to remove the ethanol. The dried starch was stored at -40 °C until analyses were completed.

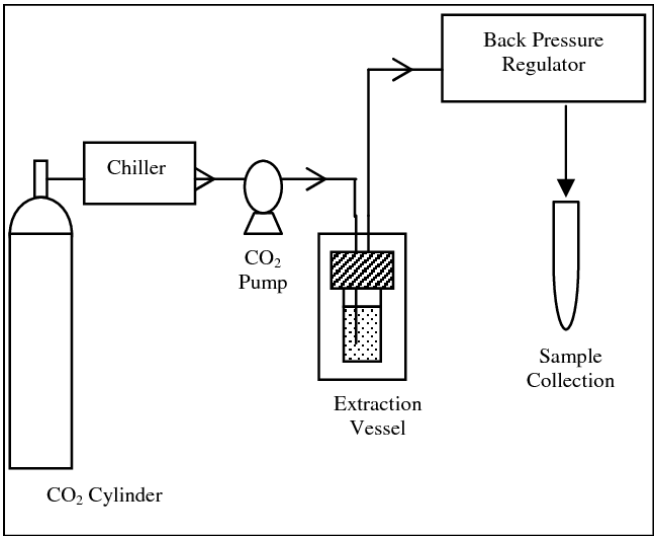

Figure 4. Flow diagram of supercritical fluid extraction system (Wei et al., 2005).

3.2.2.2. Ethanol Treatment

Raw pea starch obtained during starch isolation was subjected to extraction with aqueous ethanol (47.5%) for 63 min following the method of Gohl (2019) with some modifications. Briefly, 300 g of pea starch was extracted in approximately 3 L of 47.5% ethanol for 63 min. The supernatant was discarded by careful decanting, and the starch cake was dried at 45 °C for 18 h in air convection oven. After drying, the samples were re-milled using 0.5 mm screen - UDY cyclone mill to reduce starch particle size and stored at – 40 °C in Ziplock bags. Ethanol treated starch (ETS) were warmed to room temperature prior to composition analysis and functionality testing.

3.2.3. Proximate Composition

3.2.3.1. Moisture Content

The moisture content of the starch was determined using the official AACC Approved Methods of Analysis (44-15.02). This method is based on the moisture content as loss in weight of sample when heated under controlled conditions. In replicate, in pre-weighed metal tins with the lid (W1), 2 g of the sample was added for each duplicate. The starch, metal tins, and lid weight was recorded (W2) before placing in a 130 °C air oven for 3 h. After 3 h, the sample was taken out of the oven and allowed to cool to room temperature in a desiccator for 20 min before the final weight was taken (W3). Moisture content was determined using the following formula:

$$\text{Moisture content (\%)} = (W2 - W3) / (W2 - W1) * 100$$

3.2.3.2. Protein Content

The protein content of starch was determined using combustion method following the AACC Approved Methods of Analysis 46-30.01 (AACC 2010). A conversion factor of 6.25 was used to calculate protein content.

3.2.3.3. Lipid Content

The fat content of starch was determined using the official AACC Approved Methods of Analysis 30-10.01 (AACCI 2010). In pre-weighed filter bags (W1), 1.5 g of starch was added, and the filter bags are sealed. The weight of filter bag and samples was recorded (W2). Prior to the extraction, samples were dried at 104 °C for 3 hours in an air oven. The dried samples are placed into a desiccator to cool before the weight was recorded (W3). After that, the samples were put into the fat extractor (Buchi, Flawil, Switzerland).

The fat was extracted from the starch using hexane for 4 h and 25 min. The samples were removed from the extractor and allowed to cool to room temperature for 5 minutes in a fume hood before drying for 30 min at 103 °C in air oven to remove the residual hexane. Before taking the final weight (W4), the samples were kept in the desiccator to cool. The formula used to determine lipid content was as follows:

$$\text{Lipid Content (\%)} = (W3 - W4) / (W2 - W1) * 100$$

3.2.3.4. Ash Content

Ash content was obtained following the official AACC Approved Methods of Analysis Method 08-01.01 (AACCI 2010). In a pre-weighted crucible (W1), 1g of the starch was added. The weight of the crucible and samples were recorded (W2). To prevent burning of the sample, the oven was set to heat to 350 °C for 1 h, then 450 °C for 1 h, before reaching 590 °C overnight. The sample was kept in a desiccator before the final weight was taken (W3). The ash content was determined using the following formula:

$$\text{Ash Content (\%)} = (W3 - W1) / W2 - W1) * 100$$

3.2.3.5. Starch Analysis

Total starch and starch damage content was determined using the official AACC Approved Methods of Analysis 76-13.01 (AACCI 2010). A Megazyme K-TSTA-100A kit from Neogen (Lansing, MI) was used for the analysis of total starch. Starch (0.1 g) was weighed during the determination and each sample run in duplicate. The starch damage was estimated following the official AACC Approved Methods of Analysis

Method 76-31.01 (AACC, 2010). A Megazyme K-SDAM 06/18 kits from Neogen (Lansing, MI) was used for the analysis of starch damage. Starch (0.1 g) was used during the determination and each sample ran in duplicate.

3.2.4. Functional Properties

3.2.4.1. Water Holding Capacity

Water Holding Capacity (WHC) of pea starch samples were determined following the official AACC Approved Methods of Analysis (56-37). Briefly, 1 g of the starch sample was weighed (W1) in a test tube. The test tube, sample, filter cloth, and syringe barrel were weighed (W2). The filter cloth and syringe barrel were removed, and 20 to 22 drops of distilled water were added dropwise to the tubes and the starch mixed with glass stirrer for 1 minute. The filter cloth was used to clean the glass stirrer from the remaining starch and place inside the barrel in upside down direction. This syringe assembly was placed in a 50 ml centrifuge tube and then centrifuged at 300 *x g* for 10 min. The final weight of the test tube, sample, filter cloth, and syringe barrel were taken after removing the 50 ml tube (W3). WHC was calculated using the following formula:

$$\text{Water holding capacity} = (W3 - W2) + (W1 * mc) / (1 - mc)\ (W1)$$

Where, mc = initial moisture content of sample.

3.2.4.2. Oil Holding Capacity

Oil holding capacity (OHC) was determined using the method described by Wang et al. (2020). Sample (0.5 g) was weighed in a test tube. Then, the combination weight of filter paper, test tube and syringe barrel were recorded. Canola oil (1.5 mL) was added to the test tube and vortexed for 5 seconds every 10 minutes for 20 min (total of three times

vertex). The test tube was inverted with the filter paper at the bottom of the test tube into the syringe, then the assembly was immediately place into a 50 ml centrifuge tube and centrifuged at 600 *x g* for 25 min. after that, the whole assembles of syringe barrel, filter paper and test tube, sample and oil absorbed was weighed. A blank sample with filter paper was also included during the centrifugation to avoid the problem created by some free oil being entrapped in the filter paper and not collected at the bottom of the conical centrifuge tube. The OHC was calculated using the following formula:

$$\text{Oil holding capacity} = (W3 - W2 - W4) / (1 - mc / 100) (W1)$$

Where, W1 = weight of the sample before oil addition (g),

W2 = weight of the syringe barrel, filter paper, test tube, and sample (g),

W3 = weight of the syringe barrel, filter paper, test tube, sample, and oil absorbed after centrifugation (g),

W4 = weight of oil absorbed by the blank filter paper after centrifugation (g),

mc = initial moisture content of the sample (%).

3.2.4.3. Pasting Profiles

Pasting properties of pea starch samples were determined using Rapid Visco-Analyzer (RVA) (4800 Perten Instruments, Springfield, IL), following the method of Hyang et al. (2007). Briefly, 28 g of 6% (w/w) starch slurry was used during the following procedure. During a run, the temperature was initially set at 50 °C and increased to 95 °C over the course of 4 min. 42 s. There was then a holding time that

lasted for 7 min. 12 s. The slurry was then cooled to 50 °C at 11 min and maintained at 50 °C for the remaining time of the 23 min run.

3.2.4.4. Texture Profiles

After RVA, starch samples were kept at room temperature for 2 hours to cause gel formation. A texture analyzer (Ta.Tx, Texture Technologies Corp, South Hamilton, MA) was used to analyze the gel strength of the gels generated in the canisters. Each canister was placed upright on the metal plate, and the gel was compressed with a cylinder probe (TA-510A 10 mm dia x 45 m long) at a speed of 4 mm/s to a distance of 15 mm and a trigger force of 2 g. The compression generated a force-time curve, from which the hardness (height of the first peak) was calculated. Each sample was run in triplicate.

3.2.5. Food Application

3.2.5.1. Preparation of Yogurt

Yogurt was prepared using the starter culture of *Lactobacillus delbrueckii* subsp. bulgaricus (L. bulgaricus), *Streptococcus Thermophilus* and *Lactobacillus acidophilus*. Starch treated samples (ISE, ISC-SFE, ethanol treatment) were used with one modified corn starch sample as control. The ingredient percentage was as indicated in table 1. Briefly, the process was completed by first solubilizing the starch ingredients and heat at 75 °C for 10 mins. After complete solubilization of the starch, the mixture was brought to 40-43 °C in room temperature and then the starter cultures (3 g for 1 L of milk) were added and stirred well. The mixture was divided into glass cups and placed in an incubator at 42 °C for 4 h or until yogurt began to thicken and reach a pH of 4.5. The

samples were stored overnight at 4 °C and in the next day the hardness and syneresis were measured in duplicate as described below.

Table 1. Formula of pea starch fortified yogurt.

Ingredients	%	Weight (g)
Milk	97.7	97.7
Pea starch or (modified corn starch for the control)	2	2
Starter culture	0.3	0.3
Total	100	100

3.2.5.2. Preparation of Pudding

Pudding (Table 2) was made by mixing the drying ingredients (cocoa and sugar) with starch treated samples (ISE, ISC-SFE, ETS) and modified corn starch sample as control. Briefly, the dry ingredient mixture was added to milk and heated to reach 80 °C with stirring. After reaching 80 °C, the pudding was stirred for one min and taken off the stove. The cooked pudding was divided into plastic cup and placed in the refrigerator (4 °C) to cool overnight.

Table 2. Formula for chocolate pudding made with experimental pea starch and modified corn starch.

Ingredients	Pudding with Pea Starch (g, %)	Pudding with Corn Starch (g, %)
Milk	240 (83)	240 (83)
Sugar	22 (7.6)	22 (7.6)
Pea Starch	14 (4.8)	0 (0)
Modified Corn Starch	0 (0)	14 (4.8)
Cocoa	14 (4.8)	14 (4.8)
Total	290 (100)	290 (100)

3.2.5.3. Physical Evaluation

3.2.5.3.1. Gel Strength

Starch samples were kept at room temperature for 15 mins before the analysis. A texture analyzer (Ta.Tx, Texture Technologies Corp, South Hamilton, MA) was used to analyze the gel strength of the yogurt and pudding. Each cup, containing approximately 25 g, was placed upright on the metal plate, and the sample was compressed with a cylinder probe TA-510A at a speed of 4 mm/s to a distance of 10 mm and a trigger force of 2 g. The compression generated a force-time curve, from which the hardness (insert unit) was calculated. Each sample was run in triplicate.

3.2.5.3.2. Syneresis

Syneresis of yogurt and pudding samples was measured for everyday for 6 days for yogurt, and 5 days for pudding. After the preparation of samples, approximately 25 g of sample were poured into plastic cups and recorded (this was used to calculate the syneresis percentage). In the day of analysis, samples were kept in room temperature for 30 mins before taking the syneresis value. The surface of the samples was broken with spoon and then the separated liquid weight was recorded. The value of separated whey was taken in duplicate. The percentage syneresis was calculated using the equation below:

Syneresis (%) = [Total weight of drained whey (g) / Total weight of sample (g)] ×100

3.2.6. Statistical Analysis

Analysis of variance (ANOVA) at a 5% significance level ($p \leq 0.05$) was performed on all the outcome data from all parameters using excel program to determine the significant differences among treatments. Standard deviation was determined to measure the variation within the samples. Fisher's least significant difference (LSD) test was used to evaluate mean value differences between samples in case of more than 2 group evaluation.

4. RESULTS AND DISCUSSION

4.1. Starch Isolation

4.1.1. Optimization Of Industrial Scale Starch Isolation Process

The objective of the study was to optimize pea starch extraction and to examine the impact of dehulling prior to milling on the extraction process and the purity of the pea starch. Therefore, the different isolation starting materials were analyzed for their moisture, protein, ash, and starch content as well as water and oil holding capacity and pasting properties.

4.1.1.1. Proximate Compositions

Moisture content was determined to calculate the dry weight (dry weight basis, d.w.b.) mass of the chemical compounds. The results show higher moisture content in the flour fraction samples (WPF, DPF) compared to the whole and dehulled seed samples (Table 3). All samples had protein contents of less than 1.63% (d.w.b) compared to 18.6% for the whole pea used. These results agreed with the finding of starch isolation from corn using similar alkaline steeping approach (Palacios-Fonseca et al., 2013). This

high reduction can be attributed to alkaline steeping as an effective method to remove high amounts of protein during the initial phase (steeping) of isolation (Puchongkavarin et al., 2005). Puchongkavarin reported a high reduction in protein content in rice starch after steeping the rice overnight in 0.4% NaOH solution at 5 °C compared to enzyme and water steeping approaches. The polar side chain of the alkali-soluble protein can interact with sodium hydroxide via hydrogen bonding during soaking and thus aid to solubilize the protein that can then be removed during liquid decanting. In addition to this, alkaline solution could soften the protein-starch matrix to be separated easily by centrifugation (Lee et al., 2007). Furthermore, zein found in protein bodies is water insoluble but can be solubilized in alkaline solutions with high pH above 8.

Table 3. Chemical composition and starch recovery of isolated starches.

Treatment[1]	Moisture content %[2]	Ash%[3]	Protein %[3]	Starch%[3]	Starch recovery%[3]
WPS	7.9 (0.02)[b]	0.4 (0.004)[a]	1.6 (0.0003)[a]	91.9 (1.18)[a]	65.0[d]
DPS	6.1 (0.17)[c]	0.3 (0.005)[b]	1.4 (0.007)[b]	93.2 (1.78)[a]	79.1[b]
WPF	11.6 (0.06)[a]	0.3 (0.014)[b]	1.0 (0.0007)[c]	86.4 (1.50)[b]	75.4[c]
DPF	11.6 (0.03)[a]	0.2 (0.002)[c]	0.8 (0.0003)[d]	88.2 (1.56)[b]	87.6[a]

[1]Abbreviation: WPS, whole pea seed; DPS, dehulled pea seed; WPF, whole pea fraction (100 mesh size); DPF, dehulled pea fraction (100 mesh size).
[2]Values of mean (standard deviations).
[3]Dry weight as values as mean (Standard deviations). Values with different letters in the same column are significantly different at $p \leq 0.05$.

