

Fortification of baked products with soy protein: studying the effects of baking with soy protein
and potential use in the development of hands-on food science education modules

by

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Abstract

Soy protein ingredients are of interest for the development of high-protein bakery foods because soy is a plant-based protein that contains adequate amounts of all essential amino acids. Fortification of bakery foods with soy protein results in changes to physical properties (water absorption, volume, color, and texture) and organoleptic properties (flavor, aftertaste). Research is ongoing to explore methods for improving physical and organoleptic properties of bakery foods with added soy protein to improve consumer acceptance. Enzymatic hydrolysis breaks down proteins into smaller molecular weight peptides with different properties than their large molecular weight counterparts. Soy protein hydrolysates prepared using papain and Flavourzyme have been found to have acceptable sensory properties, however, research is needed to determine the effects of soy protein hydrolysates in food systems. The objective of this study was to observe the effect of soy protein hydrolysates on physical properties of muffins. Seven muffin treatments were prepared in a 2×3 augmented factorial treatment structure. Three protein types (soy protein isolate (SPI), papain soy hydrolysate (PSH), Flavourzyme soy hydrolysate (FSH)) were tested at two levels (10%, 20%). Additionally, a control treatment was prepared with no added protein. Batter tests included specific gravity and pH. Muffin tests included crust and crumb color, height, weight, texture profile analysis, water activity, and moisture loss. Specific gravity ranged from 0.95-1.08 with SPI10, SPI20, and FSH20 having lower ($p < 0.05$) specific gravity values than C. Batter pH ranged from 6.53-7.72 and no significant differences were found. No significant difference was found for crust lightness (70.86-74.49), crumb lightness (72.73-74.77), weight (68.2-72.1 g), hardness (6.85-11.17 N), water activity (0.500-0.580) or moisture loss (11.1-12.7%). Muffin height ranged from 41.7-46.7mm and all treatment heights were similar to control, however, PSH10 and SPI20 were significantly different from each other with heights of 41.7mm and 46.7mm, respectively. Overall, hydrolysis of SPI resulted in batters more similar to control than SPI and did not reduce physical quality as compared to C.

Hands-on laboratory exercises play an integral role in STEM education to increase knowledge of core content and facilitate the development of soft skills such as communication skills and the ability to apply knowledge. As enrollment in online courses increases, instructors of STEM courses are challenged with ensuring hands-on learning experiences are available to online students. Additionally, hands-on activities present an opportunity to students to engage with relevant issues faced in the industry to better prepare them for employment upon graduation. The addition of soy protein to bakery foods is a current issue in the food industry that

can be used in the development of a hands-on at-home laboratory for online food science students. The objective of this study was to develop an at-home laboratory exercise for online students in an undergraduate food processing lab and compare student perception and performance to a similar on-campus course. Laboratory kits containing a control muffin mix with no soy, a 50% soy flour muffin mix, and 100% soy flour muffin mix were sent to students enrolled in an undergraduate food processing course. Students prepared muffin mixes and scored muffin physical and organoleptic properties using a muffin scorecard. Students completed post-laboratory questions and wrote a scientific abstract to communicate their results. After completion of the module, students responded to a module reflection survey with a 5-point Likert scale. Seventy-two percent of online students met or exceeded expectations on the abstract assignment. Ninety percent of online students agreed or strongly agreed the laboratory improved their ability to apply knowledge to practical issues in food processing and 97% agreed the laboratory exercise improved their scientific communication skills. Student performance and perception results indicate the module was effective in teaching course content and facilitating the development of soft skills.

Secondary education teachers use materials outside the curriculum to supplement existing material and to increase student engagement with novel lessons. Supplemental lessons come from a variety of sources, however, the creation of supplemental materials that build on curriculum standards would benefit teachers. The objective of this study was to develop and test a module with hands-on laboratory exercise for use in secondary food and agriculture classrooms. A module was designed and a preliminary test was conducted in a local high school classroom. After modifications were made, an independent teacher-based trial was conducted and a module evaluation survey was sent to teacher participants. While survey response was relatively low (n=3), the module was well-received by respondents.

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Chapter 1 - Introduction

Overview

Soybeans are an important, versatile agricultural commodity for the United States and are used in a variety of industries including food, feed, biofuel, and manufacturing. Soybeans are utilized for two main coproducts: soybean oil and soybean meal. After crushing the soybean seed and extracting the oil, the remaining material is soybean meal, with 32% carbohydrates and 44% protein (Perkins, 1995). While soybean meal is used primarily as a protein source in animal feed, soybean usage in human foods could be increased to meet the nutrition needs of a growing population without dramatic negative environmental impacts.

Protein-fortified baked goods are becoming more popular, with 40% of consumers indicating they are somewhat or very interested in high protein pancakes, waffles, and muffins in a survey from Glanbia Nutritionals (2020). High-protein ingredients made from soybean meal can be included in food formulations to increase protein. Soy protein ingredients, such as soy flour, concentrate, and isolate, are of interest for the development of protein-fortified baked goods because soy is a complete, plant-based protein, while many other plant-based proteins lack essential amino acids (Gorissen et al., 2018). However, high amounts of soy protein in baked goods can reduce consumer acceptance with negative qualities including a “beany” flavor, tough texture, and gummy mouthfeel (Majzoobi et al., 2014).

Research is ongoing to explore solutions that reduce negative physical and organoleptic attributes associated with adding soy protein to bakery foods. One solution is the use of defatted soy flour which has a protein content of approximately 50%. Removal of fat produces a mild-flavored soy flour by preventing the formation of off flavors due to lipid oxidation. Another solution for improving attributes of high protein bakery foods involves the use of enzyme-modified soy protein isolate. Soy protein isolate is produced from soy flour and contains 90% or more protein, therefore lower quantities are required in formulations to achieve a desired protein content (Annor et al., 2014). Further processing through enzymatic hydrolysis has been shown to reduce bitterness and change properties (solubility, emulsifying capacity, foaming capacity) of many plant protein hydrolysates in model systems (Wouters et al., 2016). More research is needed to determine the effect of hydrolysis of soy protein isolate on functional properties in bakery foods.

Problem-based learning is a type of active learning that invites students to investigate solutions to a field-specific problem, such as the addition of soy protein to bakery foods (Yew &

Goh, 2016). Problem-based laboratory activities can be an alternative to carefully choreographed experiments often used in lab courses. Experiments designed with a specific end result are not reflective of the research process and do not allow students the opportunity to practice interpreting and discussing unexpected results. In these “cookbook” style lab activities, students miss opportunities to develop critical thinking skills that are essential in science fields (Handelsman, 2004).

Educators at secondary and post-secondary levels could utilize modules featuring relevant problems in a field to improve students’ learning experience. Secondary teachers report seeking out material to supplement district or school provided curriculum to increase student engagement, provide differentiated instruction to learners at various levels, and expand upon existing curriculum content (Wang et al., 2021). Problem-based activities meet the aforementioned needs because they are engaging and dynamic by nature, so instructors can adjust the structure and guidance to meet the needs of diverse students. Course modules with problem-based activities are also beneficial for post-secondary classes. At the undergraduate level, research shows problem-based learning facilitates better long-term retention and skills acquisition than traditional methods (Strobel & Barneveld, 2009). As a result, problem-based learning modules could improve the career readiness of students, especially in STEM fields.

Objectives

Two objectives were identified for this study. The first objective was to design course modules utilizing hands-on laboratories on the use of soy protein in food products for three class types: high schools, undergraduate online, and undergraduate on-campus. The long-term goal is to make these modules available to secondary and undergraduate instructors for use in food and agriculture science courses. The second objective was to investigate the physical properties of muffins made with soy protein isolate and two soy protein hydrolysates.

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Chapter 2 - Review of Literature

Proteins

Proteins are biological macromolecules that participate in a variety of reactions in living organisms as enzymes, hormones, and structural components. Additionally, proteins aid in regulatory and transport biological processes. The structure of proteins can be described by four levels of increasing complexity that impact protein properties (Murray et al., 2017).

A unique sequence of amino acids is chained together to form polypeptides that make up the primary structure of a protein. Amino acids generally contain an amino group, a carboxyl group, and a side chain or R group arranged around a central carbon atom. Amide bonds known as peptide bonds link amino acids to form the backbone of a protein. Amino acid properties such as pH and polarity are determined by the R group.

The secondary structure refers to the arrangement of polypeptides in α -helices, β -sheets, and turns because of hydrogen bonds. The arrangement of the polypeptide chain is known as the topology of the secondary structure. Secondary structure topology is determined by bond angles due to hydrogen bonding between amino acids (Bischoff & Schlüter, 2012).

Side chains on the polypeptide backbone interact to form a three-dimensional protein shape, and some proteins form a quaternary structure when the side chains of two or more polypeptides interact (Sanvictores & Farci, 2022). Tertiary and quaternary structures contain protein folds, that are connected to protein function, and the folds can be grouped into domains of folding units (Bischoff & Schlüter, 2012).

Protein structure may be modified to change functional properties through physical, chemical, or enzymatic methods. Thermal treatment is a type of physical change that denatures proteins to irreversibly change the spatial arrangement of the protein bonds. Enzymatic modifications use enzymes to cleave peptide bonds of proteins, resulting in an increased number of low molecular weight peptides (Wouters et al., 2016). Chemical modifications by pH adjustment alters net charge of a protein. At lower pH values, proteins have a positive net charge and at higher pH values, proteins have a negative net charge (Zink et al., 2016).

The isolation of a protein from its source is performed at the isoelectric point of a protein, or the pH at which the net charge of the protein is zero. Proteins are precipitated at the isoelectric point because it is also the point of lowest protein solubility. After precipitation, the proteins can be further purified for use as protein concentrates or isolates (Tokmakov et al., 2021).

Protein structures vary based on whether the protein is sourced from animals or plants. In general, plant proteins have more β -sheets and less α -helices relative to animal proteins, and as a result, plant-based proteins are more likely to form aggregates that reduce digestibility and are more resistant to denaturation (Carbonaro et al., 2012). Making connections between structure and function of proteins is useful in developing modified proteins for both food and non-food applications (Rasheed et al., 2020).

Soybeans

Soybeans (*Glycine max*) are classified as an oilseed, which is a type of grain legume (Annor et al., 2014). The United States is the second leading producer of soybeans globally, producing 31% of the world's soybeans in 2020, second to Brazil. Of the 114 million metric tons of soybeans produced in the U.S., 54% are exported, making soybeans a valuable commodity in the U.S. Globally, approximately six percent of soybeans are utilized whole for human foods while seven percent are used whole in animal feed. The remaining 87% of soybeans are not consumed whole but are crushed to extract the valuable soybean oil. While a small fraction of soybean oil is used as biofuel, it is primarily used in food under the label vegetable oil (United Soybean Board, 2022).

The high-protein material remaining after oil extraction is known as soybean meal or soy cake. Globally, about 97% of soybean meal is used as a protein source in animal feed, while only three percent is used for food (United Soybean Board, 2022). In Asian countries a variety of soy foods are commonly used as a source of protein, including edamame, miso, natto, and tempeh (Li and Qi, 2022). China, the leading consumer of whole soybeans, consumed 14 million metric tons from 2020-2021.

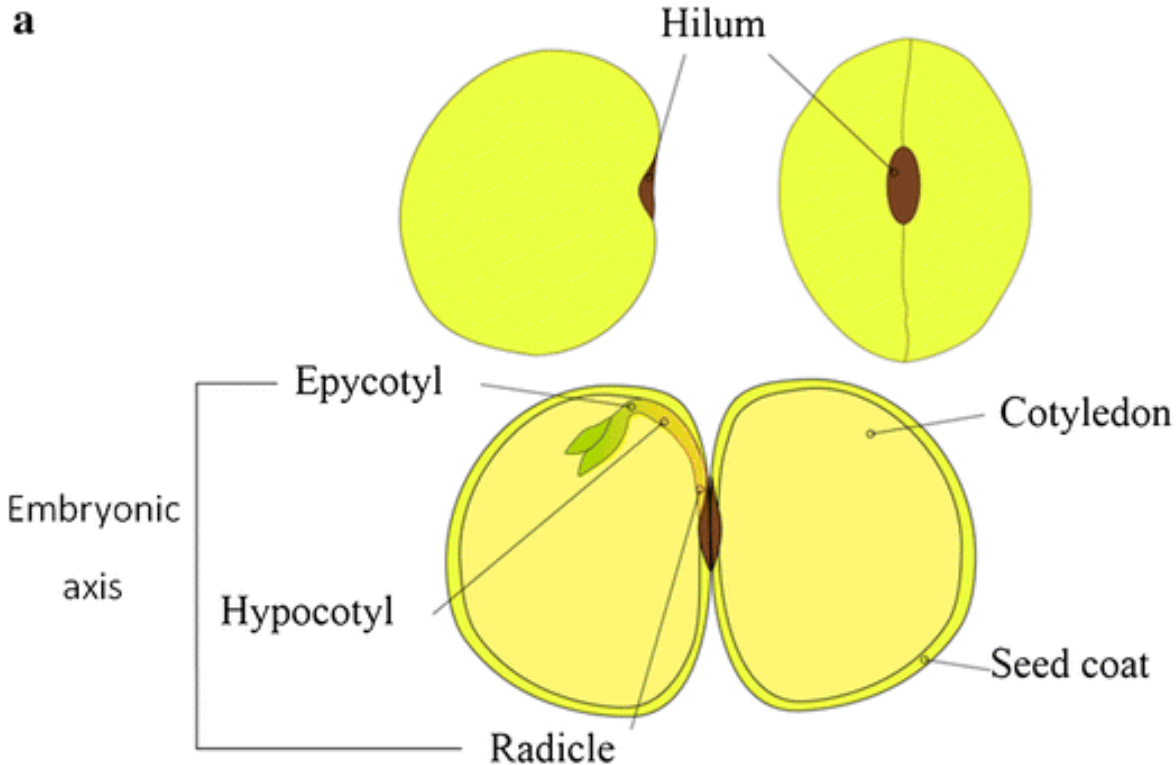
In the U.S., where soybean consumption is less common, only 125 thousand metric tons of whole soybeans were consumed in 2020-2021 (United Soybean Board, 2022). U.S. soy consumption is expected to increase as demand for plant-based foods increases. Retail sales of plant-based foods surpassed \$7 billion in 2021, with plant-based milk and plant-based meat representing the largest share of sales at \$2.6 billion and \$1.4 billion respectively (Plant Based Foods Association, 2021).

Structure

Soybeans are found in the pods of the soybean plant, typically with three beans per pod. Soybeans are seeds and contain all materials required to germinate and grow into a plant. Figure

2.1 is a diagram of the outside and cross-section of a soybean with labelled parts. On the outside of the bean, the seed coat protects the embryo until conditions are favorable for germination and contains about 86% carbohydrates (Medic et al., 2014).

Because the seed coat is impermeable to water, soybeans are considered hard seeds (Rolston, 1978). Research is ongoing to breed soybeans with more permeable seed coats because hard seed coats require more soaking time to absorb water, which can slow processing time (Ma et al., 2004).