For pea starch isolation with the alkaline procedure, a final protein content of 0.4% can be achieved (Sun et al., 2015b). However, the high protein content (0.87% to 1.63% d.w.b) in the isolated starch (Table 3) can be associated with blending and milling operation as well as the lack of a centrifugation step. As a result, the higher protein likely is the result where starch granules did not detach sufficiently from the protein matrix. Sun et al. (2015) used an impact mill which might enhance size reduction and the separation of protein bodies from starch. Unlike Sun (2015) who used centrifugation, the starch separated in this study was by gravity force and not by centrifugation, decanting likely left some of the protein at the surface of the starch and remained with the dried starch.

In both cases of dehulling (DPS and DPF), the protein content in starch obtained from these materials was less than in the starch obtained from whole pea seed and flour fraction (Table 3). This result was the same as previous reported (Saldanha do Carmo et al., 2020). As a result, a high starch recovery was observed in dehulled treatments, compared to their counterparts (Table 3). This can be attributed to the removal of the hull, which can be a contaminating material in the starch. Since most protein will be solubilized in the alkaline steeping stage, steeping the whole seed flour can limit the removal of protein by interfering hull fiber. Owing to alkaline and dehulling process, the highest starch content, at 93.20 % d.w.b, was in DPS. Also, the interaction between dehulling and alkaline steeping influences the removal of ash. Initially, the pea (i.e., Orion cultivar) used in the starch isolation had an ash content of 3.5%; thus, supporting that the isolation facilitated the ash reduction (Table 3). Other researchers support that dehulling of seed prior to starch extract aided in the removal of ash (Palacios-Fonseca et al., 2013; Saldanha do Carmo et al., 2020).

Overall, the method of alkaline steeping and dehulling was used to create a high purity starch isolation process. Use of alkaline as an extraction solvent significantly lowered the protein and ash content in all samples; however, starch from dehulled samples had the lowest protein and ash content compared to the whole seed samples. Dehulling prior to the extraction eased the separation of starch granules from the protein bodies since the fiber was removed, which generally interferes with the sieving operation.

4.1.1.2. Pasting Properties

Pasting properties of the different isolation materials were impacted by the size reduction of flour prior to the extraction of starch (Table 4). In the case of flours obtained through fractionation (WPF, DPF), they had higher peak viscosities than the starch isolated from whole (WPS) and dehulled (DPS) pea. In general, alkaline steeping increase peak viscosity and swelling power due to its ability to remove fat (Lee et al., 2007). Additionally, smaller particles provide a larger surface area to which starch may be exposed during pasting, leading to faster water absorption, and swelling, as well as higher peak and final viscosities (Vatansever, Rao, et al., 2020). Yellow pea flour also showed an inverse correlation between pasting properties and particle size (Kaiser et al., 2019). For example, the smaller the particle, the greater the viscosity of the flour paste.

Table 4. Pasting properties of the isolated pea starches.

Treatments[1]	Peak viscosity (cP)	Trough (cP)	Breakdown (cP)	Final viscosity (cP)	Setback (cP)	Peak time (min)	Pasting temp. (°C)
WPS	5759[b2]	4162[b]	1596[b]	12334[a]	8172[a]	4.5[a]	73.9[a]
DPS	6334[b]	4178[b]	2155[b]	14241[a]	10063[a]	4.4[a]	73.1[a]
WPF	8037[a]	4902[a]	3135[a]	12574[a]	7672[a]	4.3[a]	73.3[a]
DPF	8080[a]	5068[a]	3012[a]	9971[a]	4903[a]	4.3[a]	73.6[a]

[1]Abbreviation: WPS, whole pea seed; DPS, dehulled pea seed; WPF, whole pea fraction (100 mesh size); DPF, dehulled pea fraction (100 mesh size).
[2] Values (n=2) with different letters in the same column are significantly different at $p \leq 0.05$.

4.1.1.3 Functional Characteristics

Dehulling did not significantly impact the water and oil holding capacities. However, the WHC was significantly higher for the starches from the fractionated raw material, but once again dehulling did not impact WHC (Figure 5). This might be related to particle size and potentially starch damage.

Unexpectedly, dehulling did not impact the pasting or functional property of the samples. The dehulling prior to extraction improved the extraction yield and purity of starch and made available hulls to be used in other applications. Isolating starch by the dehulling method is the most suitable way to produce starch from yellow dry peas for food ingredient utilization.

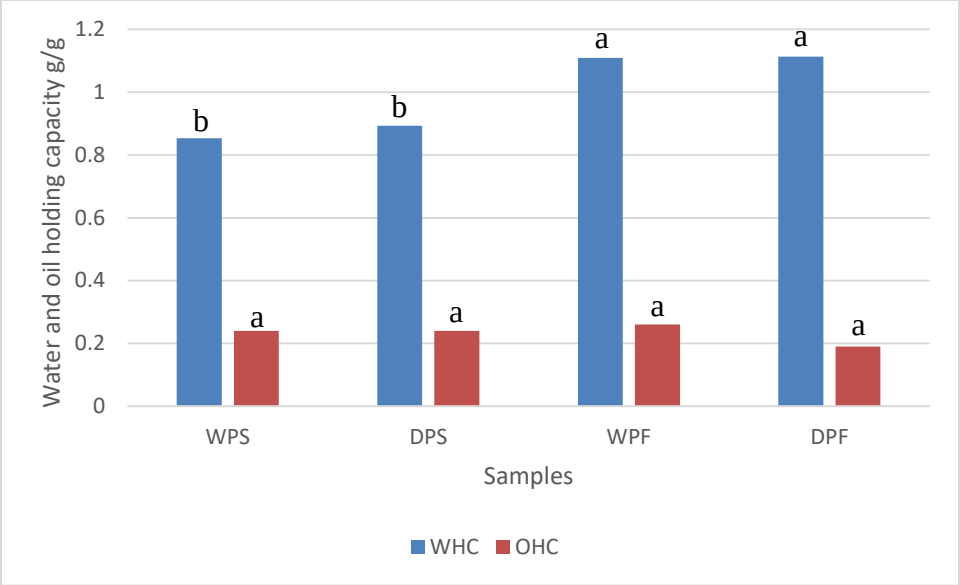

Figure 5. Water holding capacity (WHC) and oil holding capacity (OHC) of starches isolated from pea. Abbreviation: WPS, whole pea seed; DPS, dehulled pea seed; WPF, whole pea fraction (100 mesh size); DPF, dehulled pea fraction (100 mesh size). Values with different letters in the same functionality test are significantly different at $p \leq 0.05$.

4.1.2 Lab Scale Starch Isolation

A lab scale starch isolation from yellow pea flour was done to study its chemical, pasting and functional properties before and after the extraction of supercritical carbon dioxide (SC-CO_2) (Vatansever et al., 2020). This isolation was done to make general comparisons between lab prepared starch composition and functionality to that of an industrial approach as described in section 4.1.1.

4.1.2.1 Proximate Compositions

The results of the lab scale extraction (LSE) were similar to the previous starch isolation (Tables 3 and 4). A significant reduction of the moisture content due to the drying stage to remove acetone, as well as the use of ethanol and acetone might have additional drying effectiveness. The isolated starch had a high purity with low protein

(1.41% d.w.b), ash (0.11% d.w.b) and fat content (0.10 % d.w.b). A significant increase in the starch level to 87.96 % d.w.b was observed.

Table 5. Chemical composition and starch damage of lab scale starch extraction.

Sample[1]	Moisture %[2]	Starch %[3]	Protein %[3]	Ash %[3]	Fat %[3]	Damage starch %[3]
Flour	8.1 (0.044)[a]	67.9 (0.03)[c]	15.6 (0.14)[a]	1.7 (0.016)[a]	1.4 (0.04)[a]	2.4 (0.042)[a]
LSE	6.0 (1.240)[b]	87.9 (0.84)[a]	1.4 (0.14)[b]	0.1 (0.016)[b]	0.1 (0.02)[b]	1.0 (0.032)[b]

[1]Abbreviation: Flour; yellow pea flour, LSE, lab scale starch extraction
[2]Values of mean (standard deviations).
[3]Dry weight-based values of mean (standard deviations). Values (n=2) with different letters in the same column are significantly different at $p \leq 0.05$.

4.2. Supercritical Fluid Extraction

The isolated starch from the lab (LSE) and industrial approach (ISE) were subjected to SC-CO_2 + EtOH extraction to study the impact on the chemical, pasting and functional properties of yellow pea starch and its application in food system. Therefore, moisture, protein, starch, ash, and fat content and starch damage level were determined. In addition to this, pasting, gel strength and functional properties were included.

4.2.1. Physicochemical Properties

The results of the two raw pea starch samples (LSE and ISE) were discussed separately in this section with some connection regarding the impact of SC-CO_2 + EtOH extraction in the starch extraction. Furthermore, the LSE was compared with previous study in yellow pea flour (Vatansever, Rao, et al., 2020). Overall, the SC-CO_2 + EtOH extraction caused a significant reduction ($p \leq 0.05$) in moisture, total starch, and fat content. No significant ($p \leq 0.05$) effect was determined for protein and ash contents after the extraction of both pea starches (Table 6).

Table 6. Proximate composition of non-extracted and SC CO_2 extracted pea starch samples.

[1]Treatments	Moisture %	Starch %	Protein %	Ash %	Fat %	Damage starch %
LSE	6.0 (1.240)^{b2}	87.9 (0.84)^{a}	1.4 (0.14)^{b}	0.1 (0.016)^{b}	0.1 (0.022)^{b}	1.0 (0.032)^{b}
LSE-SFE	2.9 (0.067)^{c}	83.8 (0.52)^{b}	1.4 (0.05)^{b}	0.1 (0.018)^{b}	0.05 (0.008)^{c}	1.0 (0.003)^{b}
Alkaline isolation						
ISE	9.5 (0.075)^{a}	88.5 (0.911)^{a}	1.4 (0.166)^{a}	0.3 (0.019)^{a}	0.1 (0.047)^{a}	0.4 (0.007)^{a}
ISE-SFE	3.4 (0.136)^{b}	84.0 (1.637)^{b}	1.4 (0.098)^{a}	0.3 (0.042)^{a}	0.06 (0.019)^{b}	0.3 (0.049)^{a}

[1]Abbreviation: LSE; lab scale starch extraction, LSE-SFE; lab scale starch extracted with supercritical fluid extraction, ISE; industrial scale starch extraction; ISE-SFE; industrial scale starch extracted with supercritical fluid extraction. [2]Values (standard deviation (n=4)) with different letters in the same column are significantly different at $p \leq 0.05$.

A substantial moisture reduction was observed in the LSE-SFE and ISE-SFE (Table 6), were reduced to 2.91% and 3.42%, respectively was observed. The results were in the same direction with previous studies (Brown et al., 2008; Solaesa et al., 2019; Vatansever, Rao, et al., 2020). The interaction between ethanol and SC-CO_2 during the extraction is thought to have dehydration effect in the extracted materials by increasing the solubility of the polar substance, such as water, thus increasing the moisture removal rate. For example, supercritical carbon dioxide has been considered as drying technique to reduce carrot moisture while maintaining a high-quality dry product (Brown et al., 2008). Furthermore, the oven-drying stage (30 min at 70 °C) that was applied to remove the ethanol after the extraction might contribute to more moisture reduction (Vatansever et al., 2020). The reduction moisture impact on whole quinoa flour after the extraction with SC-CO_2 was reported (Solaesa et al., 2019).

Even though most of the fat content was reduced previously by the starch isolation process, a significant removal ($p \leq 0.05$) of fat content after the SC-CO2 + EtOH was observed (Table 6). LSE-SFE and ISE-SFE had less fat ~~content~~, from 0.10% (d.w.b) and 0.13% (d.w.b) down to 0.05% (d.w.b) and 0.06% (d.w.b), respectively. A SC-CO_2 extraction was reported as a successful defatting method for soy and quinoa flour (Kang et al., 2017; Solaesa et al., 2019). The introduction of ethanol as cosolvent increase the solubility of lipid in SC-CO_2. This small reduction of fat levels after the SC- CO_2 extraction is expected to have limited impact on the functional properties of pea starch since most of the fat content was removed previously during starch isolation (Vatansever, Rao, et al., 2020).

No significant change occurred in the protein content after the extraction. However, a slight increase in the protein content were in the same direction of yellow pea flour treated with SC- CO_2 + Ethanol extraction (Vatansever, Rao, et al., 2020), and corn gluten meal treated with SC- CO_2 + Ethanol extraction (Cobb et al., 2018). The loss in the fat likely contributed to the increased protein content on a percentage basis.

When compared with SC-CO_2 + EtOH on whole yellow pea flour (Vatansever, Rao, et al., 2020), the results were similar with exception of total starch and starch damage. A significant decline in the starch content after the extraction were observed in LSE-SFE and ISC-SFE samples, from 87.96 % and 88.51% to 83.89% and 84.02% (d.w.b), respectively. This reduction of starch content might be associated with the interaction between pressure and temperature. A similar result was reported when starch first extracted from quinoa flour and then treated with high pressure (Ahmed et al., 2018). Treating quinoa starch for 15 min with 450 MPa and 600 MPa reduces the total starch from 64 to 60%. Liu et al. (2016) reported a decreasing of starch content in buckwheat during 20 min extraction with high pressure.

On the other hand, an increase in the resistant starch was determine in lentil after a combination of temperature and pressure extraction, which might cause starch nuclei formation and starch recrystallization (Ahmed et al., 2016). In this study, resistant starch was not determined, however, an expected increase in resistant starch might impact the functional and pasting properties of the starch.

Furthermore, the difference in total starch content compared to yellow pea flour extraction (Vatansever, Rao, et al., 2020) could be related to the concentration of ethanol during the extraction of SC-CO_2 + EtOH. In the current project, ethanol was poured into

the vessel and thus it was difficult to maintain the 22% ethanol concentration in the carbon dioxide, as was done by Vatansever, Rao, et al., 2020, throughout the extraction. Thus, differences in outcomes compared to previous researchers was like due to processing differences. No significant ($p<0.05$) different in the starch damage level was observed (Table 6). This result was consistent with previous study on quinoa (Solaesa et al., 2019).

4.2.2 Functional Properties

For a better understanding of pea starch applications in food production, information on changes in functional properties, as well as pasting properties, is essential (Vatansever, Rao, et al., 2020). The SC-CO2 + EtOH extraction had significantly impacted the functional and pasting properties of pea starch samples.

4.2.2.1 Water Holding Capacity (WHC)

We anticipated that WHC would increase after the treatment with SC-CO2 + EtOH extraction due to the possibility of starch, protein and fiber modification and lipid removal during the extraction might increase hydrophilic group interactions with water. As a result, solubility and WHC would increase. However, the opposite happened. The WHC of the pea starch samples SC-CO_2 + EtOH extraction was lower compared to non-SFE-treated pea starch samples. The reduction might be associated with the denaturation of protein during the extraction and lose of hydrophilic group in the starch being available to interact with water (Vatansever, Rao, et al., 2020). The LSE-SFE and ISE-SFE samples contained 1.37% (d.w.b) and 1.49% (d.w.b) of protein, respectively, which participate in the holding of water (Figure 6). Any structural change in the protein, denaturation for example, causing charged amino acids to interact with adjacent charged

amino acids, can result in less ion-dipole interactions with water. This would result in less protein and water hydrogen bonding (Vatansever, Rao, et al., 2020). In addition to this, the significantly lower starch content in the extracted samples could cause lower WHC. The increase in molecular mobility and structural change in pea starch can be other reasons for lower WHC. These changes could be caused by the high temperature of operation leading to weaker hydrogen bonding between starch and water molecules. Consequently, less swelling due to hydrogen bonding between starch and water will lead to lower peak viscosity of the starch (Sippel & Quiocho, 2015; Solaesa et al., 2019).

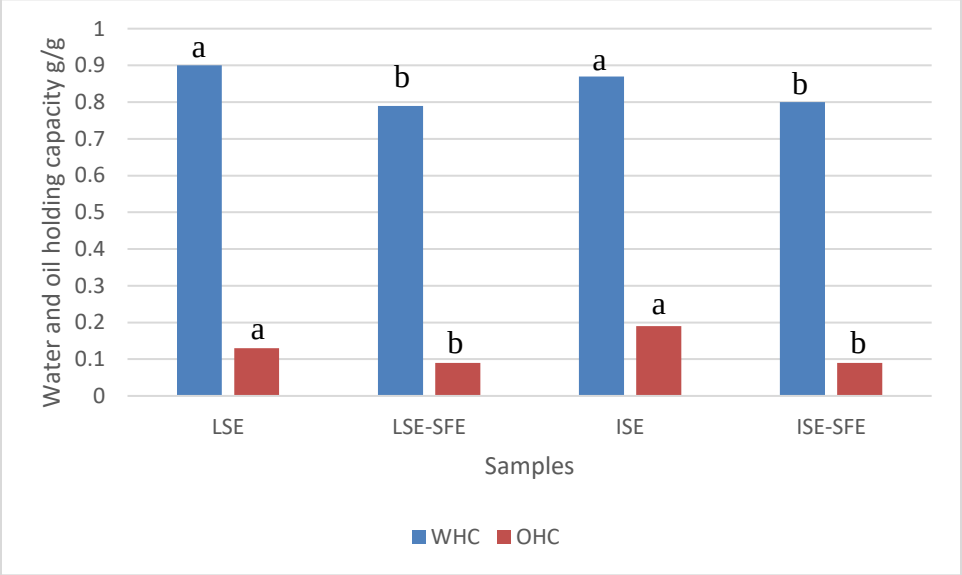

Figure 6. Functional characteristics of non-extracted and SC CO_2 extracted pea starch samples. Abbreviation: LSE; lab scale starch extraction, LSE-SFE; lab scale starch extracted with supercritical fluid extraction, ISE; industrial scale starch extraction; ISE-SFE; industrial scale starch extracted with supercritical fluid extraction, WHC; water holding capacity, OHC; oil holding capacity. Values with different letters in the same functionality test are significantly different at $p \leq 0.05$.

4.2.2.2 Oil Holding Capacity (OHC)

Oil primarily associates with the surface of hydrophobic amino acids and nonpolar chains of carbohydrates (e.g., dietary fiber) (Jitngarmkusol et al., 2008). Dietary fiber was not determined in this study, but from the literature it is expected to be between 4 to 4.5% in pea starch (L. Li et al., 2019). SC-CO_2 + EtOH extraction significantly impacted the OHC of treated pea starch samples (Figure 6). This lower OHC of LSE-SFE (0.06 g/g) and ISS-SFE (0.09 g/g), is comparable to previous study that subjected yellow pea flour to SC-CO_2 + EtOH extraction to remove undesirable flavor (Vatansever, Rao, et al., 2020). Pure starch has a very minor content of protein which will limit OHC. In this study, LSE-SFE and ISE-SFE have protein contents of 1.43 % and 1.49 % d.w.b., respectively (Table 6). However, the protein content was not significant, therefore, this reduction of OHC likely can be associated more with the denaturation of protein or the fiber in the sample (Vatansever, Rao, et al., 2020). Denaturation tends to bury hydrophobic interactions in the center of a denatured protein, and this would reduce the number of hydrophobic interactions available for oil binding.

4.2.2.3 Pasting Properties

Overall, the SC-CO_2 + EtOH extraction caused a significant reduction in all pasting parameters except pasting temperature, setback, and final viscosity (Table 7). The pasting properties of yellow pea starch samples were affected by the granule swelling rate indicated by peak viscosity and peak time (Ratnayake et al., 2001). Peak viscosity is the highest point of the pasting curve during the heating process or the maximum expansion of starch granules and represents the highest viscosity during the heating phase while

peak time is the time to reach peak viscosity (Karmakar et al., 2014). The lower peak viscosity in LSE-SFE (3634 cP) and ISE-SFE (6817 cP) compared to non-extracted pea starch samples alone is linked to the reduction in water absorption by the starch granules resulting in limited swelling along with amylose leaching during heating (Fox et al., 2014; Marta & Tensiska, 2017; Wani et al., 2016). A structural change in the treated pea starch samples, due to the high temperature (85 °C), might cause limited swelling of the starch granules (Solaesa et al., 2019). High heating treatment, over 60 °C, could result in reduction of amylose and amylopectin hydrogen bonds in the starch. When starch is heated, hydrogen bonds between polysaccharides can break. This can potentially increase leaching of amylose and thus the remaining polysaccharide would not cause as much granule swelling, resulting in a lower peak viscosity (Solaesa et al., 2019). Similar findings were observed for heat-moisture treated sweet potato starch (Marta & Tensiska, 2017) and brown rice flour (Kim et al., 2017).

Table 7. Pasting properties of non-extracted and SC CO_2 extracted pea starch samples.

Treatments[1]	Peak viscosity (cP)	Trough (cP)	Breakdown (cP)	Final viscosity (cP)	Setback (cP)	Peak time (min)	Pasting temp. (°C)
LSE	6668 (79.9)[a2]	3757 (129)[a]	2911 (55)[a]	11533 (1677)[a]	7776 (1591)[b]	4.4 (0.04)[a]	72.8 (0.34)[a]
LSE-SFE	4879 (879)[b]	1837 (141)[b]	3042 (793)[a]	10897 (701)[a]	9059 (652)[a]	4.1 (0.11)[b]	73.4 (0.26)[a]
Alkaline steeping							
ISE	8289 (175)[a]	4872 (107)[a]	3417 (100)[b]	13703 (1091)[a]	8832 (1020)[b]	4.28 (0.01)[a]	73.5 (0.08)[a]
ISE-SFE	6817 (536)[b]	2028 (77)[b]	4789 (488)[a]	12679 (540)[a]	10650 (576)[a]	4.27 (0.05)[a]	73.9 (0.52)[a]

[1]Abbreviation: LSE; lab scale starch extraction, LSE-SFE; lab scale starch extracted with supercritical fluid extraction, ISE; industrial scale starch extraction; ISE-SFE; industrial scale starch extracted with supercritical fluid extraction.
[2]Values (standard deviation (n=4)) with different letters in the same column are significantly different at $p \leq 0.05$.

Breakdown viscosity reflects the ability of starch to resist heating and shear stress during cooking (Ohizua et al., 2017). A significant increase in the breakdown of LSE-SFE (3042 cP) and ISE-SFE (4789 cP) was recorded after the extraction compared to non-extracted sample (Table 7). Typically, pea starch has high resistance to collapse due to less shear-thinning behavior of high amylose starch content; however, reducing amylose content or changing in the structure could enhance shear-thinning behavior (Simsek 2009). The SC-CO_2 + EtOH extraction might reduce resistant starch due to changes in the starch structure that would impact amylose. Amylose and resistance starch are less soluble and resist shear thinning (Delcour & Hoseney, 2010). Therefore, a reduction in them might result in higher breakdown viscosity.

The lower final viscosity after the extraction might be the result of the weak reassociation between starch polysaccharides. This might be attributed to possible starch modification during the extraction of SC-CO_2 + EtOH (Vatansever, Rao, et al., 2020). The setback is an important indicator of retrogradation and syneresis of paste (Rakshit, 2014). The setback was increased after the extraction. The higher the setback value, the more likely it is to form a gel during cooling. The high value of setback suggests a tendency of retrogradation (Rakshit, 2014).

Overall, the decreasing of peak viscosity, trough, and final viscosity was similar to yellow pea flour with SC-CO_2 + EtOH extraction (Vatansever, Rao, et al., 2020). Depending on the final use of the starch, high breakdown would not be a problem for instant products. Pasting properties determination is important to predict final product texture and palatability (Wani et al., 2016). Although the pea used in the present research was yellow, the results would be expected to be similar for other pea varieties. The

variability in protein and starch of the peas utilized in the study would like account for differences in the outcome. The results support that SC-CO_2 + EtOH extraction influenced whole yellow pea starch functionality. This extraction caused reductions in WHC and OHC of the starch, which would impact the final product quality regarding texture, acceptability, and shelf life. In addition to this, changes in functional properties most likely impacted the pasting properties of starch. Therefore, the extraction can diversify yellow pea starch application in food system.

4.3 Ethanol Extraction of Pea Starch

4.3.1 Moisture Content

The moisture content of the raw starch was (9.54 %), which is consistent with literature (Ratnayake et al., 2001) and within the range of legumes starch (Wani et al., 2016). The treated starch had a significantly ($p \leq 0.05$) lower moisture content compared to the untreated starch (Table 8). This reduction is associated with the drying stage after the treatment. In addition to this, the used of ethanol as solvent and milling is thought to have additional drying effect on the treated starch (Gohl, 2019).

Table 8. Chemical composition and starch damage of ethanol extracted starch.

Treatment[1]	Moisture%[2]	Protein %[3]	Total starch %[3]	Ash %[3]	Starch Damage %[3]
ISE	9.5 (0.075)[a]	1.4 (0.166)[a]	88.5 (0.91)[b]	0.3 (0.019)[a]	0.4 (0.007)[b]
ETS	7.2 (1.235)[b]	1.4 (0.048)[a]	90.4 (0.93)[a]	0.1 (0.016)[b]	0.5 (0.045)[a]

[1]Abbreviation: ISE; industrial scale starch extraction; ETS; ethanol treated starch.
[2]Values as it is (standard deviation (n=4)). Values (standard deviation (n=4)) with different letters in the same column are significantly at $p \leq 0.05$.

4.3.2 Protein Content

There was no significant change in the protein content between raw and treated samples (Table 8). This result agreed with the findings reported by Gohl (2019). However, there was slight increase in the protein percentage, which might be due to the decrease of moisture, and ash contents as part of the mass balance assessment.

4.3.3 Ash Content

The ash content of the raw pea starch (0.34% d.w.b) was consistence with isolated pea starch previously reported (Vatansever et al., 2021). Similar reduction in the ash content of lentil was reported when treated with 35% and 55% ethanol (Chang et al., 2019). Ash is considered as the amount of minerals in starch (King Arthur Flour, 2019). Ethanol extractions were able to reduce the ash content significantly ($p \leq 0.05$) in the treated starch to 0.18% (Table 8). Moreover, Kajihausa et al. (2014) reported that increasing the soaking time in water lowered the ash content of sesame flour. This observation suggests that water (52.5%) in the ethanol solution was sufficient to influence mineral removal in the treated pea starch.

4.3.4 Total Starch

A significantly ($p \leq 0.05$) higher total starch in the treated starch was recorded at 83.15% (Table 8). Total starch accounted for all plant starch, maltodextrins, maltose, and isomaltose present (Megazyme, 2019). There is no mechanism by which starch could have been added to the treated material, hence it is assumed that the significant increase in total starch content is attributable to the loss of moisture, ash and small molecular weight carbohydrates (sugars) from the treated starch.

4.3.5 Starch Damage

Starch damage in treated pea starch was significantly greater ($p \leq 0.05$) than in raw pea starch, with values of 0.48% and 0.36% d.w.b., respectively. While statistically significant, it is proposed that this difference is quite minor. A damaged starch granule is one that has been physically broken or fragmented, which increases water absorption (Arya, Sadawarte, & Waghmare, 2015). Starch damage happens often during the milling process because of a combination of heat produced and physical stress. The formation of minor starch degradation within the pea is thought to have occurred during the starch milling after the ethanol extraction to reduce particle size (Gohl, 2019). Similarly, Okada, Negishi, and Nagao (1986) discovered a link between the quantity of starch degradation and the number of re-grinding cycles.

4.3.6 Functional Properties

4.3.6.1 Water Holding Capacity (WHC)

The WHC results (Table 9) showed no significant difference between treated and non-treated starch. However, a slight increase in WHC was thought to be associated with the increasing amount of starch damage and total starch in the treatment sample. A similar result was observed in pea protein enrichment flour that was treated with 20% to 50% ethanol concentration (Wang et al., 2020).

Table 9. Water holding capacity (WHC) and oil holding capacity (OHC) of ethanol extracted starches.

Teatment[1]	WHC %[2]	OHC %[2]
ISE	0.876 (0.062)[a]	0.19 (0.107)[a]
ETS	0.960 (0.028)[a]	0.05 (0.008)[b]

[1]Abbreviation: ISE; industrial scale starch extraction; ETS; ethanol treated starch.
[2]Values (standard deviation (n=4)) with different letters in the same column are significantly different at $p \leq 0.05$.

4.3.6.2 Oil Holding Capacity (OHC)

The OHC (Table 9) was similar to results from a study that was done in pea protein flour enrichment (Wang et al., 2020). The results show a significant reduction in the OHC. Oil bonds with protein. Thus, this reduction might be associated with the denaturation of protein since the treated samples had higher protein content. During denaturation, the protein aggregates such that the hydrophobic parts of the protein will interact and orient to the center of the denatured protein. This leaves hydrophilic amino acids on the exterior surface where they are less likely to interact with oil. This theory also supports the higher, although not significant, WHC of the treated sample.

4.3.7 Pasting Properties

Statistically, no significant impact of the treatment was observed in the peak viscosity among samples (Table 10). However, a slight reduction was noted and might be related to the high degree of starch damage in the treated starch. With high starch damage, low viscosity is expected (Wu et al., 2018). Moreover, the denaturation of protein can be another reason (Oppong Siaw et al., 2021). Denaturation disrupts

hydrogen bonds in favor of hydrophobic interactions. Compared to raw starch, setback was significantly ($p \leq 0.05$) lower in treated pea starch. Setback is the difference between the hot paste (Trough) viscosity and the final viscosity that occurs during the retrogradation process (Balet et al., 2019b). Reduced setback has been associated with starch damage and reduced particle size (Elliot, Dang, & Bason, 2019), both of which were evident in the treated pea flours.