Note. Reprinted from Medic et al. Current Knowledge in Soybean Composition. Journal of the American Oil Chemists' Society, 2014, 91(3), pages 363–384

Figure 2.1 Diagram of outside and cross-section of soybean seed

On outside of the soybean, a small brown spot known as the hilum is visible where the seed detaches from the bean pod. The embryonic axis is inside the seed and consists of the epicotyl, hypocotyl, and the radical (Sun and Yuan, 2022). The largest structure of the soybean is the cotyledon which accounts for 90% of the seed mass. The cotyledon is about 43% protein, 23% oil, and 29% carbohydrates in dry matter (Liu, 1997).

Composition

Soybeans differ from other legumes and cereals because soybeans have relatively high amounts of both protein and oil. While the exact ratios depend on growing conditions and soybean genetics, whole soybeans contain about 40% protein and 21% oil (Perkins, 1995). In comparison, cereal grains contain eight to 15% protein, while legumes generally have 20 to 30% protein. Legumes and cereals contain small amounts of oil from one to five percent, with the exception of peanuts (48%) (Liu, 1997). The large fractions of protein and oil position soybeans as a valuable crop with versatile end uses in food and feed applications. The remaining components are 34% carbohydrates and 5% ash on a dry weight basis (Perkins, 1995).

Protein

Seed storage proteins account for approximately 90% of soy protein, which provides nitrogen, carbon, and sulfur to the seedling as it grows. Two types of storage proteins are found in soybeans: β -conglycinin and glycinin, which are named according to the Latin genus of soybeans, *Glycine* (Krishnan and Coe, 2001).

β -conglycinin and glycinin differ in size, structure, and composition. β -conglycinin is a trimer with a molecular mass of 150-200 kDa while glycinin is a hexamer with a larger molecular mass of 300–380 kDa. The two proteins contribute different functional properties to soy protein. β -conglycinin forms a transparent, soft, elastic gel while glycinin forms a turbid, hard, sturdy gel. β -conglycinin has more emulsifying capacity while glycinin has more thermal stability (Fukushima, 2011).

Proteins are large macromolecules formed from 20-22 different amino acids linked by peptide bonds. The specific amino acids that make up a particular protein are important for determining the nutritional quality of the protein source. The human body utilizes amino acids to perform necessary biological processes including muscle and tissue growth and repair as well as production of hormones, digestive enzymes, and hemoglobin. Of the 22 amino acids, some can be synthesized by the body. Nine amino acids (histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan, and valine) are considered essential because they cannot be synthesized by the body and must be consumed through diet. Proteins that contain all nine essential amino acids are considered complete proteins (Lopez and Mohiuddin, 2021). One method for measuring protein quality is the Protein Digestibility Corrected Amino Acid Score (PDCAAS). To calculate the PDCAAS of a protein, the amount of the first limiting essential amino acid is divided by the amount of the same amino acid in an essential amino acid

reference pattern. The value obtained is then corrected to represent the true fecal digestibility of the protein (Schaafsma, 2000). Cow's milk and eggs have the highest possible PDCAAS of 1.00, while beef protein scores a 0.92. Plant-based proteins typically have lower scores. For example, the PDCAAS of wheat protein is 0.2; however, soy protein has a PDCAAS of 1.00 (Hughes et al., 2011). Amounts of each essential amino acid present in the protein source are summarized in Table 2.1, adapted from Gorissen et al. (2018). Both plant-based proteins (wheat and soy) are low in methionine.

Table 2.1—Amino acid content of common protein sources

Amino Acids	Amino acid content (g/100g)			
	Wheat	Soy	Egg	Milk
Threonine	1.8	2.3	2.0	3.5
Methionine	0.7	0.3	1.4	2.1
Phenylalanine	3.7	3.2	2.3	3.5
Histidine	1.4	1.5	0.9	1.9
Lysine	1.1	3.4	2.7	5.9
Valine	2.3	2.2	2.0	3.6
Isoleucine	2.0	1.9	1.6	2.9
Leucine	5.0	5.0	3.6	7.0

Note. adapted from Gorissen et al. (2018)

On nutrition facts panels of food products, the Food and Drug Administration (FDA) requires companies include grams of protein per serving. Companies may choose to also provide information about the percent daily value of protein per serving. In order for a company to claim

a food as a good source of protein, the food must contain at least 10% of the 50g Daily Reference Value (DRV). An “excellent source of protein” claim requires the product to have 20% of the daily protein consumption recommendation. When calculating the percent daily value of protein in a food product, the FDA requires companies use the PDCAAS to take into account amino acid composition. To calculate the correct percent daily value of protein per serving, grams of protein per serving is multiplied by the PDCAAS and divided by 50g (21CFR101.9).

Lipid

Soybeans are relatively higher in fats as compared to other cereals and legumes. Fatty acid composition analysis has found 5 main fatty acids in soybean oil: palmitic (C16:0), stearic (C18:0), oleic (C18:1), linoleic (C18:2) and linolenic (C18:3). Table 2.2 lists the percent of each fatty acid. Palmitic and stearic fatty acids are considered saturated fatty acids because there are no double bonds in the structure and every carbon is bonded to a hydrogen. Oleic, linoleic, and linolenic acids are unsaturated fatty acids. Polyunsaturated fatty acids, or fatty acids with more than one double bond, reduce the shelf life of an oil by causing rancidity (Panthee et al., 2006).

Table 2.2–Average fatty acid content in soybean oil

Fatty Acid	Amount (% crude oil)
Palmitic Acid	12
Stearic Acid	4
Oleic Acid	21
Linoleic Acid	55
Linoleic Acid	5

Carbohydrates

The carbohydrates in soybeans consist of both soluble and insoluble carbohydrates. Insoluble carbohydrates, or fiber, in soybeans primarily function to provide structure to the bean in the seedcoat. Depending on method used, fiber makes up about 19.7-31.9% of the whole soybean. Much of this fiber is lost during processing when the hulls are removed, as the hulls are 86% fiber (Medic et al., 2014). To avoid waste, new end-uses are being explored for soybean hulls. Soybean hulls are commonly added to animal feed as an inexpensive energy source for

ruminants (Anderson et al., 1988). Hydrolyzed soybean hulls show promising use in active-film packaging applications (Jamróz et al., 2022). Soybean hulls can also be used to produce ethanol for biofuel through fermentation as a renewable energy source (Loman and Ju, 2016).

Soluble carbohydrates make up 11-25% of the whole soybean and assist with seed storage. Sucrose makes up about 1.1-7.4% of dry matter, followed by raffinose at 0.1-1.4% dry matter and stachyose at 1.2-6.9% dry matter. Other sugars such as verbascose, maltose, glucose, and fructose exist in lesser quantities (Medic et al., 2014). Raffinose-family oligosaccharides including sucrose, raffinose, stachyose, verbascose, and ajugose are not digested in monogastric animals and humans. Bacteria in the large intestine metabolize these sugars leading to flatulence and intestinal discomfort (Obendorf and Górecki, 2012). Current methods for reducing these undesirable effects include processing steps to remove raffinose-family oligosaccharides such as soaking, cooking, germination, dehulling, fermentation, and enzymatic treatment (Elango et al., 2022). Alternatively, selective breeding and genetic modification can be utilized to reduce soluble carbohydrate content; however, further research is needed to improve seed storage stability in soybeans with lower raffinose-family oligosaccharides (Obendorf and Górecki, 2012).

Ash

Soybeans accumulate minerals from soil as the soybean plant takes up water during the growing period. Ash, or mineral, composition of soybean seeds varies based on growing conditions and breed. Approximate values for potassium, magnesium, calcium, iron, zinc, manganese, copper, and chromium are 20.4, 3.23, 0.786, 73.0, 55.2, 33.2, 16.4, and 2.00 µg/g, respectively. Germination has no significant effect on mineral composition (Shi et al., 2010).

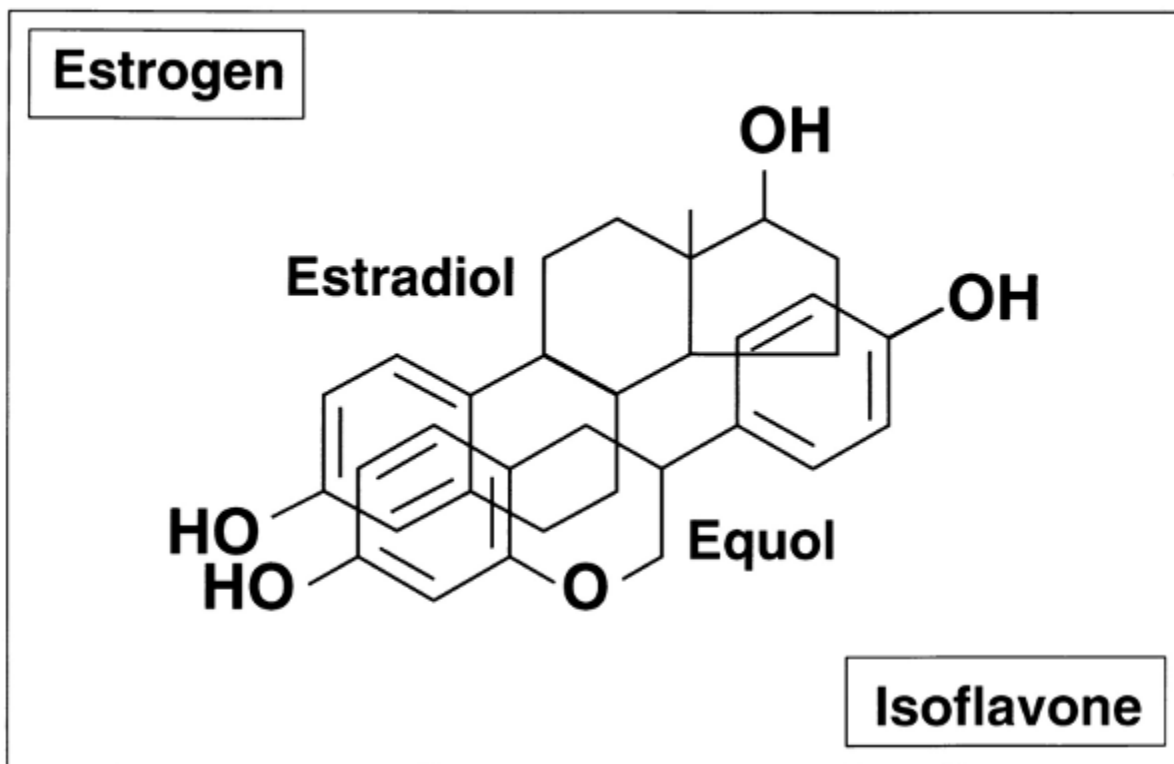
Isoflavones

Isoflavones are flavonoids found in soybeans in amounts ranging from 13.4–324.8mg per 100g while other legumes such as black beans, white beans, and peanuts contain amounts less than 1mg per 100g (Corcoran et al., 2012; Brodowska, 2017). Flavonoids are a type of polyphenol and are classified into six sub-classes: flavanols, flavones, flavonols, anthocyanidins, flavanones, and isoflavones. The two isoflavones present in soy are daidzein and genistein (Corcoran et al., 2012).

Health benefits associated with soy consumption such as decreased cancer risk, reduced menopause symptoms, and reduced risk of cardiovascular disease may be attributed in part to the presence of isoflavones (Křížová et al., 2019). Due to structural similarities (Figure 2.2),

isoflavones are also known as phytoestrogens and may bind to estrogen receptors resulting in estrogenic or antiestrogenic effects (Messina, 2016). Isoflavones are broken down through enzymatic hydrolysis during digestion or through bacterial hydrolysis during fermentation of soy foods.

SIMILARITY OF ISOFLAVONES TO ESTROGENS



Note. Setchell and Cassidy, Dietary isoflavones: biological effects and relevance to human health, Journal of Nutrition, 1999, volume 129, issue 3, Pages 758S–767S, by permission of the American Society for Nutrition

Figure 2.2—Comparison of estrogen and isoflavone chemical structures

The amount of isoflavone present in a soy food depends on the soybean cultivar and processing steps. Upon recognizing potential health benefits associated with isoflavones, researchers began efforts to breed high-isoflavone soybeans. Low isoflavone soybeans have about 8000 nmol per gram of dry matter while high isoflavone soybeans have about 15,000 nmol per gram of dry matter (Shao et al., 2009). Soy protein extraction results in isoflavone losses of 33-74% depending on extraction conditions (Wang et al., 1998; Lin et al., 2006; Shao et al., 2009).

Benefits of soy consumption

Individuals may choose plant-based proteins for health, sustainability, or ethical reasons (Beacom et al., 2021). Some individuals choose plant-based alternatives due to concerns about the environmental impact of livestock agriculture. In addition to exponential population growth globally, developing countries are consuming more animal products. For example, in China from 2000 to 2019 the average supply of animal based protein increased from 27.2 to 40.3g/capita/day (FAO, 2019). Increased consumption of protein is beneficial for nutrition and health of individuals in developing countries, however, this results in increased demand for animal products that can have detrimental impacts on the environment. Livestock agriculture has several disadvantages. Livestock production is an inefficient use of plant protein, yielding 1 kg of animal protein from 6 kg of plant protein. Additionally, 25kcal fossil energy is required to produce 1 kcal of animal protein (Pimentel and Pimentel, 2003).

Soy poses an alternative for milk, cheeses, and yogurts for those who do not consume animal products due to preferences or medical reasons. According to a 2018 Gallup poll, vegetarians make up about 5% of the U.S. adult population, while 3% are vegan (Jones and Saad, 2018). In the U.S., 36% consider themselves “flexitarians,” with a mostly plant-based diet but occasional consumption of animal products (Packaged Facts, 2020). Lastly, dairy allergies and dairy intolerances affect 0.5-3% and 65% of the global population, respectively (Storhaug et al., 2017; Flom and Sicherer, 2019).

The health benefits of soy consumption are thought to be attributed to protein and isoflavone content. In 1999, the FDA approved a health claim for products containing at least 6.25g of soy protein. A model health claim statement for eligible products is given: “25 grams of soy protein a day, as part of a diet low in saturated fat and cholesterol, may reduce the risk of heart disease,” and the amount of soy protein in each serving must be listed (21CFR101.82, 2001). Research indicates that soy consumption could reduce risk of heart disease, lower cholesterol, reduce risk of cancer, and reduce menopause symptoms from soy protein and isoflavones present in soy foods. One serving of whole, cooked soybeans contains 22g protein, while a serving of soy milk, tofu, tempeh, and soy sauce contains 7g, 6g, 5g, and 2g, respectively (Michelfelder, 2009).

The FDA approved a qualified health claim for soybean oil in July 2017. The health claim states consumption of 1 ½ tablespoons or 20.5g of soybean oil a day may reduce risk of heart disease when soybean oil replaces saturated fats. Manufacturers must list grams of soybean

oil per serving of the product (FDA, 2017). Research on behalf of the American Heart Association indicates replacing saturated fats with unsaturated fats reduces cardiovascular disease by 30% (Sacks et al., 2017). The qualified health claim was used instead of an authorized health claim because the scientific evidence did not meet FDA's required standards for "significant scientific agreement." Processors that choose to use the health claim must use qualifying language "supportive but not conclusive scientific evidence" when describing health claims (FDA, 2017).