The final viscosity values were not statistically ($P > 0.05$) different between treatments. Unlike Hillen (2016), who discovered that a 50:50 ethanol/water HPSE treatment reduced the final viscosity from 2821 to 1941 cP. In contrast to the treatment used in this investigation, Hillen (2016) used pressure, which, according to previous publications, appeared to have a detrimental influence on the final viscosity of the treated pea flour (J. Ahmed et al., 2017). Final viscosity is frequently used as the major measure to predict product quality, indicating the capacity of the flour/starch to form a paste or gel after cooking and cooling (Balet et al., 2019b). Therefore, treated starch would have less tendency to retrograde than non-treated starch due to the higher WHC (Table 10).

Table 10. Pasting properties of ethanol treated starch.

Treatment[1]	Peak viscosity (cP)	Trough (cP)	Breakdown (cP)	Final viscosity (cP)	Setback (cP)	Peak time (min)	Paste temperature. (°C)
ISE	8289 (175)[a2]	4872 (107)[a]	3417 (100)[b]	13703 (1091)[a]	8832 (1020)[b]	4.28 (0.01)[a]	73.5 (0.08)[a]
ETS	8044 (181)[a]	4877 (78.2)[a]	3167 (219)[a]	13434 (1913)[a]	8557 (1912)[a]	4.3 (0.03)[a]	74.1 (0.39)[a]

[1]Abbreviation: ISE; industrial scale starch extraction; ETS; ethanol treated starch.
[2]Values (standard deviation (n=4)) with different letters in the same column are significantly different at $p \leq 0.05$.

4.3.8. Texture Profile:

The hardness of starch gels is a significant textural parameter that reflects gel strength. The high value of gel strength (920 g) after the extraction of starch agreed with the findings of alcohol-soaked corn starch (Sun et al., 2020). The increase can be attributed to the leaching of amylose due to starch damage and ethanol treatment. Leaching of amylose can improve the strength and elasticity of the gel structure and create a more tightly packed structure. Sandhu and Kaur (2004) reported that gel hardness was primarily generated by retrogradation of starch gels, which was coupled with water syneresis and amylopectin crystallization, resulting in harder gels (N. Singh et al., 2004).

Overall, the pea starch (ISE) obtained as described in starch isolation section was treated with 47.5% ethanol for 63 min to determine the impact of the treatment on pea starch chemical, functional and pasting properties. The extraction had significantly reduced the moisture and ash contents while an increase in the total starch and starch damage was observed. The OHC of pea starch was lower after the treatment. No significant change regarding the pasting properties occurred; however, the hardness of gel was increased dramatically post ethanol treatment. The findings of this study may be utilized by processors to better understand the composition and functioning of treated pea starch. The treated starch can be used in snack that require high gel properties or noodle product.

4.4. Food Application

4.4.1. Physical Analysis

4.4.1.1. Gel Strength

In general, yogurt made with different types and treated starches exhibited harder texture with storage time. Yogurt texture is ultimately determined by the physical interaction among the casein micelles (Guven et al., 2005). A significant ($p<0.05$) higher hardness valve was recorded for pea starch samples compared to the control. This can be associated with the high amylose content in pea starch compared to the control (corn starch) (Sun & Xiong, 2014). This high amount of amylose can form a stronger network (Saleh et al., 2020). ISE-SFE had the highest hardness valve after 6 days of storage (Table 11), which can be related to the low water holding capacity and high setback viscosity value, respectively. A continual increase in hardness until day 6 was noticed. This study was done for a short time period and thus different results may occur for longer term studies. When treated starch samples were applied to pudding, the pea starch samples had significantly higher hardness compared to the control (Table 12). The greatest hardness was observed in ISE-SFE on day 5.

Table 11. Hardness of yogurt formulated with pea starch (ISE), pea starch treated with supercritical fluid extraction (ISE-SFE), ethanol extraction (ETS) and modified corn starch (control).

Treatment[1]	Day 1[2]	Day 2	Day 3	Day 4	Day 5	Day 6
Control	18.0 (0.88)^{b2}	19.7 (1.74)^{a}	20.6 (1.89)^{a}	20.2 (1.54)^{a}	20.6 (0.50)^{a}	20.3 (0.73)^{a}
ISE	36.7 (0.55)^{e}	53.8 (1.34)^{d}	55.7 (0.72)^{c}	55.0 (1.51)^{c}	57.3 (0.77)^{b}	58.5 (0.45)^{a}
ISE-SFE	38.4 (1.14)^{f}	48.7 (0.93)^{e}	51.6 (2.26)^{d}	56.8 (1.44)^{c}	58.9 (0.34)^{b}	61.0 (1.39)^{a}
ETS	40.4 (0.54)^{f}	41.6 (0.50)^{e}	43.8 (0.42)^{d}	47.3 (0.81)^{c}	49.6 (1.77)^{b}	55.2 (0.54)^{a}

[1]Abbreviation: Control; modified corn starch, ISE; industrial scale starch extraction, ISE-SFE; industrial scale starch treated with supercritical fluid extraction, ETS; ethanol treated starch.
[2]Values (standard deviation (n=6)) with different letters in the same column are significantly different at $p \leq 0.05$.

Table 12. Hardness of pudding formulated with pea starch (ISE), pea starch treated with supercritical fluid extraction (ISE-SFE), ethanol extraction (ETS) and modified corn starch (control).

Treatment[1]	Day 1[2]	Day 2	Day 3	Day 4	Day 5
Control	19.3 (1.42)^{b}	20.1 (0.64)^{b}	20.6 (1.58)^{a}	22.4 (1.34)^{a}	24.2 (0.70)^{a}
ISE	68.0 (3.02)^{e}	80.2 (1.21)^{d}	83.6 (1.71)^{c}	93.3 (2.81)^{b}	101.4 (1.81)^{a}
ISE-SFE	80.8 (2.74)^{d}	83.0 (1.27)^{d}	92.0 (1.92)^{c}	99.5 (1.95)^{b}	123.2 (1.62)^{a}
ETS	42.3 (1.83)^{e}	45.1 (1.56)^{d}	51.1 (1.43)^{c}	56.4 (1.44)^{b}	64.5 (0.67)^{a}

[1]Abbreviation: Control; modified corn starch, ISE; industrial scale starch extraction, ISE-SFE; industrial scale starch treated with supercritical fluid extraction, ETS; ethanol treated starch.
[2]Values (standard deviation (n=6)) with different letters in the same column are significantly different at $p \leq 0.05$.

4.4.1.2. Syneresis

Set yogurt exhibits a negative trait called "wheying-off," which occurs when the whey is expelled from the casein network. Spontaneous wheying-off happens when the gel network is unstable, and the whey separates without external force. This can occur due to increased rearrangements of the gel matrix or weak gel network due to mechanical damage. The ability of starch to control acidity could be another reason. Acidity has a major role in increasing whey separation in yogurt. During the storage, the lactic acid is produced and cause an increase in the acidity (Ahmed et al., 2021). This increasing acidity causes a breakdown of the calcium bonds and causes the casein to shrink and release whey. Starch can prevent lactose conversion (Altemimi, 2018). Therefore, manufacturers use stabilizers like starch, pectin, and gelatin to prevent wheying-off (Mwizerwa et al., 2017; Q. Zhao et al., 2009). Wheying-off in yogurt can be caused by several factors, including extended incubation time, imbalanced ratio of whey protein to casein, low solid content, and mishandling of the product during storage and distribution. To reduce wheying-off, yogurt can be treated with stabilizers such as 1% crosslinked cassava, corn starch, and tapioca starch, which have been found to significantly decrease yogurt syneresis (Mwizerwa et al., 2017).

Regarding wheying-off, in general, no significant differences in syneresis was observed among starches. There was a slight increase in syneresis value in ISE-SFE after 6 days compared to the other samples. A longer storage time is recommended to determine if differences in syneresis become apparent.

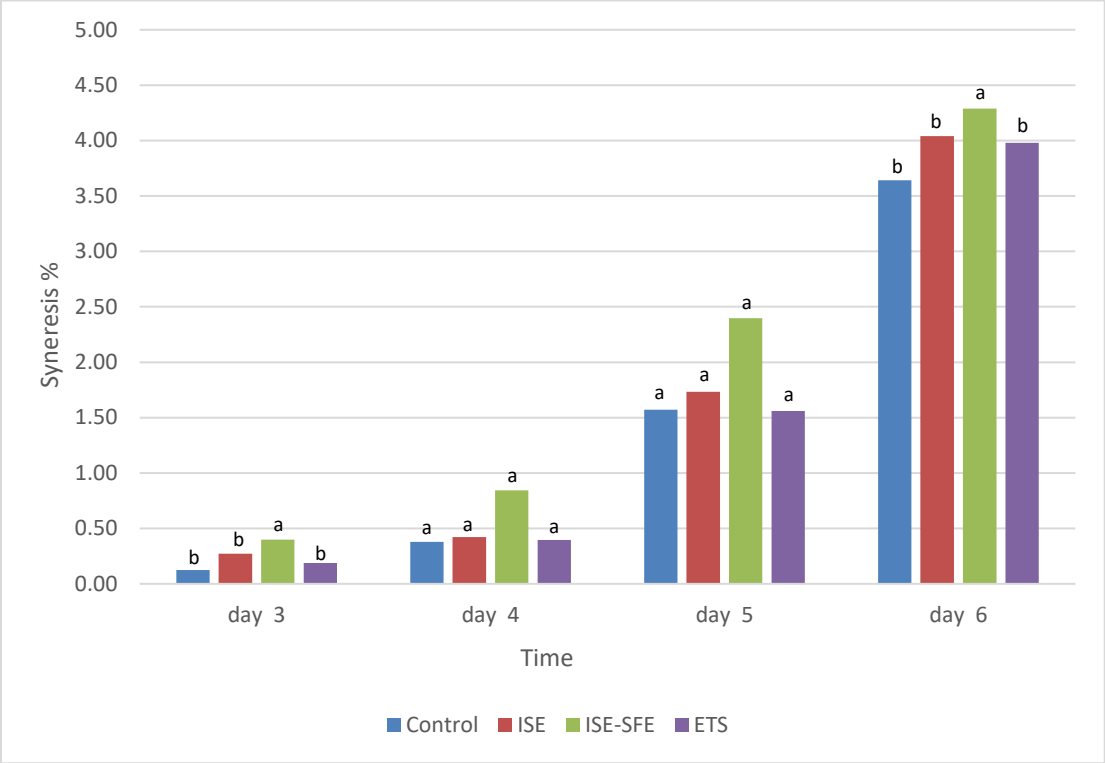

Figure 7: Syneresis of yogurt formulated with pea starch (ISE), pea starch treated with supercritical fluid extraction (ISE-SFE), ethanol extraction (ETS) and modified corn starch (control). Values (standard deviation (n=2)) with different letters in the same day are significantly different at $p \leq 0.05$.

A significant high syneresis was observed in the pudding with pea starch compared to the control (Figure 8). Pudding made with ETS treatment had the lowest syneresis compared to ISE and ISE-SFE. Unexpected result of the ISE-SFE was associated with the low water holding capacity and high setback value (Table 7, Figure 6).

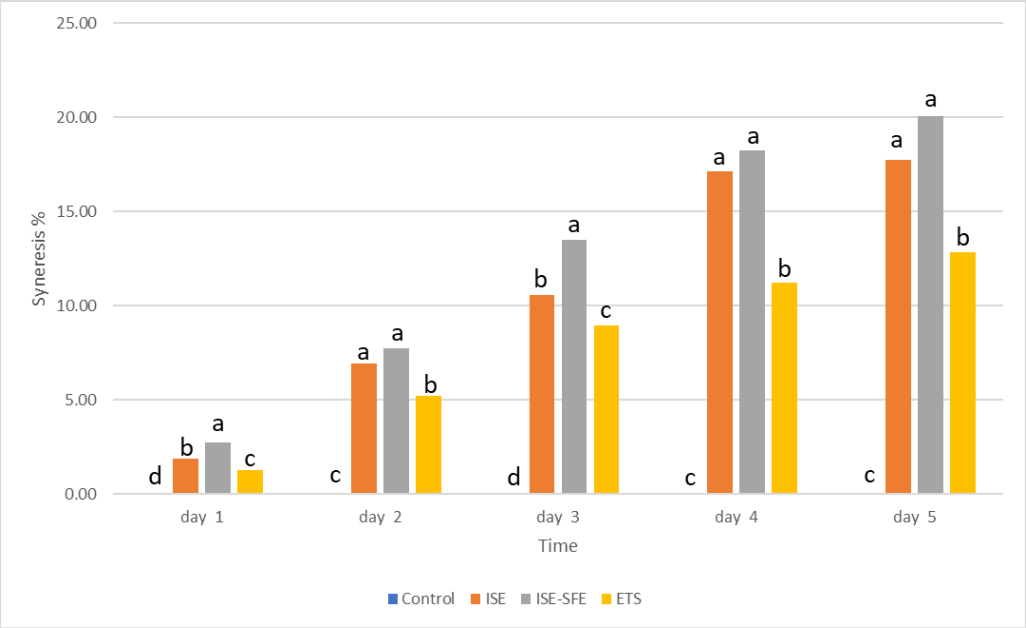

Figure 8. Syneresis of pudding formulated with pea starch (ISE), pea starch treated with supercritical fluid extraction (ISE-SFE), ethanol extraction (ETS) and modified corn starch (control). Values (standard deviation (n=2)) with different letters in the same day are significantly different at $p \leq 0.05$.

5. SUMMARY AND CONCLUSION

The method of alkaline steeping and dehulling produced a high purity starch. The approach used to support the observations includes four different pea materials, two were derived from whole pea and two from dehulled pea. Using alkaline water as solvent, a significant drop in the protein content in all samples was observed; however, dehulled sample had the lowest protein content compared to the whole seed samples. Dehulling prior to the extraction eased the separation of starch granules from the protein bodies since the fiber was removed, which interfered with the sieving operation. Dehulling did not impact the pasting or functional properties of the starch. To conclude, dehulling prior

to extraction improved the extraction yield and purity of starch and made available hulls to be used in other applications. Isolating starch by the dehulling method is the most suitable way to produce starch from yellow dry peas for food ingredient utilization.

Extracted starch with SC CO_2 + EtOH cause a reduction in peak viscosity and hot paste viscosity with significant setback in all samples. These results were unexpected compared with previous studies in yellow pea flour. This can be associated with the SFE parameters, especially ethanol that was not maintained at the desired 22% concentration during the extraction.