Concerns associated with soy consumption

Despite the health benefits associated with consumption of soy, concern has been raised about the effect of soy on breast cancer risk, hormonal effects, and intestinal discomfort. One study found that rats treated with genistein, a product of isoflavone catabolism, experienced demasculization (Wisniewski et al., 2003). Because soy isoflavones are bind to estrogen receptors, consumption of soy could have estrogenic effects that have negative health consequences for endocrine function. Systematic reviews of multiple controlled studies on the effect of soy isoflavones on thyroid function found no effect on free triiodothyronine (fT3), free thyroxine (fT4) hormones and a mild 10% increase in thyroid stimulating hormone (TSH). The study concluded that a modest 10% increase in TSH is not concerning for the general population, however, individuals with hypothyroidism may need to avoid products high in soy isoflavones until further research is conducted with these specific populations to determine whether high consumption of soy is harmful (Otun et al., 2019).

Soy is also one of the top nine allergens listed by the FDA. Companies that produce foods that include soy are required to disclose the presence of soy in the product. In the U.S. self-reported prevalence of soy allergies is between 01-0.6%, although surveys typically overestimate true allergen prevalence (Messina and Venter, 2020). Allergic response to soy is due to the α' -, α -, and β -subunits of β -conglycinin, a seed storage protein. Research is underway to develop methods to reduce allergenicity of soy products through processing techniques. Heat treatment, enzymatic hydrolysis, and fermentation are all processes that denature the protein and prevent it from binding with antibodies that cause allergic response (Meinlschmidt et al., 2015). However, denaturation results in changes to protein sensory and functional properties. In some applications, the aforementioned methods induce desired changes such as the salty taste of fermented soy sauce. For such cases where protein denaturation is undesirable, a method for reducing allergenicity of soy proteins involves conjugation with a polyphenol to alter the protein

structure. The resulting protein-polyphenol conjugate also exhibits improved antioxidant activity, emulsifying capacity, and foaming capacity (Lin et al., 2022). Soybean products that are free of soy protein, such as soy lecithin and soybean oil, have not been found to cause allergic response (Nicolson and Settineri, 2021).

Soybean Processing

Soybeans are crushed to separate the co-products: soybean meal and soybean oil. Crushing one bushel of soybeans produces approximately 48 pounds of soybean meal, 11 pounds of soybean oil, and 1 pound of waste (Irwin, 2017). To prepare soybeans for oil extraction, soybeans are dried, cleaned, cracked, dehulled, conditioned, and flaked. After flaking, soybeans are ready for mechanical or solvent extraction. Mechanical extraction involves physically compressing the flakes to express oil using a hydraulic press, continuous screw press, or extruder (Woerfel, 1995). During hydraulic pressing, flaked soybeans are compressed with a hydraulic ram at 1500-1600 psi to physically express oil. Screw press extraction is a continuous process where soybeans are forced through a chamber by a rotating screw that gradually increases pressure up to 20,000 psi to 40,000 psi. (Kenyon et al., 1948). Solvent extraction, however, is the preferred method for oil extraction because the process extracts approximately 98% of the soybean oil as opposed to mechanical methods which yield about 70% oil extraction (Rotimi, 2014). Solvent extraction involves the use of a solvent, typically n-hexane, to dissolve the oil from the soybean flakes. After extraction, the solvent must be recovered (Woerfel, 1995).

Hexane is typically used for soybean oil extraction because it is nonpolar and easy to recover from the product. However, research is ongoing to seek out new solvents because of concerns associated with environmental and health consequences. During the oil extraction process, Hexane is released into the atmosphere where it may contribute to the formation of ozone (Kumar et al., 2017). Additionally, hexane is a neurotoxicant with dangerous health consequences. Acute inhalation at 5,000 ppm causes giddiness and dizziness, while chronic inhalation as low as 30 ppm for 2 months can have long-term serious neurological effects (Brown, 2022). Individuals exposed to n-hexane as an occupational hazard have experienced peripheral neuropathy after prolonged exposure (Wilson et al., 2007). Green solvents are being explored as an alternative to n-Hexane, including water, carbon dioxide, terpenes, and petroleum (Kumar et al., 2017). In particular the terpene limonene is of interest as alternative oil extraction solvent because it is inexpensive, environmentally friendly, efficient, and safe (Virot et al., 2008).

Soy Products

Soybean Oil

In 2022, global consumption of soybeans exceeded 60 million metric tons, representing 28.6% of global vegetable oil consumption (USDA and FAS, 2022). Soybean oil is used in foods as an ingredient in bakery products, salad dressing, and cooking oil (WWF, 2014).

Soybean oil is also high in lecithin, an emulsifier. Soy lecithin has a variety of applications in food and feed production as well as cosmetic and pharmaceutical applications. The emulsifying properties of soy lecithin are attributed to phospholipids which have two parts: a hydrophilic head and hydrophobic tail. Phospholipids surround oil phase droplets in a continuous water phase with the hydrophobic tail facing inside the oil droplet and the hydrophilic head facing out towards the water. The presence of these surface-active agents lowers surface tension to increase stability of emulsions (Deng, 2021).

In baked goods soy lecithin can be used as an egg replacer to aid in emulsification (Hedayati and Mazaheri Tehrani, 2018). Lecithin also acts as a lubricant to improve handling and reduce stickiness in bread dough, pie crust, tortillas, and Asian noodles. Additionally, lecithin is commonly used in large scale chocolate production to lower the viscosity and increase processing speeds. Lecithin is also widely used in nonfood applications, including as a release agent, adhesive, stabilizer, and lubricant (List, 2015).

Soybean Protein

Soy protein is extracted from soybean meal after the lipid fraction has been removed. Soy protein ingredients include soy flour, soy protein concentrate, and soy protein isolate which contain about 50%, 60%, and 90% protein respectively (Annor et al., 2014). Partial replacement of wheat flour with soy flour in baked goods can increase protein content, while complete replacement of wheat flour non-wheat flour blends is being explored for the production of gluten-free baked goods. Gluten-free corn flour bread has been shown to have improved nutrition and sensory properties with the inclusion of 15% soy flour (Taghdir et al., 2016). To improve sensory characteristics and shelf life, soy flour can be washed with hexane to produce defatted soy flour (Annor et al., 2014). Off flavors described as bitter or beany in soy flour may be attributed to lipid oxidation from lipoxygenase, therefore, a defatted soy flour may be better suited for the development of mild-flavored high protein foods (Wolf, 1975).

Hydrolysis

Soy proteins can be modified to change functional, sensory, and bioactive properties through hydrolysis, a process that breaks down proteins into smaller peptides in the presence of water using acids, enzymes, or bacteria (Wouters et al., 2016). Hydrolysis has three effects on the protein: a decreased molecular mass, increased number of ionizable groups, and increased hydrophobicity. Degree of hydrolysis (DH) is calculated as the number of peptide bonds cleaved (h) divided by the total peptide bonds (h_{tot}) multiplied by 100 to get a percentage (Equation 3).

$$\text{DH, \%} = \frac{h}{h_{\text{tot}}} \times 100 \quad (3)$$

Higher values indicate that more peptide bonds were cleaved and the protein was broken down more. Several methods exist to determine degree of hydrolysis, including pH-stat, osmometric, soluble nitrogen after trichloroacetic acid precipitation (SN-TCA), 2,4,6-trinitrobenzenesulfonic acid (TNBS), o-phthalaldehyde (OPA), amino acid nitrogen, and formol titration methods.

Hydrolysis can be performed using bacteria, enzymes, or acid. Enzymes are the preferred method for hydrolysis because bacterial hydrolysis is more difficult to control and acid hydrolysis is a more dangerous method that may result in unsafe products and undesirable side reactions (Campbell et al., 1996). Enzymes cleave preferentially to specific sites of protein structures to release peptides of varying size and polarity, therefore, enzyme choice is an important factor in determining functional properties of soy protein hydrolysates. In addition to enzyme choice, reaction time, temperature, and pH affect the size and polarity of peptides and by extension, physicochemical properties of the resulting hydrolysates.

Previous work has focused on evaluating physical properties of plant protein hydrolysates in model systems. Changes in protein structure as a result of hydrolysis impact their solubility, water and fat holding capacity, emulsifying and foaming capacity, and gelation properties (Wouters et al., 2016). Hydrolysis conditions differed among studies, therefore findings on the effect of hydrolysis on soy protein varied and these effects are summarized in Table 2.3 adapted from Wouters et al. (2016).

Table 2.3–Summary of studies of functional properties of soy protein hydrolysates as compared to native SPI

Property¹	Effect of Hydrolysis	Reference
Solubility	Increased (p<0.05) at pH 4.5 and 7	Qi et al., 1997
	Increased (p<0.05) at pH 6-8	Were et al., 1997
	Increased at pH 1-10, >60% increase at pI	Achouri et al., 1998
	Increased, >99% solubility at pH 2-9	Chiang et al., 1999
	Increased at pH 3-7	Jung et al. 2005
	Decreased at pH <4, Decreased at pH ≥6 using pepsin	Tsumura et al. 2005
Water Holding Capacity	Approx. 1300% increase	Were et al. 1997
Fat Holding Capacity	Increased	Briones-Martínez et al., 1997
	Increased by approx. 400%	Achouri et al., 1998
Foaming properties	Increased FC	Puski, 1975
	Increased FC and FS	Babiker, 2000
	FC, FS increased (p<0.05) at pH 6 and 8,	Were et al., 1997
	Increased	Molina Ortiz & Wagner, 2002
Gelation	Significantly (p<0.05) decreased gel strength	Lamsal et al., 2007
	Decreased gel strength	Fan et al., 2005
Emulsifying properties	Increased EAI (p<0.05) Decreased ESI (p < 0.05) at DH < 15%	Qi et al., 1997
	No change in EA or ES	Were et al., 1997
	Increased (p<0.05) EC, unchanged ES at 2% DH and increased ES at 4% DH	Jung et al., 2005

¹pI = isoelectric point, WHC = Water Holding Capacity, FHC = Fat Holding Capacity, EAI=Emulsifying Activity Index, ESI= Emulsifying Stability Index, EC= Emulsifying Capacity, ES= Emulsifying Stability EA = Emulsifying Activity DH=Degree of Hydrolysis.

Structural modifications resulting from hydrolysis also affect sensory properties of proteins. In general, protein hydrolysates exhibit increased bitterness due to the liberation of hydrophobic peptides (Neklyudov et al., 2000). In a study of soy protein hydrolysis with ten proteases, beany aroma was significantly decreased ($p<0.05$) for all hydrolysates as compared to native SPI, however, only Pancreatic Trypsin Novo[®] 6.0 S, papain, and Flavourzyme[®] hydrolysates did not significantly increase in bitterness as compared to native SPI (Meinlschmidt et al., 2015). In these studies, hydrolysate solutions were used to evaluate the sensory properties of soy protein hydrolysates. Further research is needed to evaluate the sensory properties of foods prepared with soy protein hydrolysates.

Proteases

Proteases, also known as peptidases, are enzymes that break down proteins into smaller molecular weight peptides and amino acids. Endopeptidases cleave at amino acids in the middle of a peptide, while exopeptidases cleave terminal amino acids (Van Der Velden and Hulsmann, 1999). A classification system was developed in 1993 that groups statistically similar protease sequences into families and families with similar tertiary structures into clans (Rawlings and Barrett, 1993). Information on a specific protease including classification, structure, and source organism has been summarized in the *MEROPS* database hosted by the European Bioinformatics Institute (EBI) (Rawlings et al., 2014).

Papain, an enzyme produced from papaya latex is a cysteine endopeptidase with broad specificity. Applications of papain are varied across food and nonfood industries (Fernández-Lucas et al., 2017). Papain is included as Generally Recognized as Safe (GRAS) by the FDA and is allowed in foods as an enzyme for processing or tenderization with no restrictions on level within current good manufacturing practice (FDA, 2016). In the meat industry papain is used as a tenderizer to improve lower-grade cuts of meat. In the baking industry papain is utilized as a dough relaxer. Additionally, the use of papain to reduce allergenicity of wheat flour is being explored. Papain may be used to remove insoluble protein aggregates in beer without negative sensory changes. Outside the food industry papain is used to remove stains on teeth, treat acne, and remove heavy metals in water. (Fernández-Lucas et al., 2017). A study on the production of soy protein hydrolysate found papain reduced beany and bitter flavors as compared to

unhydrolyzed soy protein isolate. Functional properties including emulsifying capacity, foaming stability, and water binding capacity were increased by hydrolysis with papain. (Meinlschmidt et al., 2015). Research is needed to investigate sensory and functional properties of papain soy hydrolysate in baked goods.

Flavourzyme[®], a commercial enzyme blend from Novozymes[®], is derived from the lactic acid bacteria *Aspergillus oryzae* and contains both endo- and exo-peptidases. In some studies, Flavourzyme[®] has been found to have debittering properties for animal and vegetable protein hydrolysates (Ma et al., 2013; Rezvankhah et al., 2021). Additionally, supplier data indicates debittering properties of Flavourzyme[®] (Novozymes[®]). Conversely, a study of pea protein hydrolysates found Flavourzyme[®] increased bitterness (Humiski and Aluko, 2007). A study on soy protein hydrolyzed with various enzymes found bitterness remained the same after hydrolysis with Flavourzyme[®]. Additionally, the resulting soy hydrolysates had significant ($p < 0.05$) increases in functional properties including emulsifying capacity and oil binding capacity. Significant decreases were observed in water binding capacity and foaming capacity (Meinlschmidt et al., 2015).

Soy Milk

Plant-based milks are an alternative milk to dairy for consumers with lactose intolerance or dairy allergies and include soy, coconut, rice, hemp, almond, and cashew milks. A comparison of nutrient content for cow milk and plant-based milks is given in Table 2.4. Of these plant-based beverages, soy milk has the highest protein content with 2.88g per 100mL. Soy milk has 38% less fat and 28% less calories than cow milk, while maintaining the same calcium level. Hemp and almond milk have less calories than soy milk, however, these milks also have less than 1g protein per 100mL milk (Chalupa-Krebsdak et al., 2018).

Table 2.4–Nutrient content of plant-based milks compared to cow milk

	Per 100 mL				
	Calories (kcal)	Protein (g)	Fat (g)	Carbohydrates (g)	Calcium (mg)
Cow, whole	61.0	3.15	3.27	4.78	113
Soy	44.20	2.88	2.03	3.53	113
Coconut	72.67	1.28	5.04	6.72	58.67

Rice	47.00	0.28	0.97	9.17	118
Hemp	19.00	0.83	1.25	2.50	12
Almond	18.00	0.76	1.02	1.66	160
Cashew	52.00	1.31	3.16	4.74	98.5

Note. Adapted from Chalupa-Krebzdak et al. 2018 Nutrient density and nutritional value of milk and plant-based milk alternatives. *International Dairy Journal*, 2018, issue 87, pages 84–92.

Nutrient standards for plant-based beverage were proposed and used to measure nutritional value of plant-based beverages on the United States Department of Agriculture (USDA) Branded Food Products Database. Nutrient standards included requirements for calories, protein, saturated fat, added sugar, and vitamins and minerals found in cow milk: calcium, vitamins A, D, B-2 and B12. The only plant-based beverages that met the proposed guidelines were fortified soy milks, indicating soy milk has similar nutritional value to cow milk (Drewnowski et al., 2021).

To process soybeans into soy milk, processors soak the soybeans to soften the cotyledon and reduce cook time. Additionally, stachyose and raffinose leach into the water during soaking, which is beneficial because flatulence and intestinal discomfort are decreased in the absence of these oligosaccharides. The beans are cooked and then ground. After grinding, the beans are filtered to separate the soy milk from the okara (Annor et al., 2014). On a dry weight basis okara contains 42.4-55.5% fiber, 15.2-33.4% protein, and 8.3-9.8% lipid, 3.8-5.15 carbohydrates, and 3.0-4.5% ash. Additionally, okara contains isoflavones which may offer health benefits (Li et al., 2012). As a result, value-added products utilizing okara are emerging. Fresh, undried okara can be used to prepare gluten-free cookies without the need to dry okara (Park et al., 2015). Okara fiber can be added to ice cream as a functional fiber or prebiotic, along with *Lactobacillus* bacteria to produce an ice cream with beneficial effects on gut microflora (Ibrahim et al., 2022).

Baking

Chemical Leavening

Baked foods such as breads, cakes, muffins, cookies, and biscuits all require leavening from yeast or chemical leavening agents. Leavening allows dough or batter to rise before setting

in a light and porous structure that gives desirable sensory properties including a fluffy texture and soft crumb (Pyler, 1988). Leavening agents cannot create new air cells, they can only expand existing cells. During mixing, air is physically trapped in the dough or batter by protein and/or solid fat (Baker and Mize, 1941; Smith, 1972). In chemically leavened products, carbon dioxide is liberated from bicarbonate by an acid through an acid base neutralization reaction. In the oven, carbon dioxide increases in volume due to an increase in temperature, causing the dough or batter to rise (Pyler, 1988).

Baking powder is considered a complete leavening system because it contains a source of carbon dioxide and an acid in the exact amounts required to neutralize. When the acid and base are reacted completely in an aqueous solution, the reaction produces salts. The acid and carbon dioxide source vary based on the product and process. The most common carbon dioxide source is sodium bicarbonate (baking soda), and the acid or acids are selected based on processing parameters and desired product characteristics (Pyler, 1988). Acid choice is primarily based on dough rate of reaction, which measures the speed of carbon dioxide released in dough prior to baking. Home baking powders are usually double acting, with one acid reacting during mixing and another acid reacting in the heat of the oven during baking (Brodie and Godber, 2007).

Maillard Browning Reaction

Maillard browning is a complex reaction that is responsible for non-enzymatic browning of thermally processed foods including bread, coffee, and seared meats. During the Maillard Reaction, amino groups react with reducing sugars to form Maillard reaction products including aroma compounds and melanoidins, which are responsible for brown color change.

In addition to color and flavor changes, Maillard reaction products induce changes to protein functionality and results in beneficial and potentially harmful health impacts (Tamanna and Mahmood, 2015).

In bakery products, the Maillard reaction is primarily of interest during the final stages of baking because it responsible for flavor and color changes from crust browning. The Maillard browning reaction is influenced by pH, temperature, water activity, and composition of food. An increase in amino acids or proteins in baked goods has been shown to increase browning. Maillard browning also can be increased by increasing the amount of reducing sugars (Lund and Ray, 2017).

Structure Formation in Bakery Products

Gluten proteins are also partially responsible for the structure set in chemically leavened bakery foods. Starch granules are embedded in the gluten matrix which sets into a semi-rigid structure during heat denaturation. After gluten proteins are denatured, they are no longer able to bind water, and excess water is taken up by starch granules during starch gelatinization (van der Sman and Renzetti, 2021).

Starch gelatinization is the irreversible process where starch granules absorb water while being heated, destroying molecular order within the granule. The granules rupture and leach starch molecules, primarily amylose, into solution (Schirmer et al., 2015). Structural setting of bakery products is partly attributed to starch gelatinization. The starch gelatinization temperature for a particular system varies based on formula factors such as salt, sugar, fat and protein content as well as starch type. Starch gelatinization temperature can be measured using Differential Scanning Calorimetry (DSC) or Thermogravimetric Analysis (TGA) (Goranova et al., 2019).

Protein-Starch Interactions During Baking

The interaction of protein with other macromolecules is important to investigate when formulating high-protein bakery foods. Protein-starch interactions have been investigated in a variety of applications. Whey and casein have been highly researched, and some papers compare the properties of animal proteins to plant proteins. Few studies exist to investigate the interactions between soy protein and starch. In general, protein and starch are found to interact through hydrogen bonds (Yang et al., 2019).

Muffins

Muffins are a cake-like product made from a batter that is chemically leavened rather than yeast leavened, so proof time before baking is not required. A leavening acid reacts with a carbon dioxide source to release carbon dioxide gas that causes oven spring (Pyler, 1988). Muffins are baked in round baking pans with or without paper liners to obtain discrete serving sizes. The FDA Reference Amounts Customarily Consumed (RACC) for muffins is 55g (21CFR101.12). Muffins typically contain inclusions such as fruits, nuts, chocolate chips, spices, meats and/or herbs.

Muffins are formulated and prepared in a similar way as cakes, which use a solid, plastic fat such as shortening, butter, or margarine to physically trap air during mixing. Flour, leavening agents, and liquids (water, milk, oil) are added at specific times depending on if a single or multi-stage mix is used. Trapped air during mixing can be measured using specific gravity. Specific

gravity is calculated as the weight of batter in a cup divided by the weight of water in the same cup. The optimum specific gravity for similar bakery foods, yellow and white cakes, has been found to range from 0.94-0.97 and 0.95-0.97, respectively (Pyler, 2009).

The baking process for cakes and cake-like products is the process by which fluid batter is transformed into a set, porous structure. Heat causes the structure to expand, increasing the volume of the product. At 98.4-98.9 °C, moisture evaporation, starch gelatinization, and protein coagulation occur in the system at the fastest rate. When enough water evaporates to form a sufficiently dry crust, Maillard reactions may occur to cause browning (Pyler, 1988). If the baking parameters including time and temperature are appropriately selected, the structure of the crumb also will be formed. Enough water will evaporate from the crumb to leave a stable network of protein and starch which makes up the air-continuous foam after gas escapes during baking (Schwartzberg and Hartel, 1992).

A Commercial Item Description (CID) defined by USDA’s Agricultural Marketing Service (AMS) outlines expected characteristics of muffins (USDA and AMS, 2021). Muffins may be plain or contain inclusions such as chocolate, fruit, vegetables, nuts, or spices. Muffins may also be topped with sugar streusel topping. Table 2.5 summarizes information about expected sensory characteristics for muffins.

Table 2.5–Summary of AMS Commercial Item Description (CID) for muffins

Attribute	Expectation
Appearance and Color	Round top, crust and crumb color corresponds to labelled muffin flavor, evenly baked without burning
Flavor and Aroma	Flavor and aroma correspond to labelled muffin flavor, no off flavors such as burnt, scorched, stale, rancid, or moldy
Texture	Slightly moist, light and tender crumb

Protein in Baked Goods Trend

The baking industry has reported an increase in demand for protein in baked goods, with an expected compound annual growth rate (CAGR) of 5% through 2025 (GMI, 2019). Consumers are interested in boosting protein intake for a variety of health benefits, including muscle development, increased satiety, weight loss, and appetite stimulation (Henchion et al., 2017). In particular, the market for soy protein ingredients is expected to experience a CAGR of

6.5% from 2019-2025 (GMI, 2019). Research is ongoing to investigate the effect of added soy protein on physical and organoleptic properties of baked products.

Soy Protein in Baked Goods

Added soy protein in baked products has been found to increase water absorption of batters and doughs. Sana et al. (2012) found addition of 7-22% soy flour to bread increased water absorption. A control bread dough without soy flour had a water absorption of 64.5% while addition of 7% soy flour increased water absorption to 65.5% and 22% soy flour increased water absorption to 72.5%. Without adding extra water to compensate for increased absorption, the resulting dough or batter can be dry or crumbly. To address quality concerns water should be added when reformulating for the addition of soy protein (Nogueira and Steel, 2018).

The addition of soy protein can also interfere with starch gelatinization, which is a key element of the structure of baked goods. Increased protein content dilutes the starch and creates competition for water. Collapse of baked products during cooling is thought to be attributed to decreased starch gelatinization (de la Hera et al., 2012).

Soy protein is a highly refined product and many flavor compounds are removed during processing, however some flavor compounds remain. A “beany” flavor is often identified in the final product when higher amounts of soy protein are added. Brewer et al. (1992) tested the addition of 40%, 50%, and 60% SPI to muffins and found “beany” and “grain-like” flavors were significantly increased by addition of SPI. Research is ongoing to reduce off flavors in soybean products, including thermal or chemical denaturation, thermal or chemical inactivation of protease inhibitors, or enzymatic hydrolysis (Barac et al., 2004). Further research is needed to address challenges associated with the addition of soy protein to baked goods found by studies in order to meet consumer demand for high-protein bakery products.

Education

Hands-on Learning

Hands-on learning increases student engagement, encourages higher-order thinking, and supports development of soft skills. Engagement can be defined using three dimensions of engagement. Behavioral engagement describes the extent of student participation and effort. Emotional engagement is related to student’s feelings about the subject and topic, classmates, and teachers. Cognitive engagement involves effort in mental processing of content (Fredricks et al., 2016). Increased student engagement is correlated with increased academic achievement,

higher test scores, reduced drop-out rates, and reduced behavioral problems (Fredricks et al., 2004; Scheidler, 2012; Hao Le et al., 2018).

Hands-on activities designed to address all dimensions of engagement are beneficial for students. During hands-on activities students are actively involved and physically interact with materials, which facilitates behavioral engagement. Choosing a hands-on activity that is personally relevant or interesting to students encourages emotional engagement. Lastly, hands-on activities can support cognitive engagement by creating opportunities to develop higher order cognitive skills with appropriate educational supports (Krzic et al., 2018).

In addition to boosting student engagement, hands-on learning promotes higher-order thinking. Bloom's taxonomy is used in curriculum and course design to measure depth of learning from the lowest level (remember) to the highest level (create). Table 2.6 shows the hierarchy of cognitive processes in the revised Bloom's taxonomy. Categories are in order from least to most complex with some overlap allowed in the complexity of cognitive processes (Krathwohl, 2002). During hands-on activities, students are encouraged to use more complex cognitive skills found in higher levels of Bloom's taxonomy. For example, in a laboratory class students execute a series of planned steps, analyze results, and produce a report communicating the results. These hands-on learning opportunities foster higher order thinking.

Table 2.6—Revised cognitive processes of Bloom's taxonomy

Categories	Definition	Cognitive Processes
Remember	Retrieving relevant knowledge from long-term memory.	1.1 Recognizing 1.2 Recalling
Understand	Determining the meaning of instructional messages, including oral, written, and graphic communication.	2.1 Interpreting 2.2 Exemplifying 2.3 Classifying 2.4 Summarizing 2.5 Inferring 2.6 Comparing 2.7 Explaining

Apply	Carrying out or using a procedure in a given situation.	3.1 Executing 3.2 Implementing
Analyze	Breaking material into its constituent parts and detecting how the parts relate to one another and to an overall structure or purpose.	4.1 Differentiating 4.2 Organizing 4.3 Attributing
Evaluate	Making judgments based on criteria and standards.	5.1 Checking 5.2 Critiquing
Create	Putting elements together to form a novel, coherent whole or make an original product.	6.1 Generating 6.2 Planning 6.3 Producing

Note. Adapted from Krathwohl 2002

Upon graduation students are expected to have attained both subject knowledge and skills necessary for success in the industry. The Institute of Food Technologists' (IFT) core competencies were released in 2001 and are grouped into five categories: food chemistry and analysis, food processing and engineering, food safety and microbiology, applied food science knowledge, and success skills. Morgan et al. (2006) surveyed industry professionals who rated applied food science knowledge and success skills as most important. While the IFT core competencies are now referred to as "standards," implications from the 2006 survey are still useful in determining how effective Food Science programs are. Both categories involve less subject knowledge and more soft skills. Table 2.7 indicates the core competencies in each category. In addition to general subject knowledge, employers in the food industry expect graduates to have certain soft skills, such as the ability to work on a team, to apply knowledge from one situation to another, and the ability to troubleshoot problems.

Table 2.7—Institute of Food Technologist's core competencies for applied food science knowledge and success skills

Category	Core Competency
Applied food science knowledge	Is able to apply and incorporate the principles of food science in practical, real-world situations and problems.

Is able to apply the principles of food science to control and assure the quality of food products.

Knows how to use computers to solve food science problems.

Understands government regulations required for t
manufacture and sale of food products.

Is aware of current topics of importance to the food industry.

Is able to apply statistical principles to food science applications.

Understands the basic principles of sensory analysis.

Success Skills

Defines a problem, identifies potential causes and possible solutions, and make thoughtful recommendations.

Commits to the highest standards of professional integrity and ethical values.

Handles multiple tasks and pressures.

Demonstrates the use of oral and written communication skills. This includes such skills as writing technical reports, letters and memos; communicating technical information to a nontechnical audience; and making formal and informal presentations.

Manages time effectively.

Works effectively with others.

Able to apply critical thinking skills to new situations.

Deals with individual and/or group conflict.

Works and/or interacts with individuals from diverse cultures.

Provides leadership in a variety of situations.

Can independently research scientific and nonscientific information.

Facilitates group projects.

Is able to explain the skills necessary to continually educate oneself.

Laboratory courses provide opportunities for students to develop and improve these soft skills. Students may work in teams to complete laboratory objectives. Students also may observe unexpected results and generate possible explanations for deviations. In food science specifically, hands-on learning allows students to become more familiar with qualitative observations during food processing, such as the strength of a bread dough during mixing or the color of meat during thermal processing. Additionally, students learn common methods outlined by field specific scientific associations and basic analysis of data.

Online Learning

In the 2019 fall semester, 36% of U.S. undergraduate students were enrolled in at least one online course (Irwin et al., 2021). During the COVID 19 pandemic, 73% of U.S. students were enrolled in online courses (NCES, 2020). Students in STEM majors take laboratory courses to gain hands-on experience in their field. STEM majors who enroll in an online course may lack opportunities to gain necessary hands-on experience.

Non-traditional students exhibit a preference for online courses, therefore, improving quality of online courses may increase enrollment and reduce dropout rates for this demographic. Students with one or more of the following risk-factors for non-completion of their degree are considered non-traditional students: delayed enrollment, no high school diploma, part-time enrollment, financially independent, have dependents, single parent status, working full-time while enrolled. Non-traditional students are more likely to enroll in distance courses and report higher satisfaction with distance courses than traditional students. Online courses allow students with time constraints due to family and work the flexibility required to successfully participate in courses (Pontes et al., 2010).

Online courses often lack hands-on components. In a survey of STEM faculty and administrators during the COVID-19 pandemic, 34% of respondents selected inadequate online laboratories as a barrier to online STEM education (Seaman et al., 2021). Implementing hands-

on activities in online courses can pose challenges as students living situations vary considerably. Some students may have access to household kitchen tools and appliances, while others may be living in university housing and lack access to these resources. Labs designed for on-campus courses often utilize special equipment and reagents to give students specialized experience. To address concerns with access to lab materials, kits can be mailed to students containing all or some materials necessary for the lab. These kits can be assembled by on-campus instructors or ordered from science laboratory suppliers.