Ethanol treatment show similar results to previous study that was done in yellow pea flour (Gohl, 2019). ISE sample was treated with 47.5% ethanol for 63 min to determine the impact of the treatment on pea starch chemical, physicochemical and pasting properties. The extraction reduced moisture and ash contents while an increase in the total starch and starch damage. The OHC of pea starch was lower after the treatment. No significant changes regarding the pasting properties were observed in ETS; however, the hardness of gel increased dramatically post ethanol treatment. The findings of this study may be utilized by processors to better understand the composition and functioning of treated pea starch, allowing them to decide which products are most suited for usage with treated pea starch as an ingredient.

Regardless of the starch treatment, correlation between time (day of storage) and syneresis was observed. The pudding sample made with pea starch had higher syneresis compared to the control. The ETS had syneresis and hardness values most similar to the control. On the other hand, ISE-SFE had substantial release of water with increasing

hardness after 3 days of storage. Therefore, ISE-SFE could be used in other food applications that are not affected by high syneresis rates and required hardness. For example, ISE-SFE could be applied to extruded snacks and noodles where high setback value is preferred. Finally, based on the results of the extracted samples with SC CO_2 + EtOH and with comparison with previous studies, extraction of the whole flour instead of starch might have less impact on the starch properties. However, if high setback value is preferable, SC CO_2 + EtOH extraction (temperature (85 °C), pressure (427 bar), CO_2 flow rate (1L/min)) is recommended. These unexpected results, as compared to previous reports, of SC CO_2 extraction were likely related to the difference in the ethanol concentration than the aimed parameters.

6. FUTURE DIRECTIONS OR RECOMMENDATIONS

Results indicated that pea starch had a positive final yogurt texture and syneresis; however, a comparison with control sample (without starch) with longer period of time (15 days), is needed to determine the impact of the additional of pea starch in the shelf life compared with the control.

In this study, the impact of isolation of pea starch, ethanol treatment, and SC CO_2 extraction on chemical, functional and pasting properties were determined. Further research is needed to study the impact of these processes on the amylose/amylopectin ratio, resistant starch, sugar and fiber content, particle size determination, thermal characteristics, starch digestibility, and x-ray diffraction. This extended research would give more understanding of pea starch characteristics. Expansion of treated pea starch

application with sensory evaluation is needed to examine the starch in different food products, for example noodles, and consumer acceptability.

LITERATURE CITED

AACCI. 2010. Method 08-01.01, 44-15.02, 46-30.01, 30-10.01, 56-35.01, 61-02.01, 56-37.01, 56-36.01 and 76-13.01. in: Approved methods of analysis (11th ed.). St. Paul, MN, USA: American Association of Cereal Chemists.

Ahmed, J., Mulla, M. Z., & Arfat, Y. A. (2017). Particle size, rheological and structural properties of whole wheat flour doughs as treated by high pressure. *International Journal of Food Properties*, *20*(8), 1829–1842. https://doi.org/10.1080/10942912.2016.1220394

Ahmed, J., Thomas, L., Arfat, Y. A., & Joseph, A. (2018). Rheological, structural and functional properties of high-pressure treated quinoa starch in dispersions. *Carbohydrate Polymers*, *197*, 649–657. https://doi.org/10.1016/j.carbpol.2018.05.081

Ahmed, J., Thomas, L., Taher, A., & Joseph, A. (2016). Impact of high-pressure treatment on functional, rheological, pasting, and structural properties of lentil starch dispersions. *Carbohydrate Polymers*, *152*, 639–647. https://doi.org/10.1016/j.carbpol.2016.07.008

Ahmed, M. E., Rathnakumar, K., Awasti, N., Elfaruk, M. S., & Hammam, A. R. A. (2021). Influence of probiotic adjunct cultures on the characteristics of low-fat Feta cheese. *Food Science and Nutrition*, *9*(3), 1512–1520. https://doi.org/10.1002/fsn3.2121

Alavitalab, H. (2016). *Starch-protein extraction and separation it from green pea Preparation of PUFA from Hypophthalmichthys molitrix oil via urea complexation View project Starch-protein extraction and separation it's from green pea View project*. www.european-science.com

Altemimi, A. B. (2018). Extraction and optimization of potato starch and its application as a stabilizer in yogurt manufacturing. *Foods*. https://doi.org/10.3390/foods7020014

Amarakoon, D., Thavarajah, D., McPhee, K., & Thavarajah, P. (2012). Iron-, zinc-, and magnesium-rich field peas (Pisum sativum L.) with naturally low phytic acid: A potential food-based solution to global micronutrient malnutrition. *Journal of Food Composition and Analysis*, *27*(1), 8–13. https://doi.org/10.1016/j.jfca.2012.05.007

Amaral, G. v., Silva, E. K., Cavalcanti, R. N., Cappato, L. P., Guimaraes, J. T., Alvarenga, V. O., Esmerino, E. A., Portela, J. B., Sant' Ana, A. S., Freitas, M. Q., Silva, M. C., Raices, R. S. L., Meireles, M. A. A., & Cruz, A. G. (2017). Dairy processing using supercritical carbon dioxide technology: Theoretical fundamentals, quality and safety aspects. In *Trends in Food Science and Technology*, (64), 94-101. https://doi.org/10.1016/j.tifs.2017.04.004

Anderson, V. L., Lardy, G. P., & Uffelman, B. R. (2014). Field Pea Grain and Forage for Beef Cattle. www.ag.ndsu.edu

Asif, M., Rooney, L. W., Ali, R., & Riaz, M. N. (2013). Application and Opportunities of Pulses in Food System: A Review. *Critical Reviews in Food Science and Nutrition*, *53*(11), 1168–1179. https://doi.org/10.1080/10408398.2011.574804

Balet, S., Guelpa, A., Fox, G., & Manley, M. (2019). Rapid Visco Analyser (RVA) as a Tool for Measuring Starch-Related Physiochemical Properties in Cereals: A Review. In *Food Analytical Methods*. https://doi.org/10.1007/s12161-019-01581-w

Baugreet, S., Kerry, J. P., Brodkorb, A., Gomez, C., Auty, M., Allen, P., & Hamill, R. M. (2018). Optimisation of plant protein and transglutaminase content in novel beef restructured steaks for older adults by central composite design. *Meat Science*, *142*, 65–77. https://doi.org/10.1016/j.meatsci.2018.03.024

Boukid, F., Rosell, C. M., & Castellari, M. (2021). Pea protein ingredients: A mainstream ingredient to (re)formulate innovative foods and beverages. In *Trends in Food Science and Technology*. https://doi.org/10.1016/j.tifs.2021.02.040

Bourré, L., McMillin, K., Borsuk, Y., Boyd, L., Lagassé, S., Sopiwnyk, E., Jones, S., Dyck, A., & Malcolmson, L. (2019). Effect of adding fermented split yellow pea flour as a partial replacement of wheat flour in bread. *Legume Science*, *1*(1). https://doi.org/10.1002/leg3.2

Boye, J., Zare, F., & Pletch, A. (2010). Pulse proteins: Processing, characterization, functional properties and applications in food and feed. In *Food Research International* 43(2), 414–431. https://doi.org/10.1016/j.foodres.2009.09.003

Brown, Z. K., Fryer, P. J., Norton, I. T., Bakalis, S., & Bridson, R. H. (2008). Drying of foods using supercritical carbon dioxide - Investigations with carrot. *Innovative Food Science and Emerging Technologies*, *9*(3), 280–289. https://doi.org/10.1016/j.ifset.2007.07.003

Brummer, Y., Kaviani, M., & Tosh, S. M. (2015). Structural and functional characteristics of dietary fibre in beans, lentils, peas and chickpeas. *Food Research International*, *67*, 117–125. https://doi.org/10.1016/j.foodres.2014.11.009

Campos-Vega, R., Loarca-Piña, G., & Oomah, B. D. (2010). Minor components of pulses and their potential impact on human health. In *Food Research International*. https://doi.org/10.1016/j.foodres.2009.09.004

Capuzzo, A., Maffei, M. E., & Occhipinti, A. (2013). Supercritical fluid extraction of plant flavors and fragrances. In *Molecules*, 18(6),7194–7238. https://doi.org/10.3390/molecules18067194

CDC. (2020). *National Diabetes Statistics Report 2020. Estimates of diabetes and its burden in the United States.* National Diabetes Statistics Report 2020. Estimates of diabetes and its burden in the United States. (cdc.gov).

Chandra, S., Singh, S., & Kumari, D. (2015). Evaluation of functional properties of composite flours and sensorial attributes of composite flour biscuits. *Journal of Food Science and Technology*, *52*(6), 3681–3688. https://doi.org/10.1007/s13197-014-1427-2

Chang, C., Stone, A. K., Green, R., & Nickerson, M. T. (2019). Reduction of off-flavours and the impact on the functionalities of lentil protein isolate by acetone, ethanol, and isopropanol treatments. *Food Chemistry*, *277*, 84–95. https://doi.org/10.1016/j.foodchem.2018.10.022

Chung, H. J., Liu, Q., & Hoover, R. (2010). Effect of single and dual hydrothermal treatments on the crystalline structure, thermal properties, and nutritional fractions of pea, lentil, and navy bean starches. *Food Research International*, *43*(2), 501–508. https://doi.org/10.1016/j.foodres.2009.07.030

Cobb, B. F., Kallenbach, J., Hall, C. A., & Pryor, S. W. (2018). Optimizing the Supercritical Fluid Extraction of Lutein from Corn Gluten Meal. *Food and Bioprocess Technology*, *11*(4), 757–764. https://doi.org/10.1007/s11947-017-2052-7

Coon, R. C., Olson, F., Bangsund, D. A., & Hodur, N. M. (2015). *Give to AgEcon Search Contribution of the Pulse Crop Industry To the Economies of North Dakota and Eastern Montana*. http://agecon.lib.umn.edu/.

Cruz-Suarez, L. E., Ricque-Marie, D., Tapia-Salazar, M., McCallum, I. M., & Hickling, D. (2001). Assessment of differently processed feed pea (Pisum sativum) meals and canola meal (Brassica sp.) in diets for blue shrimp (Litopenaeus stylirostris). Aquaculture, 196(1–2), 87–104. https://doi.org/10.1016/S0044-8486(00)00572-X

Dahl, W. J., Foster, L. M., & Tyler, R. T. (2012). Review of the health benefits of peas (Pisum sativum L.). In *British Journal of Nutrition*, 108, (1). https://doi.org/10.1017/S0007114512000852

Damian, C. & O. A. (2014). *influence of dietary fiber from peas on some quality characteristics of yoghurts*.

Davydova, N. I., Leont'ev, S. P., Genin, Y. V., Sasov, A. Y., & Bogracheva, T. Y. (1995). Some physico-chemical properties of smooth pea starches. *Carbohydrate Polymers*, *27*(2), 109–115. https://doi.org/10.1016/0144-8617(95)00052-9

De Almeida Costa, G. E., Da Silva Queiroz-Monici, K., Pissini Machado Reis, S. M., & De Oliveira, A. C. (2006). Chemical composition, dietary fibre and resistant starch contents

of raw and cooked pea, common bean, chickpea and lentil legumes. *Food Chemistry*, *94*(3), 327–330. https://doi.org/10.1016/j.foodchem.2004.11.020

de Souza Gomes, D., do Prado Cordoba, L., Rosa, L. S., Spier, M. R., Schnitzler, E., & Waszczynskyj, N. (2018). Thermal, pasting properties and morphological characterization of pea starch (Pisum sativum L.), rice starch (Oryza sativa) and arracacha starch (Arracacia xanthorrhiza) blends, established by simplex-centroid design. *Thermochimica Acta*, *662*, 90–99. https://doi.org/10.1016/j.tca.2018.02.011

Debet, M. R., & Gidley, M. J. (2006). Three classes of starch granule swelling: Influence of surface proteins and lipids. *Carbohydrate Polymers*, *64*(3), 452–465. https://doi.org/10.1016/j.carbpol.2005.12.011

Do, Q. D., Angkawijaya, A. E., Tran-Nguyen, P. L., Huynh, L. H., Soetaredjo, F. E., Ismadji, S., & Ju, Y. H. (2014). Effect of extraction solvent on total phenol content, total flavonoid content, and antioxidant activity of Limnophila aromatica. *Journal of Food and Drug Analysis*, *22*(3), 296–302. https://doi.org/10.1016/j.jfda.2013.11.001

Dueñas, M., Estrella, I., & Hernández, T. (2004). Occurrence of phenolic compounds in the seed coat and the cotyledon of peas (Pisum sativum L.). *European Food Research and Technology*, *219*(2), 116–123. https://doi.org/10.1007/s00217-004-0938-x

Durstine, J. L., Gordon, B., Wang, Z., & Luo, X. (2013). Chronic disease and the link to physical activity. In *Journal of Sport and Health Science*, 2(1),3–11. Elsevier. https://doi.org/10.1016/j.jshs.2012.07.009

Espinosa-Ramírez, J., Garzon, R., Serna-Saldivar, S. O., & Rosell, C. M. (2018). Mimicking gluten functionality with β-conglycinin concentrate: Evaluation in gluten free yeast-leavened breads. *Food Research International*, *106*, 64–70. https://doi.org/10.1016/j.foodres.2017.12.055

FAO. (2016). *pulses: nutritious seeds for a sustainable future*.

Ferreira, C. D., Ziegler, V., Lindemann, I. da S., Hoffmann, J. F., Vanier, N. L., & Oliveira, M. de. (2018). Quality of black beans as a function of long-term storage and moldy development: Chemical and functional properties of flour and isolated protein. *Food Chemistry*, *246*, 473–480. https://doi.org/10.1016/j.foodchem.2017.11.118

Fox, G., Visser, J., Skov, T., Meijering, I., & Manley, M. (2014). Effect of different analysis conditions on Rapid Visco Analyser malt viscograms in relation to malt of varying fermentability. *Journal of the Institute of Brewing*, *120*(3), 183–192. https://doi.org/10.1002/jib.137

Godswill Awuchi, C., Kate Echeta, C., Godswill, C., Somtochukwu, V., & Kate, C. (2019). The Functional Properties of Foods and Flours. In *International Journal of Advanced Academic Research | Sciences*, 5(11). https://www.researchgate.net/publication/337403804

Gohl, M. T. (2019). Flavor Modification of Pea Flour Using Ethanol-Based Deodorization. https://library.ndsu.edu/ir/handle/10365/31568

Gularte, M. A., Gómez, M., & Rosell, C. M. (2012). Impact of Legume Flours on Quality and In Vitro Digestibility of Starch and Protein from Gluten-Free Cakes. *Food and Bioprocess Technology*, *5*(8), 3142–3150. https://doi.org/10.1007/s11947-011-0642-3

Guven, M., Yasar, K., Karaca, O. B., & Hayaloglu, A. A. (2005). The effect of inulin as a fat replacer on the quality of set-type low-fat yogurt manufacture. *International Journal of Dairy Technology*, *58*(3), 180–184. https://doi.org/10.1111/j.1471-0307.2005.00210.x

Gwirtz, J. A., & Garcia-Casal, M. N. (2014). Processing maize flour and corn meal food products. *Annals of the New York Academy of Sciences*, *1312*(1), 66–75. https://doi.org/10.1111/nyas.12299

Hall, C. (2019). *2019 U.S. Pulse Quality Survey*.

Hall, C. (2020). *2020 U.S. Pulse Quality Survey*.

Hall, C. (2021). *2021 U.S. Pulse Quality Survey*.

Hanan, E., Rudra, S. G., Sagar, V. R., & Sharma, V. (2020). Utilization of pea pod powder for formulation of instant pea soup powder. *Journal of Food Processing and Preservation*, *44*(11). https://doi.org/10.1111/jfpp.14888

Haros, M., Tolaba, M. P., & Suárez, C. (2003). Influence of corn drying on its quality for the wet-milling process. *Journal of Food Engineering*, *60*(2), 177–184. https://doi.org/10.1016/S0260-8774(03)00038-4

Herrero, M., Mendiola, J. A., Cifuentes, A., & Ibáñez, E. (2010). Supercritical fluid extraction: Recent advances and applications. In *Journal of Chromatography A*, 1217(16),2495–2511. https://doi.org/10.1016/j.chroma.2009.12.019

Hillen, C. E. (2016). Sensory and Quality Attributes of Deodorized Pea Flour Used in Gluten-Free Food Products. https://library.ndsu.edu/ir/handle/10365/28135

Hoover, R., Hughes, T., Chung, H. J., & Liu, Q. (2010). Composition, molecular structure, properties, and modification of pulse starches: A review. In *Food Research International* 43(2), 399–413. https://doi.org/10.1016/j.foodres.2009.09.001

Hoover, R., & Senanayake, S. P. J. N. (1996). Composition and physicochemical properties of oat starches. In *Food Research International*, 29(1), 15-26.

Hrncic, M. K., Cör, D., Verboten, M. T., & Knez, Z. (2018). Application of supercritical and subcritical fluids in food processing. In *Food Quality and Safety*, 2(2),59–67. Oxford University Press. https://doi.org/10.1093/fqsafe/fyy008

Ivanovic, J., Milovanovic, S., & Zizovic, I. (2016). Utilization of supercritical CO2 as a processing aid in setting functionality of starch-based materials. In *Starch/Staerke*, 68(9–10), 821–833. Wiley-VCH Verlag. https://doi.org/10.1002/star.201500194

Jacobs, D. R., & Gallaher, D. D. (2004). Whole grain intake and cardiovascular disease: a review. *Current Atherosclerosis Reports*, *6*(6), 415–423. https://doi.org/10.1007/S11883-004-0081-Y

Jitngarmkusol, S., Hongsuwankul, J., & Tananuwong, K. (2008). Chemical compositions, functional properties, and microstructure of defatted macadamia flours. *Food Chemistry*, *110*(1), 23–30. https://doi.org/10.1016/j.foodchem.2008.01.050

Jodhner, J. C. F., & Meireles, M. A. de A. (2016). Construction of a supercritical fluid extraction (SFE) equipment: Validation using annatto and fennel and extract analysis by thin layer chromatography coupled to image. *Food Science and Technology (Brazil)*, *36*(2), 210–247. https://doi.org/10.1590/1678-457X.0027

Johnson, E. L., Feldman, H., Butts, A., Billy, C. D. R., Dugan, J., Leal, S., Rhinehart, A. S., Shubrook, J. H., Trujillo, J., Neumiller, J. J., Cannon, C., de Boer, I., Crandall, J., D'Alessio, D., de Groot, M., Fradkin, J., Kreider, K., Maahs, D., Maruthur, N., … Uelmen, S. (2019). Standards of medical care in diabetes—2019 abridged for primary care providers. *Clinical Diabetes*, *37*(1), 11–34. https://doi.org/10.2337/cd18-0105

Joshi, P. K., & Rao, P. P. (2017). Global pulses scenario: status and outlook. *Annals of the New York Academy of Sciences*, *1392*(1), 6–17. https://doi.org/10.1111/nyas.13298

Kajihausa, *, Fasasi, R. A., & Atolagbe. (2014). Effect of Different Soaking Time and Boiling on the Proximate Composition and Functional Properties of Sprouted Sesame Seed Flour. In *Nigerian Food Journal Official Journal of Nigerian Institute of Food Science and Technology www.nifst.org NIFOJ* (Vol. 32, Issue 2). www.nifst.org

Kang, S. W., Rahman, M. S., Kim, A. N., Lee, K. Y., Park, C. Y., Kerr, W. L., & Choi, S. G. (2017). Comparative study of the quality characteristics of defatted soy flour treated by supercritical carbon dioxide and organic solvent. *Journal of Food Science and Technology*, *54*(8), 2485–2493. https://doi.org/10.1007/s13197-017-2691-8

Karim, A. A., Norziah, M. H., & Seow, C. C. (2000). Methods for the study of starch retrogradation. *Food Chemistry*, *71*(1), 9–36. https://doi.org/10.1016/S0308-8146(00)00130-8

Karmakar, R., Ban, D., Karmakar, *, & Ghosh, D. K. (2014). Comparative study of native and modified starches isolated from conventional and nonconventional sources Interaction of nanostructures with bio-molecules, and cells View project sensors View project. In *Article in International Food Research Journal*, 21(2).

Kim, M. J., Oh, S. G., & Chung, H. J. (2017). Impact of heat-moisture treatment applied to brown rice flour on the quality and digestibility characteristics of Korean rice cake. *Food Science and Biotechnology*, *26*(6), 1579–1586. https://doi.org/10.1007/s10068-017-0151-x

Kirkegaard, J., Christen, O., Krupinsky, J., & Layzell, D. (2008). Break crop benefits in temperate wheat production. In *Field Crops Research*, 107, Issue 3, pp. 185–195). https://doi.org/10.1016/j.fcr.2008.02.010

Knez, Markočič, E., Leitgeb, M., Primožič, M., Knez Hrnčič, M., & Škerget, M. (2014). Industrial applications of supercritical fluids: A review. *Energy*, *77*, 235–243. https://doi.org/10.1016/j.energy.2014.07.044

Knez, Ž., Škerget, M., & KnezHrnčič, M. (2010). Principles of supercritical fluid extraction and applications in the food, beverage and nutraceutical industries. *Separation, Extraction and Concentration Processes in the Food, Beverage and Nutraceutical Industries*, 3–38. https://doi.org/10.1533/9780857090751.1.3

Lam, A. C. Y., Can Karaca, A., Tyler, R. T., & Nickerson, M. T. (2018). Pea protein isolates: Structure, extraction, and functionality. In *Food Reviews International*, 34(2),126–147. Taylor and Francis Inc. https://doi.org/10.1080/87559129.2016.1242135

Lee, H. C., Htoon, A. K., & Paterson, J. L. (2007). Alkaline extraction of starch from Australian lentil cultivars Matilda and Digger optimised for starch yield and starch and protein quality. *Food Chemistry*, *102*(3), 551–559. https://doi.org/10.1016/j.foodchem.2006.03.042

Li, J. H., & Vasanthan, T. (2003). Hypochlorite oxidation of field pea starch and its suitability for noodle making using an extrusion cooker. *Food Research International*, *36*(4), 381–386. https://doi.org/10.1016/S0963-9969(02)00230-2

Li, L., Yuan, T. Z., Setia, R., Raja, R. B., Zhang, B., & Ai, Y. (2019). Characteristics of pea, lentil and faba bean starches isolated from air-classified flours in comparison with commercial starches. *Food Chemistry*, *276*, 599–607. https://doi.org/10.1016/j.foodchem.2018.10.064

Linares-García, L., Repo-Carrasco-Valencia, R., Paulet, P. G., & Schoenlechner, R. (2019). Development of gluten-free and egg-free pasta based on quinoa (Chenopdium quinoa Willd) with addition of lupine flour, vegetable proteins and the oxidizing enzyme POx. *European Food Research and Technology*, *245*(10), 2147–2156. https://doi.org/10.1007/s00217-019-03320-1

Liu, H., Wang, L., Cao, R., Fan, H., & Wang, M. (2016). In vitro digestibility and changes in physicochemical and structural properties of common buckwheat starch affected by high hydrostatic pressure. *Carbohydrate Polymers*, *144*, 1–8. https://doi.org/10.1016/j.carbpol.2016.02.028

Lu, Z. H., Donner, E., & Liu, Q. (2018). Effect of roasted pea flour/starch and encapsulated pea starch incorporation on the in vitro starch digestibility of pea breads. *Food Chemistry*, *245*, 71–78. https://doi.org/10.1016/j.foodchem.2017.10.037

Lu, Z. X., He, J. F., Zhang, Y. C., & Bing, D. J. (2020). Composition, physicochemical properties of pea protein and its application in functional foods. In *Critical Reviews in Food Science and Nutrition* (Vol. 60, Issue 15, pp. 2593–2605). Taylor and Francis Inc. https://doi.org/10.1080/10408398.2019.1651248

Lunde, M. S. H., Hjellset, V. T., Holmboe-Ottesen, G., & Høstmark, A. T. (2011). Variations in postprandial blood glucose responses and satiety after intake of three types of bread. *Journal of Nutrition and Metabolism*, *2011*. https://doi.org/10.1155/2011/437587

Marinangeli, C. P. F., Kassis, A. N., & Jones, P. J. H. (2009). Glycemic responses and sensory characteristics of whole yellow pea flour added to novel functional foods. *Journal of Food Science*, *74*(9). https://doi.org/10.1111/j.1750-3841.2009.01347.x

Mariotti, M., Zardi, M., Lucisano, M., & Pagani, M. A. (2005). Influence of the heating rate on the pasting properties of various flours. *Starch/Staerke*, *57*(11), 564–572. https://doi.org/10.1002/star.200500425

Marta, H., & Tensiska, T. (2017). Functional and Amylographic Properties of Physically Modified Sweet Potato Starch. *KnE Life Sciences*, *2*(6), 689. https://doi.org/10.18502/kls.v2i6.1091

McKee, L. H., & Latner, T. A. (2000). Underutilized sources of dietary fiber: A review. *Plant Foods for Human Nutrition*, *55*(4), 285–304. https://doi.org/10.1023/A:100814431098

Mizelman, E., Chilibeck, P. D., Hanifi, A., Kaviani, M., Brenna, E., & Zello, G. A. (2020). A low-glycemic index, high-fiber, pulse-based diet improves lipid profile, but does not affect performance in soccer players. *Nutrients*, *12*(5). https://doi.org/10.3390/nu12051324

Mollard, R. C., Zykus, A., Luhovyy, B. L., Nunez, M. F., Wong, C. L., & Anderson, G. H. (2012). The acute effects of a pulse-containing meal on glycaemic responses and measures of satiety and satiation within and at a later meal. *British Journal of Nutrition*, *108*(3), 509–517. https://doi.org/10.1017/S0007114511005836

Morales-Polanco, E., Campos-Vega, R., Gaytán-Martínez, M., Enriquez, L. G., & Loarca-Piña, G. (2017). Functional and textural properties of a dehulled oat (Avena sativa L) and pea (Pisum sativum) protein isolate cracker. *LWT*, *86*, 418–423. https://doi.org/10.1016/j.lwt.2017.08.015

Muljana, H., Picchioni, F., Heeres, H. J., & Janssen, L. P. B. M. (2009). Supercritical carbon dioxide (scCO2) induced gelatinization of potato starch. *Carbohydrate Polymers*, *78*(3), 511–519. https://doi.org/10.1016/j.carbpol.2009.05.005

Mwizerwa, H., Abong, G. O., Okoth, M. W., Ongol, M. P., Onyango, C., & Thavarajah, P. (2017). Effect of resistant cassava starch on quality parameters and sensory attributes of yoghurt. *Current Research in Nutrition and Food Science*, *5*(3), 353–367. https://doi.org/10.12944/CRNFSJ.5.3.21

NDSU. (2021). *Field Pea Production Reviewed by Gregory Endres Extension Cropping Systems Specialist Hans Kandel Extension Agronomist/Broadleaf Crops*. www.ag.ndsu.edu

Noyori, R. (1999). Supercritical Fluids: Introduction. *Chemical Reviews*, *99*(2), 353–354. https://doi.org/10.1021/cr980085a

Ocheme, O. B., Adedeji, O. E., Chinma, C. E., Yakubu, C. M., & Ajibo, U. H. (2018). Proximate composition, functional, and pasting properties of wheat and groundnut protein concentrate flour blends. In *Food Science and Nutrition*, 6(5),1173–1178. Wiley-Blackwell. https://doi.org/10.1002/fsn3.670

Ohizua, E. R., Adeola, A. A., Idowu, M. A., Sobukola, O. P., Afolabi, T. A., Ishola, R. O., Ayansina, S. O., Oyekale, T. O., & Falomo, A. (2017). Nutrient composition, functional, and pasting properties of unripe cooking banana, pigeon pea, and sweetpotato flour blends. *Food Science and Nutrition*, *5*(3), 750–762. https://doi.org/10.1002/fsn3.455

Oppong Siaw, M., Wang, Y. J., McClung, A. M., & Mauromoustakos, A. (2021). Effect of protein denaturation and lipid removal on rice physicochemical properties. *LWT*, *150*. https://doi.org/10.1016/j.lwt.2021.112015

Ottenhof, M. A., & Farhat, I. A. (2004). Starch retrogradation. *Biotechnology and Genetic Engineering Reviews*, *21*(1), 215–228. https://doi.org/10.1080/02648725.2004.10648056

Palacios-Fonseca, A. J., Castro-Rosas, J., Gómez-Aldapa, C. A., Tovar-Benítez, T., Millán-Malo, B. M., Del Real, A., & Rodríguez-García, M. E. (2013). Effect of the alkaline and acid treatments on the physicochemical properties of corn starch. *CYTA - Journal of Food*, *11*(SUPPL.1), 67–74. https://doi.org/10.1080/19476337.2012.761651

Patil, S. S., Brennan, M. A., Mason, S. L., & Brennan, C. S. (2016). The effects of fortification of legumes and extrusion on the protein digestibility of wheat based snack. *Foods*, *5*(2), 1–8. https://doi.org/10.3390/foods5020026

Pietrasik, Z., Sigvaldson, M., Soladoye, O. P., & Gaudette, N. J. (2020). Utilization of pea starch and fibre fractions for replacement of wheat crumb in beef burgers. *Meat Science*, *161*. https://doi.org/10.1016/j.meatsci.2019.107974

Pietrasik, Z., & Soladoye, O. P. (2021). Use of native pea starches as an alternative to modified corn starch in low-fat bologna. *Meat Science*, *171*. https://doi.org/10.1016/j.meatsci.2020.108283