Some laboratory courses utilize virtual laboratories such as virtual reality experiences, video experiments, or animated interactive laboratories. These virtual experiences can be engaging and encourage active learning; however, they lack opportunities for learners to receive sensory input about the experiment.

Another challenge for online at-home laboratory exercises is the lack of supervision from instructors. Students in on-campus labs typically have questions while performing the experiment, however these questions may not be answered in time to support the at-home lab. In a study comparing online and on-campus chemistry laboratories, several students responded in a reflective survey that on-campus laboratories were preferred over virtual due to instructor availability (Rowe et al., 2018). Additionally, lack of supervision may pose safety concerns when dealing with hazards. Students may not have basic skills surrounding hazards such as cutting, heating, or chemical handling. In an on-campus course, these skills can be addressed prior to the lab in a demonstration or corrected as the instructor supervises the lab activities. Instructions for the laboratory must be detailed and easy to understand to reduce frustration and errors in at-home laboratories. At-home labs must consider the safety of assigned activities and consider providing appropriate support in the form of video demonstrations or written instructions (Holmes and Wieman, 2016).

Hands-on activities for online learners have been implemented successfully in many undergraduate science courses. Yeerum et al. (2022) developed an at-home analytical chemistry lab using guava leaf extract as a color indicator to measure iron solution concentrations. Lab kits with materials and reagents were prepared and sent to students in advance, and the laboratory was conducted synchronously over an online video communication platform. Post-lab quiz scores were higher ($p < 0.05$) for online students compared to on-campus students and mean overall satisfaction for the at-home lab was above 4 on a Likert scale (1=least satisfied, 5=most satisfied). When designing the lab activity, the authors selected materials and equipment that

posed little to no safety risks. Laboratory procedures were designed to be easily completed by students lacking formal lab experience.

Santiago et al. (2022) developed an at-home laboratory exercise for chemical engineering students. Students investigated reaction kinetics involved in treating wastewater containing methylene blue dye. Students also were provided a lab kit with reagents and materials necessary for completion of the lab. After completing the laboratory, students responded to a survey with a 5-point Likert scale (1=strongly disagree, 5=strongly agree). Post-lab survey results indicated the activity was positively received by students overall, and all students responded the instructions provided were clear enough to facilitate completion of the activity.

In both studies, the authors designed laboratory activities that are minimally hazardous and do not require advanced laboratory skills. Students were provided lab kits with specialized materials to reduce the burden of acquiring materials. Additionally, the instructors wrote detailed instructions for laboratory procedures to support students working remotely. These strategies are useful in the design and implementation of an at-home lab activity for food science courses.

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Chapter 3 - Comparing Online Soybean Processing Module Including a Laboratory Component to On-Campus Module

Introduction

In the fall of 2019, 36% of all undergraduate students at United States (U.S.) universities were enrolled in at least one online course, and 15% of all undergraduate students were enrolled exclusively in online courses (Irwin et al., 2021). During the COVID-19 pandemic, online course enrollment increased further as universities closed physical campuses. In the fall 2020 semester, 61% of undergraduates reported a change in class format from in-person to online or hybrid models.

During the COVID-19 pandemic faculty rapidly adapted course material for online courses. Laboratory components pose unique challenges when adapted for online learning, however, the inclusion of hands-on learning is important. Hands-on activities increase retention of learning outcomes and develop soft skills desired by potential employers (Hollis & Eren, 2016). In a 2005 survey by Purdue University, food science industry members rated the importance of the core competencies given by the Institute of Food Scientists (IFT). Success Skills plus Applied Food Science Knowledge were rated as most important content categories (Morgan et al., 2006). Success skills include oral and written communication, critical thinking, professionalism, information acquisition, teamwork, and organization (Hartel, 2001). In addition to these success skills, employers also expect graduates to be able to apply food science principles to practical issues. The demand for graduates with these skills presents an opportunity for undergraduate programs to improve the career readiness of their graduates. Online courses that include hands-on activities designed for a home environment provide experiences that are typically gained in on-campus courses. Educational programs that implement these activities better serve students by preparing them to meet industry demands.

One approach for hands-on activities that also encourages application of food science principles is problem-based learning. Instructors that implement problem-based learning present students with an issue and encourage them to explore solutions (Yew & Goh, 2016). A current problem the food industry faces is developing products that meet demand for protein-rich convenience foods (Sloan, 2020). Brands that feature high protein foods have experienced an increase in sales. For example, total sales for the high-protein bakery brand, Kodiak Cakes, grew from \$6.7 million in 2014 to \$160 million (Peckenpaugh, 2020). A 2021 Food and Health study found that 62% of Americans ages 18-80 are trying to consume more protein (International Food

Information Council, 2021). In addition to demanding more protein, consumers are more informed when choosing a protein source. Consumption of plant-based, complete proteins is important to 67% of adults that purchase protein foods (USB 2019). To meet the demand, food companies must develop plant-based, protein-rich foods that maintain quality characteristics of non-fortified products.

Soy protein is of interest in baked goods with added protein because it is a highly digestible plant-based protein (Annor et al., 2014). Soy protein has a Protein Digestibility Corrected Amino Acid Score (PDCAAS) of 1.0, similar to meat, dairy, and eggs (Hughes et al., 2011). Quick breads such as muffins are inexpensive and easy to prepare, and these qualities allow online undergraduate students to prepare multiple treatments with added soy protein and observe differences at home. The objectives of this study were to 1) develop an online module with a laboratory component covering the addition of soy to foods and 2) compare students' perceptions and performance with a similar on-campus module in an undergraduate food processing course.

Methods

The online module was first implemented in fall 2020 in the online Fundamentals of Food Processing course (FDSCI 305) at Kansas State University and repeated across four additional semesters (spring 2021, summer 2021, fall 2021, and spring 2022). FDSCI 305 is taught at the sophomore level; however, the course is open to all undergraduate students. A total of 94 students were enrolled across all five semesters. Students accessed course materials via the online learning platform Canvas (Instructure, Inc., Salt Lake City, UT).

The module consisted of an audio lecture with slides that covered background information on soy products processing, a companion textbook chapter (see lecture section), a hands-on laboratory exercise, a discussion board question, and exam and quiz questions (Figure 3.1). At the end of the module in both semesters, students voluntarily completed a student reflective survey to assess module efficacy.

Online Module Components

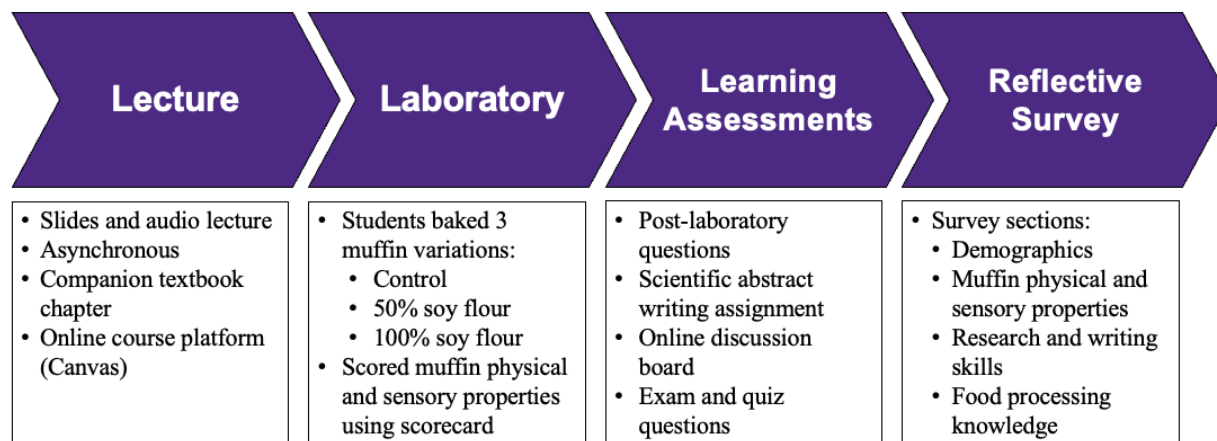


Figure 3.1–Summary of module components

Lecture

Instructors (authors) provided students with a prerecorded audio lecture with slides on Canvas. A companion textbook chapter from Food Processing: Principles and Applications titled “Crops — Legumes” by Annor, Ma, & Boye (2014) was used to develop the lecture. Topics covered included soybean production, composition, nutrition, and processing methods. The lecture also included an overview of common ingredients made from soybeans, including soybean oil, soy flour, soy milk, and soy protein. To connect the lecture to the laboratory exercise (preparation of soy muffins), the advantages and disadvantages of soy as an ingredient in foods were discussed.

Laboratory Exercise

Online students completed a hands-on laboratory exercise in their home kitchens. The purpose of the exercise was to encourage students to explore soy protein as an ingredient for increasing the protein content in muffins and to evaluate the effect on muffin physical and sensory properties. Laboratory instructions were provided to students including formulations, muffin preparation steps, a muffin score card, post-laboratory questions, abstract writing instructions, abstract grading rubric, and supporting material. The full instructions handout is available in Appendix A1 and the supporting material is available in Appendix A2.

Students were informed about the increased demand for protein-rich baked goods and explored muffins enriched with defatted soy flour to fill this demand. Students investigated the effect of defatted soy flour (Prolia® FLR-200/70), donated by Cargill (Minneapolis, MN), on

physical and organoleptic properties of muffins. Baking kits containing three muffin mixes (Table 3.1) were prepared by the teaching assistants and mailed to students to reduce the chances of experimental error. Each mix contained a commercial muffin base (Muffin Base 10 2.0; Product #139037) donated by Corbion (Lenexa, KS) plus added flour and granulated sugar. For the control mix, 100% of the added flour was all-purpose flour. The added flour in the 50% soy flour mix contained 50% soy flour and 50% all-purpose flour. The added flour in the 100% soy flour mix contained 100% soy flour. Students followed the laboratory instructions to prepare muffins in their home kitchens using their own oil, eggs, and water (Table 3.2).

Table 3.1–Muffin mix formula variations with gravimetric measurements

Ingredients	Weight (g)		
	Control	SF50	SF100
Soy Flour	0.0	50.6	101.2
AP Flour	101.2	50.6	0.0
Sugar	124.6	124.6	124.6
Muffin Base ¹	59.4	59.4	59.4

¹Muffin Base Ingredients: Enriched Wheat Flour (Wheat Flour, Niacin, Reduced Iron, Thiamine Mononitrate, Riboflavin, Folic Acid), Modified Corn Starch, Corn Syrup Solids, Whey (Milk), Soybean Oil, Sodium Aluminum Phosphate, Salt, Sodium Bicarbonate and 2% or Less of Each of the Following: Propylene Glycol Esters of Fatty Acids, Xanthan Gum, Mono- and Diglycerides, Sodium Stearoyl Lactylate (SSL), Sodium Carboxymethyl Cellulose, Diacetyl Tartaric Acid Esters Of Mono-Diglycerides (DATEM), Artificial Flavor.

Table 3.2–Muffin formula with volumetric measurements included in laboratory instructions

Ingredients	Amount		
	Control	SF50	SF100
Muffin Mix ^a	1 package	1 package	1 package
Oil ^b	1/2 cup	1/2 cup	1/2 cup
Eggs ^b	2 whole eggs	2 whole eggs	2 whole eggs

Water^b

1/3 cup

1/3 cup

1/3 cup

^aSee Table 3.1 for muffin base ingredients, ^bPurchased and measured by students

After preparing the muffins, students evaluated physical and organoleptic properties according to a muffin score card (Appendix A3) adapted from Foods: Experimental Perspectives (McWilliams, 2001). The scorecard was divided in two sections: external and internal qualities. External qualities included volume, contour, and crust color while external qualities included crumb color, cell uniformity and size, thickness of cell walls, texture, flavor, and aftertaste. Within each section muffin quality descriptions with corresponding numerical scores were given. The last section of the scorecard includes a rating for overall acceptability from 1 (very unacceptable) to 5 (very acceptable). Upon completing the lab, students were assigned post-laboratory questions to guide their reflection of the hands-on exercise (Appendix A4). As a resource for answering the post-laboratory questions, the laboratory instructions included background information on muffin quality parameters and a muffin commercial item description (CID) from the United States Department of Agriculture's (USDA) Agricultural Marketing Service (AMS) (2021).

Student Learning Assessment

Students wrote a scientific abstract to present their findings and make a recommendation on a level of soy flour for further research. The laboratory instructions provided students with information on writing scientific abstracts and the grading rubric (Appendix A5). A sample abstract from a relevant paper was included with annotations explaining different sections of an abstract to assist students in writing their abstracts. Abstracts were graded using the rubric with the following sections: introduction, objectives, materials and methods, results, professional writing, and length. Student understanding also was assessed with post-laboratory questions, a discussion question assignment, ten-question quiz, essay exam question (Table 3.3). Answers to post-laboratory and quiz questions are available in Appendix A6.

Table 3.3–Learning Assessment Questions

Post Laboratory Questions

Explain the difference between a variation and a replication.

Create a flow diagram of how a muffin processing line might look in a commercial setting. This can be handwritten and attached as a picture, created in excel and attached as a separate document, or pasted below. Make sure you include receiving of ingredients AND packaging material and storage of ingredients AND packaging material.

List 2 advantages and 2 disadvantages of soy as an ingredient in food.

Use the sample abstract from “Physicochemical and nutritional properties of a healthy snack chip developed from germinated soybeans” above to answer the following questions:

What are the variations in this experiment? This will be different levels of the independent variable.

What is being measured in this experiment? List at least 3 measurements.

In the sample abstract taken from “Physicochemical and nutritional properties of a healthy snack chip developed from germinated soybeans” was the glycemic index statistically different between the germination variations? How do you know? (Hint: check the P-value!)

Use the Pre-Lab Information and Supporting Material to answer the following questions. Circle the answer.

T or F Muffins made with bread flour are likely to have tunnels because of the high gluten potential.

T or F Excessive baking soda results in a muffin with a soapy, bitter flavor and a yellow color and coarse texture.

T or F Wrapping muffins before cooling will increase the shelf life by increasing the moisture content.

T or F According to the information and your observations, the protein content in soy flour causes more Maillard browning reactions to occur.

T or F Adding ham, cheese, or dried fruits to muffin batter will decrease the shelf life of muffins.

Undermixing muffins will result in a product with the following characteristics:

- A. Thick cell walls
- B. Pockets of unmoistened dry ingredients
- C. Low volume
- D. Both b & c are correct
- E. All of the above are correct

Emulsifiers such as monoglycerides and diglycerides function as fat substitutes by:

- A. Binding water
- B. Inhibiting hydration
- C. Promoting gelatinization
- D. Forming an outer crust that retains moisture

Maillard browning results from a reaction between the aldehyde group of glucose and other reducing sugars and:

- A. The hydroxyl group of water
- B. Sulfide groups in gluten
- C. The amino group of protein
- D. Carboxyl group of amino acids

The quantity of alternative flours that may be substituted for all-purpose flour for an acceptable muffin is:

- A. 10% to 20%
- B. 33% to 50%
- C. 75% to 100%
- D. 100%

An expected characteristic of muffins made by substituting all whole wheat flour for all-purpose flour is:

- A. Peaked Top
- B. Tunnels
- C. Tough
- D. Crumbly

T or F The CID for muffins does not require the use of Grade A Pasteurized Milk.