Puchongkavarin, H., Varavinit, S., & Bergthaller, W. (2005). Comparative study of pilot scale rice starch production by an alkaline and an enzymatic process. *Starch/Staerke*, *57*(3–4), 134–144. https://doi.org/10.1002/star.200400279

Rakshit, S. K. (2014). Physicochemical properties of heat moisture treated sweet potato starches of selected Indonesian varieties. In *Article in International Food Research Journal*. https://www.researchgate.net/publication/285996149

Rasskazova, I., & Kirse-Ozolina, A. (2020). Field pea Pisum sativum L. as a perspective ingredient for vegan foods: a review. *Research for Rural Development*, *35*, 125–131. https://doi.org/10.22616/rrd.26.2020.019

Ratnayake, W. S., Hoover, R., Shahidi, F., Perera, C., & Jane, J. (2001). *Composition, molecular structure, and physicochemical properties of starches from four field pea (Pisum sativum L.) cultivars*. www.elsevier.com/locate/foodchem

Ratnayake, W. S., & Warkentin, T. (2002). *Pea Starch: Composition, Structure and Properties - A Review*. https://doi.org/10.1002/1521-379X(200206)54:63.0.CO;2-R

Rebello, C. J., Greenway, F. L., & Finley, J. W. (2014). Whole grains and pulses: A comparison of the nutritional and health benefits. In *Journal of Agricultural and Food Chemistry*, 62(29),7029–7049. American Chemical Society. https://doi.org/10.1021/jf500932z

Ren, Y., Setia, R., Warkentin, T. D., & Ai, Y. (2021). Functionality and starch digestibility of wrinkled and round pea flours of two different particle sizes. *Food Chemistry*, *336*. https://doi.org/10.1016/j.foodchem.2020.127711

Rennie, M. J. (2005). Body maintenance and repair: How food and exercise keep the musculoskeletal system in good shape. *Experimental Physiology*, *90*(4), 427–436. https://doi.org/10.1113/expphysiol.2005.029983

RincÓn, J., De Lucas, A., & Gracia, I. (2000). Isolation of rock rose essential oil using supercritical CO2 extraction. *Separation Science and Technology*, *35*(16), 2745–2763. https://doi.org/10.1081/SS-100102366

Roland, W. S. U., Pouvreau, L., Curran, J., Van De Velde, F., & De Kok, P. M. T. (2017). Flavor aspects of pulse ingredients. In *Cereal Chemistry* (Vol. 94, Issue 1, pp. 58–65). American Association of Cereal Chemists. https://doi.org/10.1094/CCHEM-06-16-0161-FI

Rong, L., Shen, M., Wen, H., Xiao, W., Li, J., & Xie, J. (2022). Effects of xanthan, guar and Mesona chinensis Benth gums on the pasting, rheological, texture properties and microstructure of pea starch gels. *Food Hydrocolloids*, *125*. https://doi.org/10.1016/j.foodhyd.2021.107391

Saberi, B., Thakur, R., Vuong, Q. V., Chockchaisawasdee, S., Golding, J. B., Scarlett, C. J., & Stathopoulos, C. E. (2016). Optimization of physical and optical properties of biodegradable edible films based on pea starch and guar gum. *Industrial Crops and Products*, *86*, 342–352. https://doi.org/10.1016/j.indcrop.2016.04.015

Saldanha do Carmo, C., Silventoinen, P., Nordgård, C. T., Poudroux, C., Dessev, T., Zobel, H., Holtekjølen, A. K., Draget, K. I., Holopainen-Mantila, U., Knutsen, S. H., & Sahlstrøm, S. (2020). Is dehulling of peas and faba beans necessary prior to dry fractionation for the production of protein- and starch-rich fractions? Impact on physical properties, chemical composition and techno-functional properties. *Journal of Food Engineering*, *278*. https://doi.org/10.1016/j.jfoodeng.2020.109937

Saleh, A., Mohamed, A. A., Alamri, M. S., Hussain, S., Qasem, A. A., & Ibraheem, M. A. (2020). Effect of different starches on the rheological, sensory and storage attributes of non-fat set yogurt. *Foods*, *9*(1). https://doi.org/10.3390/foods9010061

Sasaki, T., & Matsuki, J. (1998). Effect of wheat starch structure on swelling power. *Cereal Chemistry*, *75*(4), 525–529. https://doi.org/10.1094/CCHEM.1998.75.4.525

Schoch, T., & Maywald, E. (1968). *Preparation and Properties of Various Legume Starches*.

Seewi, G., Gnauck, G., Stute, R., Chantelau, E., Gnauck, · G, & Stute, · R. (1999). Effects on parameters of glucose homeostasis in healthy humans from ingestion of leguminous versus maize starches. In *Eur J Nutr* (Vol. 38).

Sessa, D. J., Eller, F. J., Palmquist, D. E., & Lawton, J. W. (2003). Improved methods for decolorizing corn zein. *Industrial Crops and Products*, *18*(1), 55–65. https://doi.org/10.1016/S0926-6690(03)00033-5

Shahwar, D., Ansari, M. Y. K., & Choudhary, S. (2018). Evaluation of Genotoxic Potential of Heavy Metal in a Proteinaceous Crop (Lens culinaris Medik) in Aspect to Cyto-morphological Parameters. *Journal of Biological Sciences*, *18*(5), 208–215. https://doi.org/10.3923/jbs.2018.208.215

Shand, P. J., Ya, H., Pietrasik, Z., & Wanasundara, P. K. J. P. D. (2007). Physicochemical and textural properties of heat-induced pea protein isolate gels. *Food Chemistry*, *102*(4), 1119–1130. https://doi.org/10.1016/j.foodchem.2006.06.060

Simsek, S., Tulbek, M. C., Yao, Y., & Schatz, B. (2009a). Starch characteristics of dry peas (Pisum sativum L.) grown in the USA. *Food Chemistry*, *115*(3), 832–838. https://doi.org/10.1016/j.foodchem.2008.12.093

Simsek, S., Tulbek, M. C., Yao, Y., & Schatz, B. (2009b). Starch characteristics of dry peas (Pisum sativum L.) grown in the USA. *Food Chemistry*, *115*(3), 832–838. https://doi.org/10.1016/j.foodchem.2008.12.093

Singh, B., Singh, J. P., Shevkani, K., Singh, N., & Kaur, A. (2017). Bioactive constituents in pulses and their health benefits. In *Journal of Food Science and Technology* (Vol. 54, Issue 4, pp. 858–870). https://doi.org/10.1007/s13197-016-2391-9

Singh, M., Manickavasagan, A., Shobana, S., & Mohan, V. (2021). Glycemic index of pulses and pulse-based products: a review. In *Critical Reviews in Food Science and Nutrition*, 61(9),1567–1588. https://doi.org/10.1080/10408398.2020.1762162

Singh, N. (2020). Functional and physicochemical properties of pulse starch. In *Pulse Foods: Processing, Quality and Nutraceutical Applications*. https://doi.org/10.1016/B978-0-12-818184-3.00005-2

Singh, N., Sandhu, K. S., & Kaur, M. (2004). Characterization of starches separated from Indian chickpea (Cicer arietinum L.) cultivars. *Journal of Food Engineering*, *63*(4), 441–449. https://doi.org/10.1016/j.jfoodeng.2003.09.003

Sippel, K. H., & Quiocho, F. A. (2015). Ion-dipole interactions and their functions in proteins. In *Protein Science*, 24(7),1040–1046. https://doi.org/10.1002/pro.2685

Smith, C. J., & Chalk, P. M. (2020). Grain legumes in crop rotations under low and variable rainfall: are observed short-term N benefits sustainable? In *Plant and Soil*, 453(1–2),271–279. https://doi.org/10.1007/s11104-020-04578-1

Solaesa, Á. G., Villanueva, M., Beltrán, S., & Ronda, F. (2019). Characterization of Quinoa Defatted by Supercritical Carbon Dioxide. Starch Enzymatic Susceptibility and Structural, Pasting and Thermal Properties. *Food and Bioprocess Technology*, *12*(9), 1593–1602. https://doi.org/10.1007/s11947-019-02329-4

Stoughton-Ens, M. D., Hatcher, D. W., Wang, N., & Warkentin, T. D. (2010). Influence of genotype and environment on the dietary fiber content of field pea (Pisum sativum L.) grown in Canada. *Food Research International*, *43*(2), 547–552. https://doi.org/10.1016/j.foodres.2009.07.011

Sun, Q., Chu, L., Xiong, L., & Si, F. (2015a). Effects of different isolation methods on the physicochemical properties of pea starch and textural properties of vermicelli. *Journal of Food Science and Technology*, *52*(1), 327–334. https://doi.org/10.1007/s13197-013-0980-4

Sun, Q., Chu, L., Xiong, L., & Si, F. (2015b). Effects of different isolation methods on the physicochemical properties of pea starch and textural properties of vermicelli. *Journal of Food Science and Technology*, *52*(1), 327–334. https://doi.org/10.1007/s13197-013-0980-4

Sun, Q., Li, P., Li, Y., Ji, N., Dai, L., Xiong, L., & Sun, Q. (2020). Rapid production of corn starch gels with high mechanical properties through alcohol soaking. *International Journal of Biological Macromolecules*, *163*, 1557–1564. https://doi.org/10.1016/j.ijbiomac.2020.08.042

Sun, Q., & Xiong, C. S. L. (2014). Functional and pasting properties of pea starch and peanut protein isolate blends. *Carbohydrate Polymers*, *101*(1), 1134–1139. https://doi.org/10.1016/j.carbpol.2013.10.064

Thys, R. C. S., Westfahl, H., Noreña, C. P. Z., Marczak, L. D. F., Silveira, N. P., & Cardoso, M. B. (2008). Effect of the alkaline treatment on the ultrastructure of C-type starch granules. *Biomacromolecules*, *9*(7), 1894–1901. https://doi.org/10.1021/bm800143w

Vander Pol, M., Hristov, A. N., Zaman, S., & Delano, N. (2008). Peas can replace soybean meal and corn grain in dairy cow diets. *Journal of Dairy Science*, *91*(2), 698–703. https://doi.org/10.3168/jds.2007-0543

Vanier, N. L., El Halal, S. L. M., Dias, A. R. G., & da Rosa Zavareze, E. (2017). Molecular structure, functionality and applications of oxidized starches: A review. In *Food Chemistry* (Vol. 221, pp. 1546–1559). Elsevier Ltd. https://doi.org/10.1016/j.foodchem.2016.10.138

Vatansever, S., & Hall, C. (2020). Flavor modification of yellow pea flour using supercritical carbon dioxide + ethanol extraction and response surface methodology. *Journal of Supercritical Fluids*, *156*. https://doi.org/10.1016/j.supflu.2019.104659

Vatansever, S., Rao, J., & Hall, C. (2020). Effects of ethanol modified supercritical carbon dioxide extraction and particle size on the physical, chemical, and functional properties of yellow pea flour. *Cereal Chemistry*, *97*(6), 1133–1147. https://doi.org/10.1002/cche.10334

Vatansever, S., Tulbek, M. C., & Riaz, M. N. (2020). Low- and High-Moisture Extrusion of Pulse Proteins as Plant-Based Meat Ingredients: A Review. *Cereal Foods World*, *65*(4). https://doi.org/10.1094/cfw-65-4-0038

Vatansever, S., Whitney, K., Ohm, J. B., Simsek, S., & Hall, C. (2021). Physicochemical and multi-scale structural alterations of pea starch induced by supercritical carbon dioxide + ethanol extraction. *Food Chemistry*, *344*. https://doi.org/10.1016/j.foodchem.2020.128699

Voormeij, D., Simandl, G. J., & Voormeij, D. A. (2003). *Geological and Mineral CO2 Sequestration Options: A Technical Review Ongoing research on Specialty Metal deposits View project Geological Sequestration of CO2 View project Geological and Mineral CO 2 Sequestration Options: A Technical Review*. https://www.researchgate.net/publication/246314679

Wang, K., & Arntfield, S. D. (2016). Modification of interactions between selected volatile flavour compounds and salt-extracted pea protein isolates using chemical and enzymatic approaches. *Food Hydrocolloids*, *61*, 567–577. https://doi.org/10.1016/j.foodhyd.2016.05.040

Wang, N., & Daun, J. K. (2006). Effects of variety and crude protein content on nutrients and anti-nutrients in lentils (Lens culinaris). *Food Chemistry*, *95*(3), 493–502. https://doi.org/10.1016/j.foodchem.2005.02.001

Wang, N., Hatcher, D. W., Warkentin, T. D., & Toews, R. (2010). Effect of cultivar and environment on physicochemical and cooking characteristics of field pea (Pisum sativum). *Food Chemistry*, *118*(1), 109–115. https://doi.org/10.1016/j.foodchem.2009.04.082

Wang, N., Maximiuk, L., Fenn, D., Nickerson, M. T., & Hou, A. (2020). Development of a method for determining oil absorption capacity in pulse flours and protein materials. *Cereal Chemistry*, *97*(6), 1111–1117. https://doi.org/10.1002/cche.10339

Wang, N., Maximiuk, L., & Toews, R. (2012). Pea starch noodles: Effect of processing variables on characteristics and optimisation of twin-screw extrusion process. *Food Chemistry*, *133*(3), 742–753. https://doi.org/10.1016/j.foodchem.2012.01.087

Wang, S., & Copeland, L. (2012). Effect of alkali treatment on structure and function of pea starch granules. *Food Chemistry*, *135*(3), 1635–1642. https://doi.org/10.1016/j.foodchem.2012.06.003

Wang, Y., Guldiken, B., Tulbek, M., House, J. D., & Nickerson, M. (2020). Impact of alcohol washing on the flavour profiles, functionality and protein quality of air classified pea protein enriched flour. *Food Research International*, *132*. https://doi.org/10.1016/j.foodres.2020.109085

Wani, I. A., Sogi, D. S., Hamdani, A. M., Gani, A., Bhat, N. A., & Shah, A. (2016). Isolation, composition, and physicochemical properties of starch from legumes: A review. In *Starch/Staerke* (Vol. 68, Issues 9–10, pp. 834–845). https://doi.org/10.1002/star.201600007

Wei, P. C., May, C. Y., Ngan, M. A., & Hock, C. C. (2005). Supercritical Fluid Extraction of Palm Carotenoids. *American Journal of Environmental Sciences*, *1*(4), 264–269. https://doi.org/10.3844/ajessp.2005.264.269

Wu, F., Li, J., Yang, N., Chen, Y., Jin, Y., & Xu, X. (2018). The Roles of Starch Structures in the Pasting Properties of Wheat Starch with Different Degrees of Damage. *Starch/Staerke*, *70*(5–6). https://doi.org/10.1002/star.201700190

Yoshida, H., Tomiyama, Y., Saiki, M., & Mizushina, Y. (2007). Tocopherol content and fatty acid distribution of peas (Pisum sativum L.). *JAOCS, Journal of the American Oil Chemists' Society*, *84*(11), 1031–1038. https://doi.org/10.1007/s11746-007-1134-5

Zhao, J., Liu, X., Bai, X., & Wang, F. (2019a). Production of biscuits by substitution with different ratios of yellow pea flour. *Grain & Oil Science and Technology*, *2*(4), 91–96. https://doi.org/10.1016/j.gaost.2019.09.004

Zhao, J., Liu, X., Bai, X., & Wang, F. (2019b). Production of biscuits by substitution with different ratios of yellow pea flour. *Grain & Oil Science and Technology*, *2*(4), 91–96. https://doi.org/10.1016/j.gaost.2019.09.004

Zhao, Q., Zhao, M., Yang, B., & Cui, C. (2009). Effect of xanthan gum on the physical properties and textural characteristics of whipped cream. *Food Chemistry*, *116*(3), 624–628. https://doi.org/10.1016/j.foodchem.2009.02.079

Zhou, D., Ma, Z., Yin, X., Hu, X., & Boye, J. I. (2019). Structural characteristics and physicochemical properties of field pea starch modified by physical, enzymatic, and acid

treatments. *Food Hydrocolloids*, *93*, 386–394. https://doi.org/10.1016/j.foodhyd.2019.02.048

Zia-ud-Din, Xiong, H., & Fei, P. (2017). Physical and chemical modification of starches: A review. In *Critical Reviews in Food Science and Nutrition* 57(12), 2691–2705. https://doi.org/10.1080/10408398.2015.1087379

APPENDIX

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	19.42981	1	19.42981	25.18232	0.002408	5.987378
Within Groups	4.629392	6	0.771565			
Total	24.0592	7				

Table 1. ANOVA of moisture content comparison of starch sample before and after SC CO_2 + EtOH treatment.

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	4330625	1	4330625	27.1754	0.00199	5.987378
Within Groups	956149.5	6	159358.3			
Total	5286774	7				

Table 2. ANOVA of peak viscoisty comparison of starch sample before and after SC CO_2 + EtOH treatment.

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	0.008333	1	0.008333	6.386097	0.044857	5.987378
Within Groups	0.007829	6	0.001305			
Total	0.016162	7				

Table 3. ANOVA of fat content comparison of starch sample before and after SC CO_2 + EtOH treatment.

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	0.000506	1	0.000506	0.474988	0.516453	5.987378

Within Groups	0.006394	6	0.001066
Total	0.0069	7	

Table 4. ANOVA of ash content comparison of starch sample before and after SC CO_2 + EtOH treatment.

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	40.22437	1	40.22437	22.92407	0.003038	5.987378
Within Groups	10.52807	6	1.754679			
Total	50.75244	7				

Table 5. ANOVA of total starch content comparison of starch sample before and after SC CO_2 + EtOH treatment.

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	0.943791	3	0.314597	1.36817	0.372761	6.591382
Within Groups	0.91976	4	0.22994			
Total	1.863551	7				

Table 6. ANOVA of day 5 of yogurt synersis comparison of starch samples.

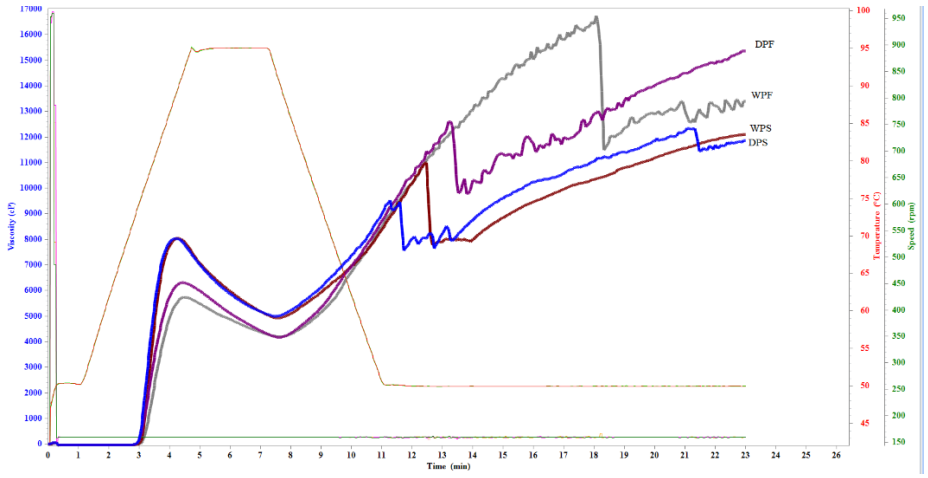

Figure 1. Rapid visco analyzer (RVA) plot of starch obtained from different starting materials using alkaline and dehulling. Abbreviation: WPS, whole pea seed; DPS, dehulled pea seed; WPF, whole pea fraction (100 mesh size); DPF, dehulled pea fraction (100 mesh size).

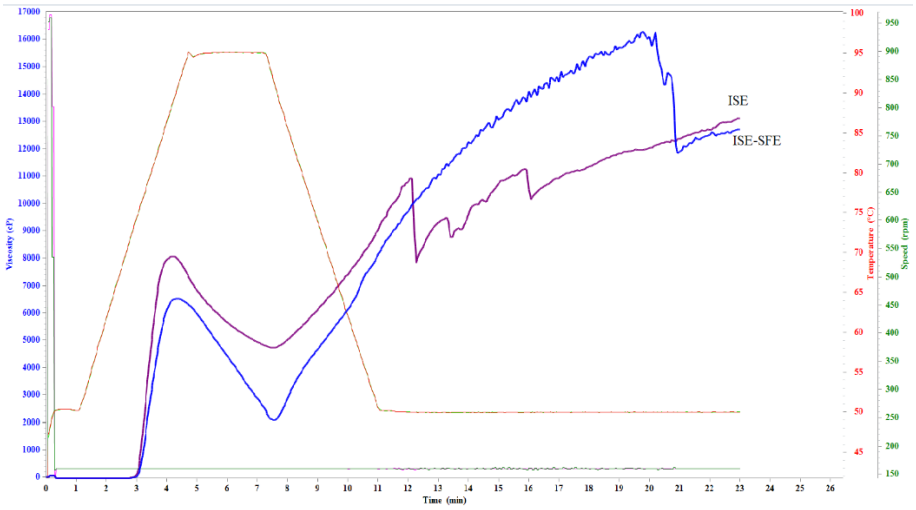

Figure 2. Rapid visco analyzer (RVA) plot of SC CO_2 + EtOH extracted starch. Abbreviation: ISE; industrial scale starch extraction, ISE-SFE; industrial scale starch extracted with supercritical fluid extraction.

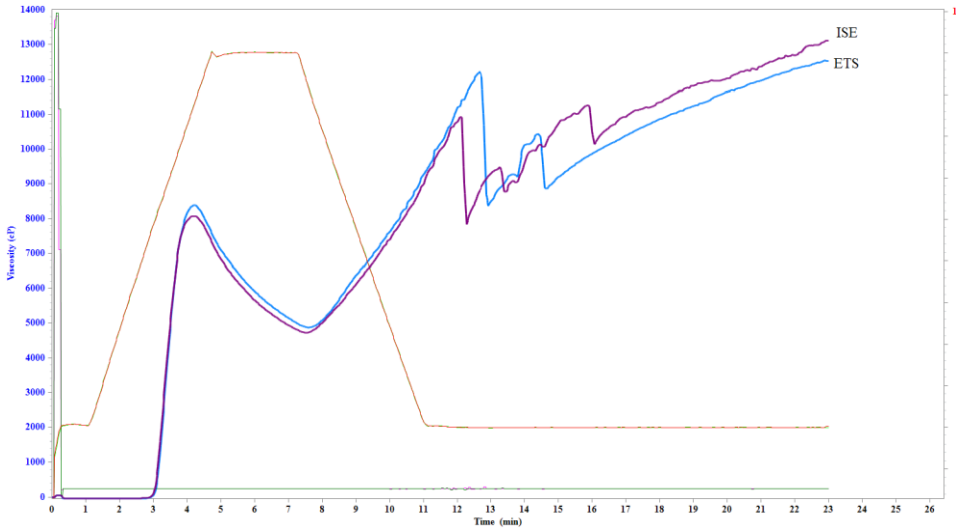

Figure 3. Rapid visco analyzer (RVA) plot of ethanol extracted starch. Abbreviation: ISE; industrial scale starch extraction, ETS; ethanol treated starch.

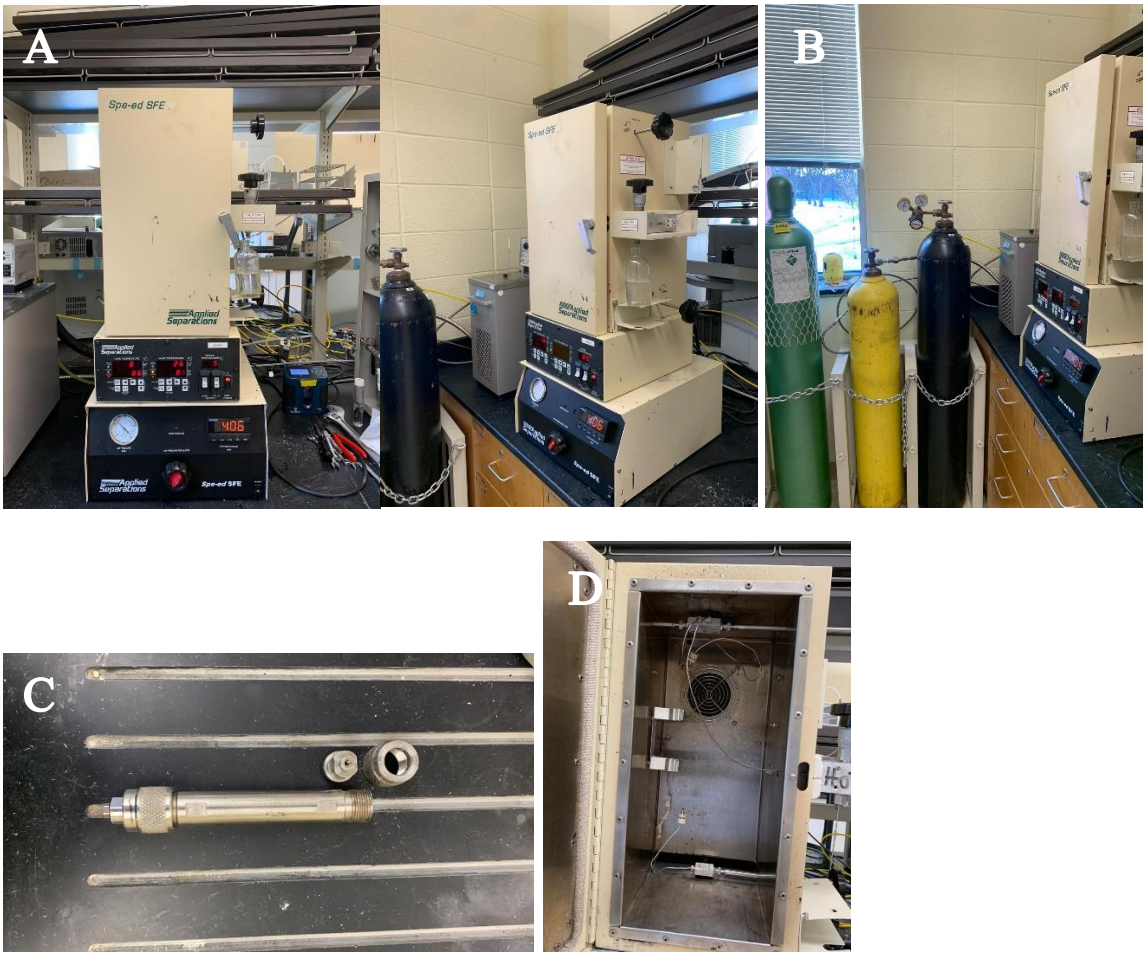

Figure 4. Supercritical fluid extractor (Spe-ed SFE, Applied Seperation) (A) used in starth extraction. The system includes CO_2 cylinder (B), a 25 ml stainless steel vessel (C) and an oven (D).

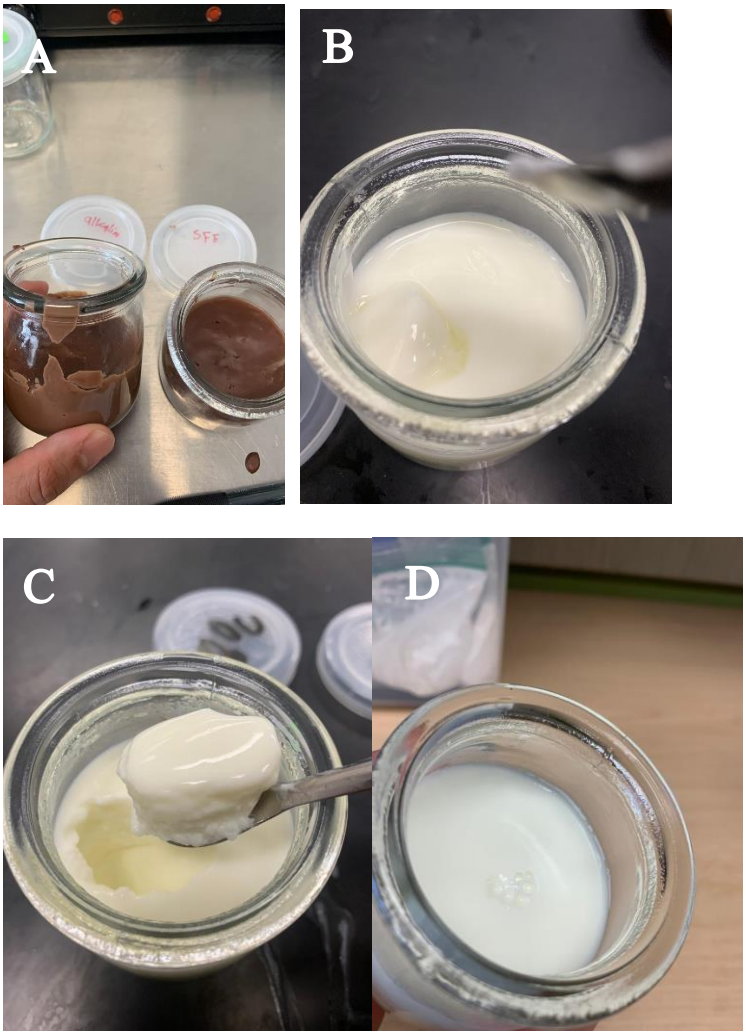

Figure 5. Starch-added pudding (A) and yogurt (B) product. Picture (C) shows the texture of the yogurt, and picture (D) shows the surface and syneresis of yogurt.