T or F According to the picture provided for SF100, it would not be acceptable by CID due to evidence of scorching or burning.

T or F There are specific age requirements for both fresh and frozen muffins that are determined by time and temperature parameters.

T or F It is acceptable for a nuts or fruit be to unevenly distributed in the muffin.

Discussion Questions

After watching the soybean lecture and completing the soy muffins lab, how has your perception of soybeans or soy as an ingredient changed? What benefits do you see for consuming soybeans and soy products? Are there any barriers for you personally consuming soy?"

Essay Question

Identify one product made from soybeans. Describe the processing steps required to make the product. List two nutritional benefits of consuming soy. List one barrier to soy consumption discussed in the lecture.

Quiz Questions

A product extracted from soybeans similar to eggs that serves as a binding agent:

- | | |
|----------------|-----------------|
| A. Soybean oil | C. Lecithin |
| B. Soy protein | D. Soybean meal |

Fermentation assists with the following:

- | | |
|---|-------------------------------------|
| A. Deactivating the antioxidant activity | C. Improving nutrient digestibility |
| B. Decreasing the phenolic compound concentration | D. All of the above |

True or False: Soybeans may turn a purple color due to drought and a specific fungus problem.

Match the soy product with a specific food application.

- | | |
|--------------------------|-----------------------|
| 1. Traditional Foods | A. Miso |
| 2. Okara | B. High fiber Breads |
| 3. Non-traditional foods | C. Soynut butter |
| 4. Lecithin | D. Emulsifying agents |
| | E. Isoflavones |

Match the term with the definition.

- | | |
|--------------------------|--|
| 1. Canning | A. Commercially Sterile |
| 2. High-Pressure Cooking | B. Hydrostatic Pressure |
| 3. Extrusion | C. Twin-screw pushes product through a die |
| 4. Soaking | D. Softens cotyledon |
-

On-Campus Module Summary

Student data for the on-campus module was collected in the Spring 2021 and Spring 2022 on-campus Fundamentals of Food Processing courses. Students attended an in-person lecture with the same slides, content, and verbal explanations as the online lecture. The laboratory exercise was modified to include more treatments to accommodate a large class and to utilize lab equipment not typically available in a home setting such as digital calipers and scales (Appendix A7). Post-laboratory questions, discussion questions, quiz questions, and the exam essay question were kept consistent between online and on-campus modules. The post-laboratory assignment with questions is available in Appendix A8. Students responded to similar reflective surveys anonymously online. The main difference between the surveys was Question 13 which references the students' ability to work independently (in the online class) or as a group (in the on-campus class).

Student Reflective Survey

A survey was modified from Heerman et al. (2020) to assess students' perception of the soybean module and at-home laboratory exercise. The study was reviewed and approved by the Kansas State University Institutional Review Board under IRB number 10248 (Appendix A9). Informed consent was obtained (Appendix A10) from each subject prior to completing the reflective survey. All students enrolled in Fundamentals of Food Processing completed the

module assignments (exam and quiz questions, laboratory exercise, report) as part of the Food Processing course, while participation in the reflective survey was optional and anonymous. The survey was distributed to online and on-campus students using a link to the online survey platform Qualtrics (Provo, UT; Seattle, WA). Students who completed the survey earned five bonus points towards their class grade. To maintain anonymity, bonus points were awarded based on submission of a screenshot of the “End of Survey” webpage. Bonus points may have created some bias but was consistent for both modules. The debriefing statement is available in Appendix A11 and the full survey is available in Appendix A12.

Results and Discussion

Student Learning Assessment

Scientific Abstract Assignment

All abstract assignments were graded by the same course teaching assistant and author (Brown). Some bias may have been introduced due to the lack of an independent grader, however, the grader remained consistent for all students and a rubric was used to standardize scoring. In the online course 72-93% of students met or exceeded expectations on every rubric section (Figure 3.2). In the on-campus course 92-98% or more of students met or exceeded expectations on every rubric section. Students in the online course scored lowest on the length rubric section with 73% of students meeting or exceeding expectations. In both sections the professional writing section had the highest scores.

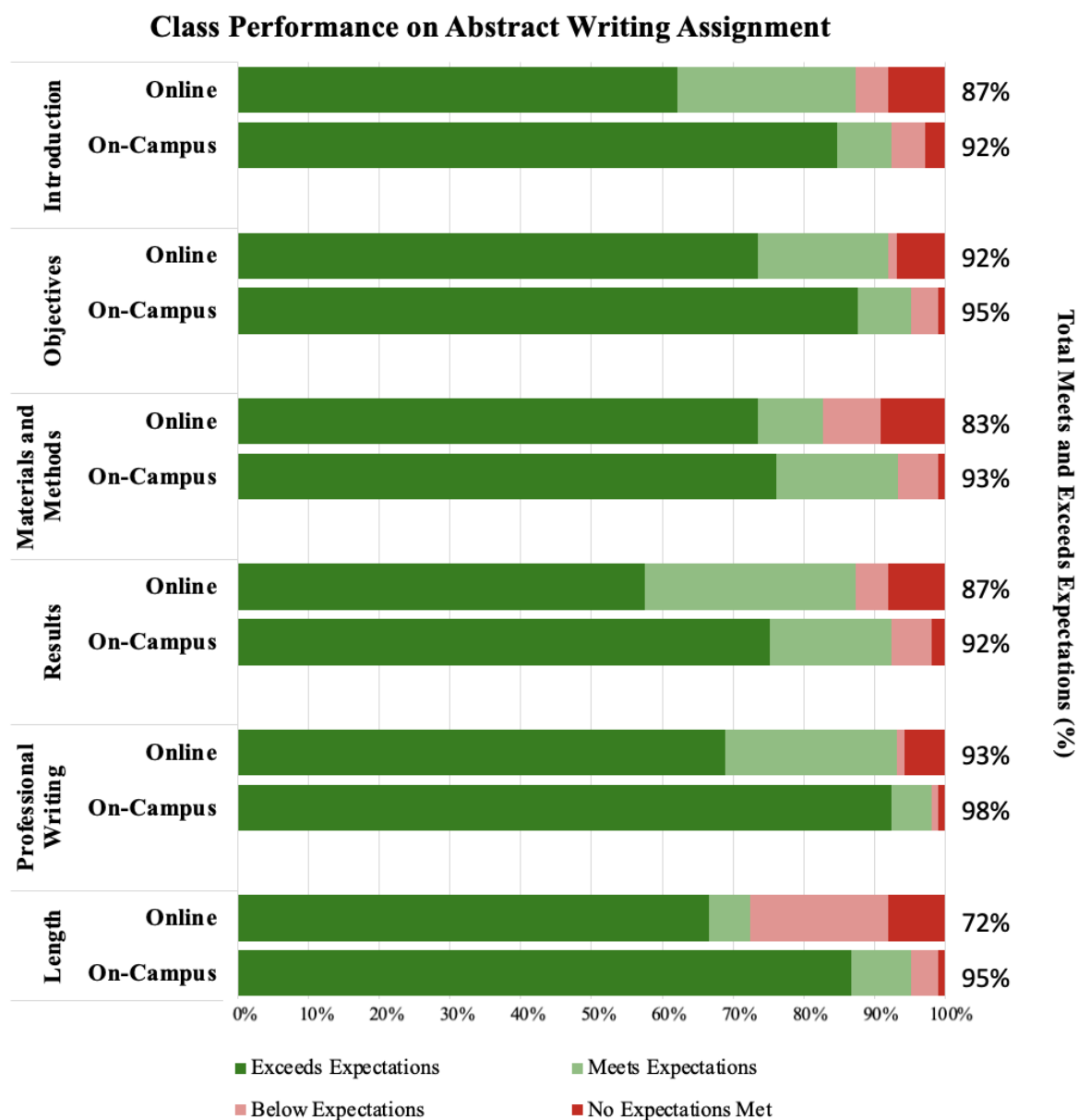


Figure 3.2—Comparison of online and on-campus class performance on abstract writing assignment

Students in the online course may have scored lower as they had written and received feedback on one scientific laboratory report prior to the muffin laboratory exercise. The on-campus students had written three scientific laboratory reports prior to the muffin exercise. Therefore, on-campus students had more opportunities to improve their scientific writing skills prior to this assignment.

Discussion Boards

After completing the lecture and laboratory activity, students participated in an online discussion board on Canvas. Table 3.4 lists the questions and summarizes student responses. Overall, students reported an increased awareness of food products that contain soy and methods for processing soybeans into ingredients. Students also reported an improvement in the perception of the taste of soy products. A common response was that students would be more likely to try soy products in the future because they were less concerned with off flavors and interested in the nutritional benefits. The discussion board results are consistent with research that indicates taste has the largest impact on demand for soy products (Chang et al., 2012). Defatted soy flour was used in the muffins because the flavor is less beany and more neutral. The laboratory exercise was likely students' first experience with defatted soy flour, and many students were surprised at the mild flavor of the soy muffins. As a results, they were more likely to try soy products in the future.

Table 3.4—Online soybean discussion board questions and summarized responses from online students

Question	Summary of Responses	Selected Response
How has your perception of soybeans or soy as an ingredient changed?	Increased awareness of end uses Improved perception of taste	“Before the soy lecture and muffin lab, I had a negative connotation whenever I heard of soy because I knew tofu was made from soy, which I am not a fan of, but so many great things come from soy including soy sauce, tempeh, soy milk, and many meat alternative uses.”
What benefits do you see for consuming soybeans and soy products?	Good source of protein	“Soy is a high-quality protein and contains all the essential amino acids like those found in meat. For vegetarians this is very beneficial.”
Are there any barriers for you personally consuming soy?	Concerns about effects of estrogen consumption, cost	“The reason I've had a negative connotation of soy is because I've been told it has a negative effect on estrogen levels which I decided to

look into after this lecture. Soy has high isoflavone levels which historically have been thought to decrease estrogen levels in the body. However recent research has shown that the levels one would need to consume to have a negative effect on one's estrogen levels would be extremely high and therefore this is not a risk for most people...”

Many students indicated that the exercise removed barriers to future soy consumption such as lack of familiarity with the product and preparation, however, a few students listed concerns about estrogen consumption and possible hormonal effects. Concerns about estrogenic effects present an opportunity for discussion in future implementations of the module. Consumption of phytoestrogens have been found to have benefits for the cardiovascular, immune, and nervous systems and risks that necessitate further research (Petrine & Del Bianco-Borges, 2021).

Student Reflective Survey Results

Demographics

Eighty-nine students in the online course (95% response rate) completed the reflective survey (n=89) (Figure 3.3). One hundred and five students in the on-campus course (95% response rate) completed the reflective survey (n=105). Distribution of grade levels between the two course modes was similar. Juniors and seniors accounted for 71% or more of students in both courses. Seven percent or less of students in either course were freshmen. While 14% of students in the online course were sophomores, the on-campus course contained 26% sophomores.

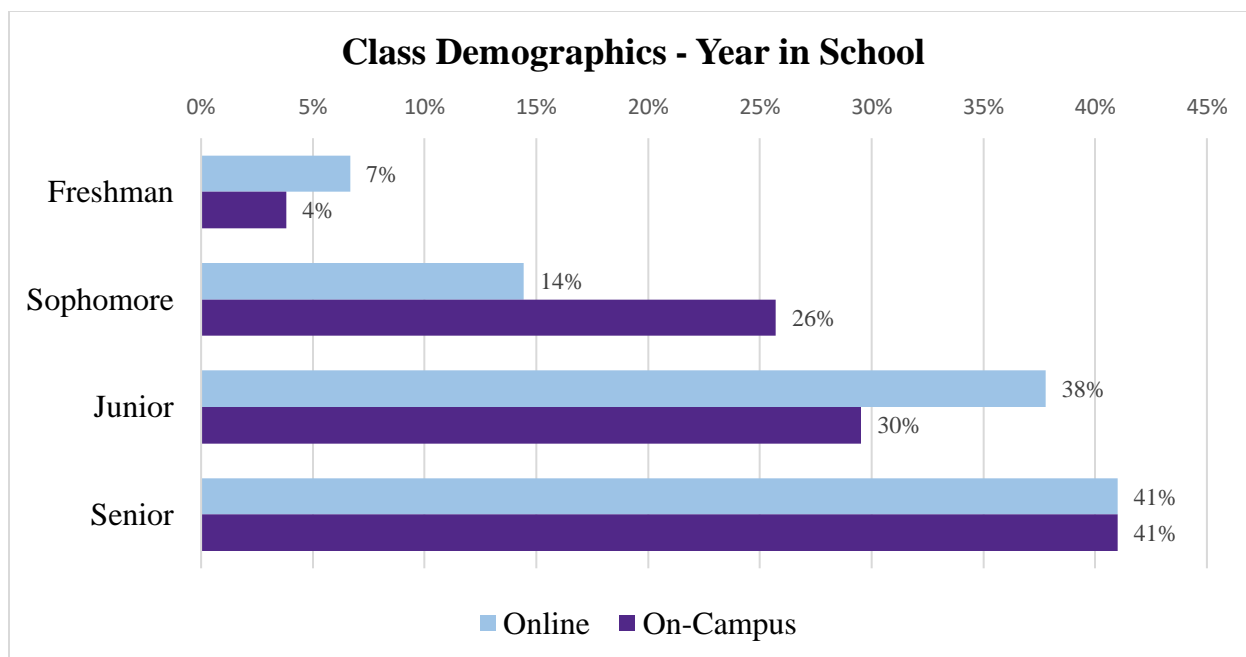


Figure 3.3—Class demographics from reflective Survey: year in school

Distribution of majors was more varied between the two course delivery methods (Figure 3.4). In both courses Animal Sciences & Industry majors were most represented, with 40% online and 55% on-campus. The animal science students were likely juniors and seniors because animal science students typically take the required food processing elective as upperclassmen. For the online course 42% of students were food science majors and 28% of students in the on-campus course were food science majors. The remaining students were Bakery Science & Management majors and other majors. Historically, students in other majors are typically in the College of Agriculture.

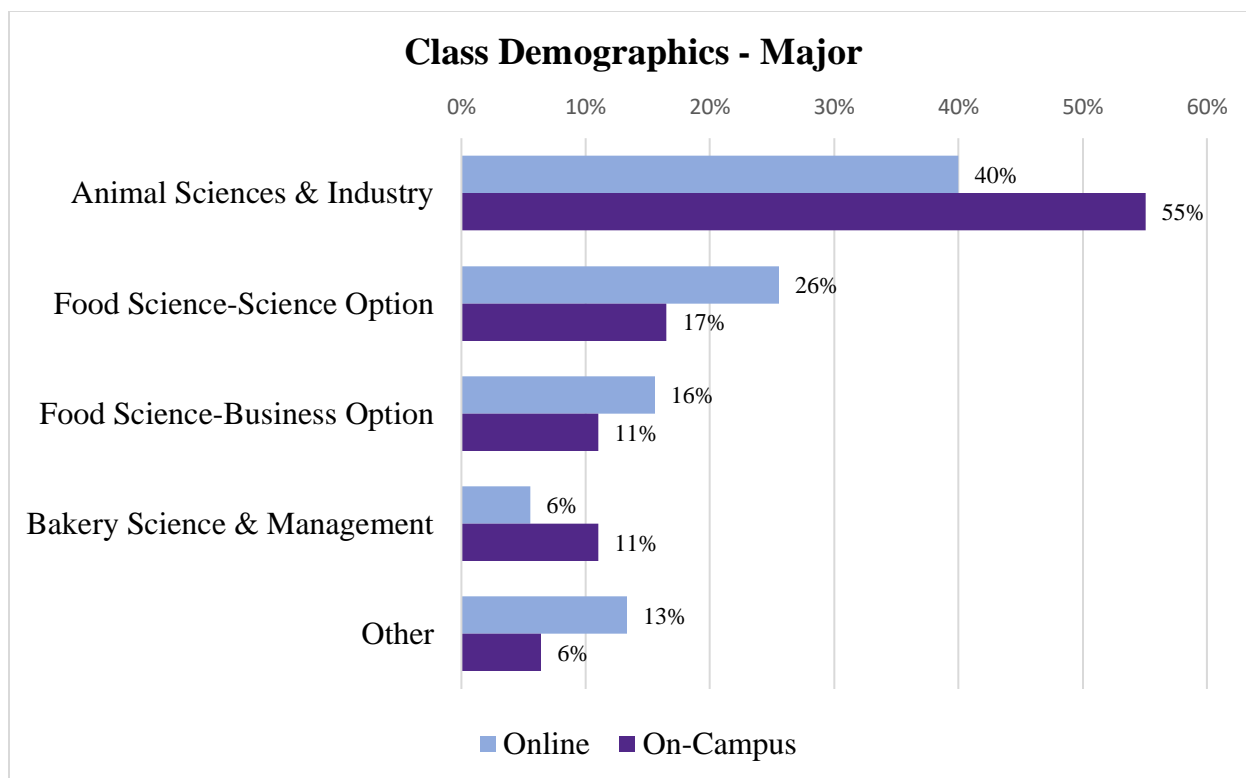


Figure 3.4–Class demographics from reflective Survey: major

Muffin Physical and Sensory Properties

A majority of responses in the “Muffin Physical and Sensory Properties” section was positive for both online and on-campus courses (Figure 3.5). The “Muffin Physical and Sensory Properties” section features questions about the physical and organoleptic properties of muffins and how these properties are affected by the addition of soy flour. All questions in this section received 77% or more responses of “strongly agree” or “somewhat agree” for both course delivery methods. At least 92% of online and on-campus respondents strongly or somewhat agreed with questions four, five, and seven. Question six received the most negative responses in both the online and on-campus courses with 9% and 8% negative responses respectively. Question five asked if the exercise introduced respondents to standard preparation procedures for muffins. Students may have responded negatively to this question if they had prior experience with muffin preparation and the exercise was not an introduction. For students already familiar with basic muffin preparation, the module expanded on prior knowledge by including commercial item descriptions given by the USDA’s AMS and including a video of a commercial muffin production line. Additionally, the muffin scorecard was introduced as a quality and

sensory evaluation method. For both course delivery methods, over 96% of students indicated that this was their first experience using a muffin scorecard. Overall, students indicated that the exercise improved their understanding of muffin physical and organoleptic properties with and without added soy.

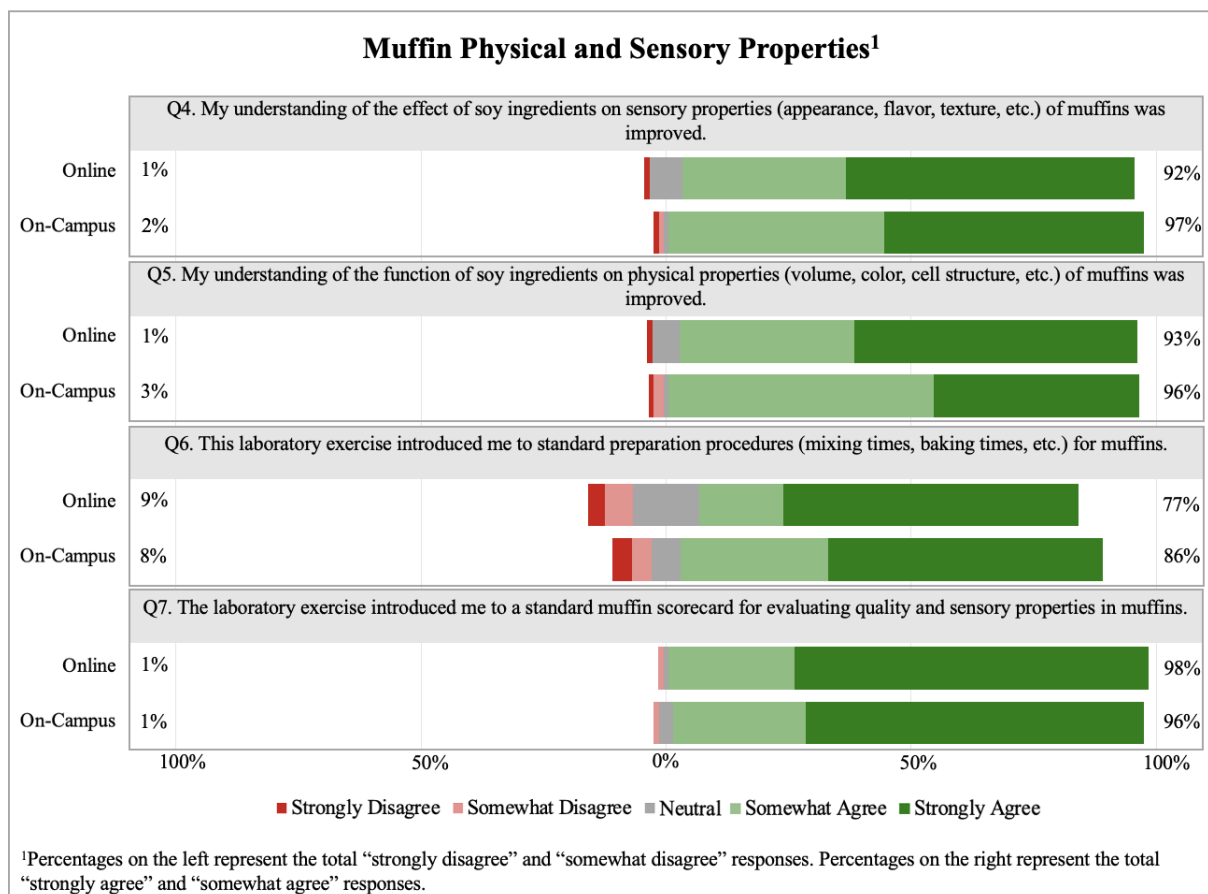


Figure 3.5–Student responses for “Muffin Physical and Sensory Properties” reflective survey section

Responses were similar for online and on-campus courses. Differences between courses ranged from 2-9%. On-campus students responded more positively overall for each question; however, responses in both course delivery methods were very positive. The efficacy of the online module at teaching about muffin physical and sensory properties was similar to the on-campus course, and both were well received by students.

Active learning, where students are engaged in the learning process as opposed to passively listening, improves understanding and retention of course material (Hollis & Eren,

2016). The online laboratory exercise successfully created an active learning opportunity similar to an on-campus laboratory exercise by allowing students to carry out every step of the baking process. Students scored the final product based on physical (texture, color, shape) and sensory properties (taste, smell, mouthfeel). An immersive experience such as the muffin laboratory adds value to the online course.

Research and Writing Skills

Students' responses in the "Research and Writing Skills" section varied more than the other sections. Questions in this section received between 44% and 97% positive responses overall (Figure 3.6). The question with the least positive responses was question nine, which asks respondents if the exercise introduced them to abstract-writing for the first time. Negative responses to this question are expected, as 71% or more of the respondents in both course delivery methods were juniors or seniors who may have read or written abstracts in previous courses; however, 79% of online respondents and 65% of on-campus respondents indicated that participation in the exercise improved their abstract writing skills. The positive responses to this question indicate that students found the abstract writing portion of the exercise valuable, regardless of prior experience writing abstracts. Furthermore, 97% and 92% on online and on-campus students, respectively, indicated that their ability to communicate scientific data was improved by participation in the exercise. The scientific abstract assignment was the main method used by students to communicate data. Fewer positive responses to the abstract writing questions as opposed to the scientific writing question could be attributed to a lack of confidence from the students in their abstract-writing skills. Regardless of self-reported improvement, 72% or more of students in the online course met or exceeded expectations in all rubric categories of the abstract assignment.

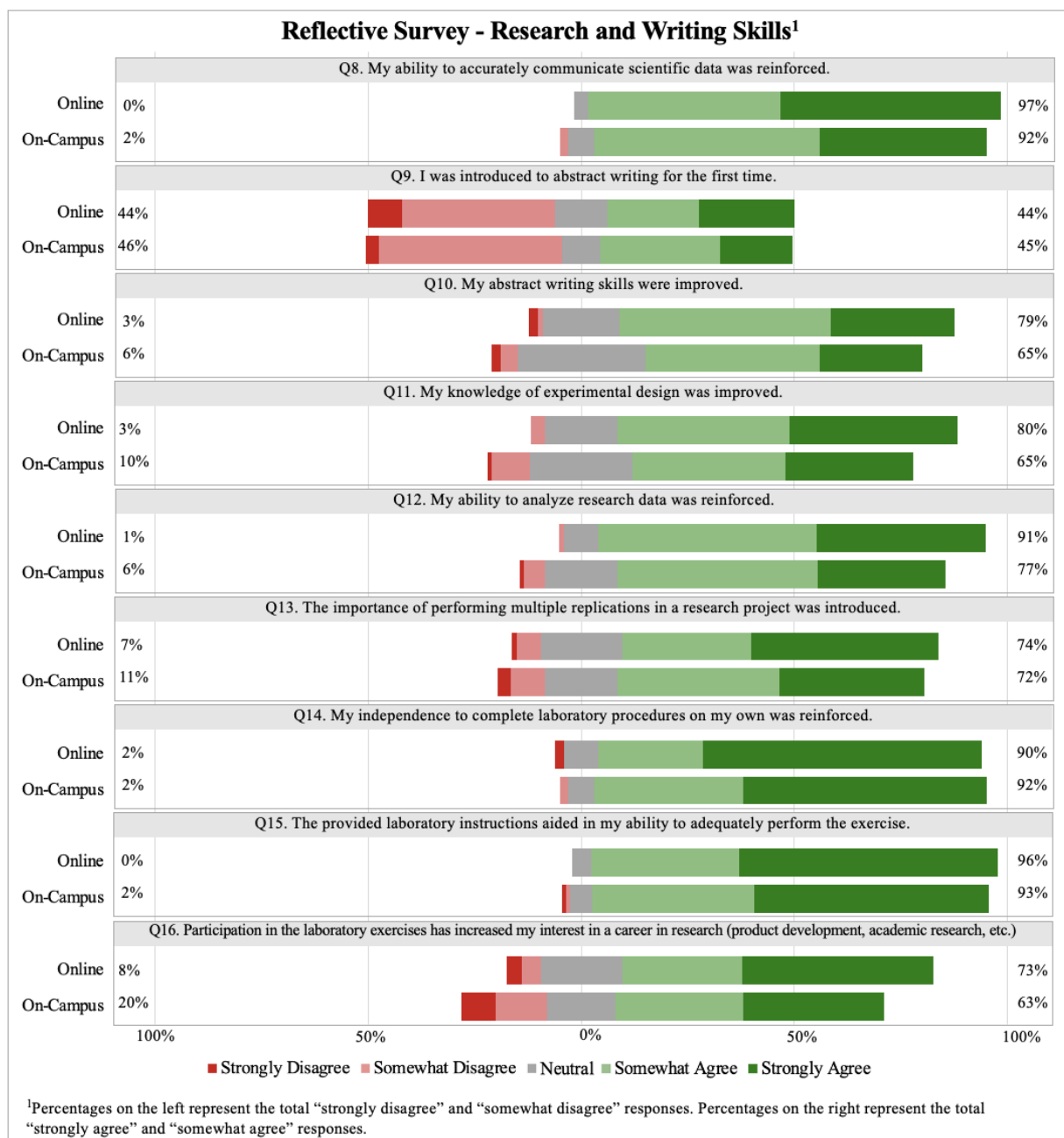


Figure 3.6—Student responses for “Research and Writing Skills” reflective survey section

A key difference between the online and on-campus course is the ability of instructors to guide students through the laboratory. In on-campus laboratory activities, the instructors typically provide verbal instructions alongside written instructions to enhance students’ experience. If students are unsure about a step, instructors are present to help. Online courses lack these opportunities for instructors to aid students, so thorough written instructions are

essential. The instructions must guide students through the activity, providing adequate information without overwhelming students. Ninety-six percent of students in the online course responded that the provided instructions were useful in completing the exercise, compared to 93% of on-campus students. The provided laboratory instructions successfully allowed students to complete the laboratory exercise, whether instructors were present to provide additional guidance or not.

Food Processing Knowledge

Questions in this section asked about how the module impacted students understanding of food processing principles and applications with 80% or more positive responses in both course formats (Figure 3.7). Students in both courses agreed or strongly agreed (90%) that the exercise helped apply principles of food science to practical issues associated with food processing. Online courses typically lack opportunities to practice applying knowledge, and survey results indicate that activities such as the soy muffin laboratory exercise provide these essential opportunities (Hollis & Eren, 2016). Ninety-four percent or more students in both courses agreed or strongly agreed that the exercise increased understanding of the advantages and disadvantages of adding soy ingredients to food. Survey results are consistent with responses to the discussion board questions, where many students were surprised at the possible uses of soy products in food and curious about including soy into their diet more frequently. The discussion board questions were likely useful in the module because understanding and retention of course material increases when students relate learning to their personal life (Schmidt, 2020).

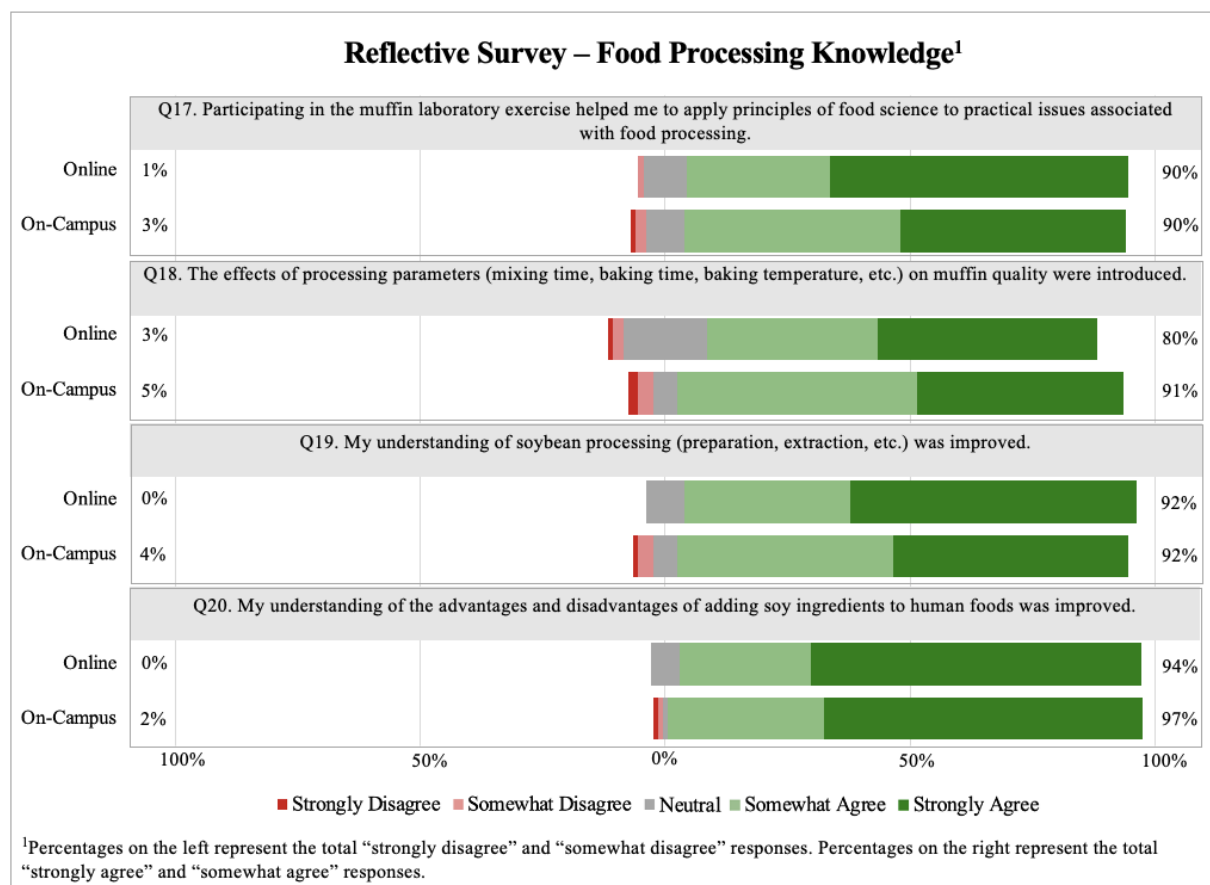


Figure 3.7–Student responses for “Food Processing Knowledge” reflective survey section

Conclusion

Data from the student abstract scores, discussion board comments, and reflective survey results indicates that the module was effective in teaching students about soybean products processing and end uses. At the completion of the module, students were more aware of the advantages and disadvantages of adding soy ingredients to foods and were more open to trying soy products in the future. Students were exposed to the research process and communicated results in a scientific abstract. The laboratory exercise encouraged students to apply basic knowledge of food science principles to explore solutions to industry challenges. The soybean product processing module serves as a model for future online modules with hands-on activities to improve content knowledge and skills acquisition in food science and agriculture courses.

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Chapter 4 - Development and Evaluation of Module with Hands-On Component for Secondary Classrooms

Introduction

When students are interested and engaged in course content, understanding and retention of course material increases. In a seven-year study of factors impacting students' attitudes towards lab, excitement was found to have the greatest impact of all factors measured, including time efficiency, lack of difficulty, ease of understanding, relevance to lecture, open-endedness, and amount of experimenting (Basey et al., 2008). One way to engage students is to involve them in practical applications of course material. In problem-based learning, students are invited to work on developing solutions to real problems industry members face while working in the field. Problem-based learning has been shown to support better understanding and utilization of course concepts (Pease & Kuhn, 2011).

In the field of food science, consumers are becoming more educated about the benefits of protein (Sloan, 2020). As a result, food companies have the opportunity to create products that meet the increasing demand for high-protein foods. In particular, high-protein snacks are beneficial for individuals with busy lifestyles who do not have time for a full meal. In 2021, the total value of the protein bar market was \$4.68 billion and is projected to exceed \$7 billion by 2029 (Fortune Business Insights, 2022). Despite the nutritional value of protein bars, taste is a barrier to consumption for many consumers (Thakur et al., 2022). Fortifying popular snacks with protein may be a way for food companies to meet market demand for high-protein snacks with desirable sensory properties.

A relevant and interesting problem such as the demand for high-protein snacks provides secondary food, nutrition, and agriculture classes with an opportunity to implement problem-based learning. Teachers invite students to use higher order cognitive skills by testing formulas and scoring finished products in the development of high-protein snacks. The objective of this study was to design and evaluate a module for secondary students utilizing soy protein in muffins as a hands-on problem-based learning experience.

Methods

A module over baking with soy protein was modified from a soybean products processing module developed for the Fundamentals of Food Processing Course at Kansas State

University. The authors Getty and Brown partnered with a local high school (Manhattan, KS) to conduct a preliminary test of the module. Further modifications were made based on observations from the preliminary test. The baking with soy protein module was distributed to a group of Kansas teachers for an independent teacher-based trial along with a module evaluation survey.

Preliminary Module Test

In the Spring of 2021, the module preliminary test was conducted at a local high school in the Baking and Pastry I and II classes. Module components included a lecture with background information and laboratory exercise preparing muffins with soy protein. Due to class scheduling, the hands-on laboratory activity was completed first during a 90-minute class period and the lecture was given later in the week during a 50-minute class period. The authors lead both the laboratory activity and the lecture to observe module effectiveness directly. While students baked, the authors supervised students and answered questions as needed. Students scored muffin physical and sensory characteristics using a muffin scorecard, and authors were available to assist students in evaluating muffins as the scorecard contained attributes new to students.

Because secondary students typically lack laboratory experience, the soy muffin laboratory was designed to support students in making unbiased observations that are crucial for scientific experiments. Students were instructed to prepare six muffins of their assigned treatment (control-no soy, low soy, high soy) without inclusions. The soy muffins were compared to the control muffins in order to isolate the effects of soy on the physical and sensory properties of muffins. The muffins were scored using the muffin scorecard. After six muffins were scooped from the group's assigned treatment batter, students then were encouraged to add desired inclusions (frozen fruits and chocolate chips) to the remaining batter. Providing instructionally incidental choices to students has been shown to significantly ($p < 0.05$) increase intrinsic motivation, task involvement, and perceived confidence (Cordova & Lepper, 1996).

The next class period was a 50-minute class, and the lesson was designed to span about 40 minutes. A lecture covering soy products, processing, and nutrition was presented with smart phone interactive slides using Mentimeter (Stockholm, Sweden) to increase engagement. Figure 4.1 is an example of the poll given at the beginning of the lecture.



Figure 4.1–Poll from in-person preliminary lecture

Students were asked if they would try an Impossible Burger from Burger King, a meatless burger made from soy. Poll type responses allow all students to respond while minimizing peer pressure and social anxiety (Price, 2021). More outgoing students were willing to share the reasoning behind their answers allowed for a short class discussion of the benefits and drawbacks of plant-based meats to lead into the next lecture topic.

Best practices in classroom polls indicate having a variety of question types is beneficial for student engagement (Price, 2021). Another type of interactive slide used was an open-ended question asking students to name products featuring soy. The screen was frozen while students submitted responses and then unfrozen to show a word cloud generated by student responses (Figure 4.2). Frequency of word submission is represented by font size, with words submitted most frequently shown in the largest font size.



Figure 4.2–Question from in-person preliminary test lecture

Interactive slides were also used to share individual opinions about muffins with the class (Figure 4.3). A sliding bar was used for each question for more variety in interactions with students' smartphones. The last question used humor to engage students in a discussion on tradeoffs for nutrition and product quality. Overall, the module was positively received, and the authors spoke personally with many students about their interest in baking and food science.

Overall Muffin Acceptability

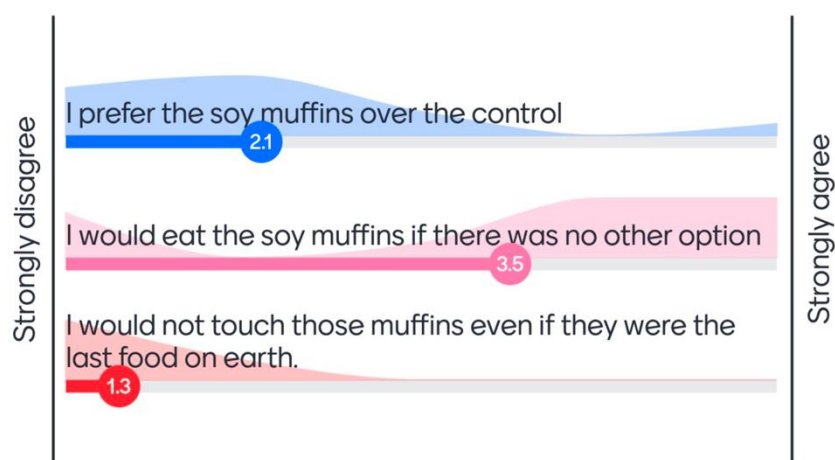


Figure 4.3–Interactive slide results for overall acceptability of soy muffins

Changes Made Before Teacher-Based Trial

Four changes were made to the module before teacher-based trials due to the authors' experiences from the preliminary testing.

1. Interactive slides were created using Slido rather than Mentimeter.
2. Lecture content was adjusted to allow high school teachers with less specialized knowledge give the lesson. Teacher notes were added to each slide.
3. Pictures with examples of muffin characteristics were included in slides.
4. Research questions were added at the end of lecture to prepare students for the laboratory activity.

Slides were edited for use on Slido (Webex, San Jose, CA), which is similar to Mentimeter as a smart-phone interactive slide system. Slido is available as a Google add-on and the free version allows 3 free interactive slides, while Mentimeter only allows 2 free interactive slides. Additionally, non-interactive slides were created via PowerPoint (Microsoft, Redmond, WA) with identical question slides for teachers who did not wish to incorporate smart phones into the classroom. Teachers were encouraged to lead class discussions over the content instead.

Lecture content in the preliminary module test covering soybean processing, products, and nutrition was highly specific and consequently above level for high school students. Relating lecture material to the laboratory exercise was also difficult as the connection required more information. A revised lecture was designed to allow food, nutrition, and agriculture teachers to reach high school students at an appropriate level without extensive subject-specific background knowledge. Speaker notes were added to each slide to aid teachers in presenting the information and a supporting information document was provided (Appendix B1).

In the preliminary testing of the module, the authors were present to assist students with scoring muffin attributes, however, teachers may not be familiar with terms used in evaluation of baked goods. Pictures of muffin characteristics such as texture (soft versus tough) and crust contour (well-rounded versus very pointed) were included in lecture slides for use during evaluation of muffin attributes. The muffin scorecard is still useful as a learning opportunity, and appropriate information about muffin scoring with example photos provided should help both teachers and students in understanding attributes.

Slides were added to the end of the lecture to introduce the laboratory exercise. Research questions were introduced before students baked muffins so students could make relevant observations during the exercise. Students answered research questions as part of the post-lab assignment.

Teacher-Based Trial

Module Design

At the beginning of the module, students were given a lecture with background information necessary to understand the laboratory exercise. Teachers were given two lecture slide formats. The first presentation used a Google add-on feature “Slido” to include interactive questions that students responded to using their devices. The second lecture slide was a normal PowerPoint presentation with identical slides that featured appropriate moments for teachers to engage verbally with students (Appendix B2). The lecture included details about grain foods and their nutritional value, protein foods and their nutritional value, and background information on soybeans and the products that can be made from soybeans.

After completing the lecture, students completed a hands-on laboratory exercise where they baked muffins with three levels of soy protein powder and evaluated muffin nutritional, physical, and sensory properties. Students, individually or in groups, prepared three muffin variations: a control with no soy protein, a low soy protein muffin with 10% soy protein, and a high soy protein with 20% soy protein. Complete laboratory handout instructions are available in Appendix B3 and Appendix B4. Group size and number of muffin variations prepared by each group was determined by the classroom teacher to accommodate various class sizes and times. Groups that prepared only one muffin variation were instructed to trade muffins so groups could evaluate all variations. Students measured muffin physical qualities using available kitchen equipment. All classrooms measured the muffin height of each muffin using a standard 12-inch ruler and calculated an average muffin height for each muffin variation. Classrooms that had access to kitchen scales also measured muffin weight and calculated an average muffin weight for each of muffin variation. Muffin sensory characteristics were evaluated using a muffin scorecard provided in the laboratory instructions adapted from *Foods: experimental perspectives* (McWilliams, 2001). The scorecard was divided into two sections, external qualities (volume, surface contour, crust color) and internal qualities (crumb color, cell uniformity and size, cell wall thickness, texture, flavor, and aftertaste), and included numerical scores associated with qualitative observations.

Students were assigned post-lab questions to guide reflection on the results. Students are asked to summarize the results, apply their knowledge, and make inferences. Because inquiry-based labs promote higher-order thinking, they can be frustrating to students. Using reflective questions after the laboratory activity is a deliberate design choice to reduce frustration and negative attitudes towards the activity (Basey et al., 2008; Cox & Junkin, 2002). Multiple choice quiz questions and answers based on the lecture and laboratory were provided for teachers to use,

as well as a short essay question. Quiz and essay questions are available in Appendix B5. Table 4.1 lists selected National Standards for Family and Consumer Science (FCS) and Agriculture, Food, and Natural Resources (AFNR) career cluster summarizes how each standard is addressed in the module.

Table 4.1–National standards for Family and Consumer Science (FCS); Agriculture, Food, and Natural Resources

FCS Standards	AFNR Standards	How standard is applied
8.2.4 Use the Hazard Analysis Critical Control Point (HACCP) and crisis management principles and procedures during food handling processes to minimize the risks of foodborne illness.	FPP.01.02. Apply food safety and sanitation procedures in the handling and processing of food products to ensure food quality.	Minimum time and temperature needed to kill <i>Salmonella</i> referenced in baking instructions.
8.2.5 Practice standard personal hygiene and wellness procedures.	FPP.01.02. Apply food safety and sanitation procedures in the handling and processing of food products to ensure food quality.	Students should follow GMP for handwashing and proper handling of food products at all stages of preparation.
8.5.1 Demonstrate professional skills in safe handling of knives, tools, and equipment.	FPP.01.01. Analyze and manage operational and safety procedures in food products and processing facilities.	Students will utilize a variety of equipment including mixers, scoops, and ovens during muffin preparation.
8.5.2 Demonstrate professional skill for a variety of cooking methods including roasting, broiling, smoking, grilling, sautéing, pan frying, deep frying, braising, stewing, poaching, steaming, and baking using professional	FPP.03.02. Design and apply techniques of food processing, preservation, packaging and presentation for distribution and consumption of food products.	Students will practice skills such as measuring, mixing, and baking.

equipment and current technologies.

8.5.4 Apply the fundamentals of time, temperature, and cooking methods to cooking, cooling, reheating, and holding of a variety of foods.	FPP.01.02. Apply food safety and sanitation procedures in the handling and processing of food products to ensure food quality.	Appropriate internal temperature for muffins is referenced in baking instructions
8.5.10 Prepare breads, baked goods and desserts using safe handling and professional preparation techniques.	FPP.03.02. Design and apply techniques of food processing, preservation, packaging and presentation for distribution and consumption of food products.	Students prepare muffins as a food lab.
8.5.14 Demonstrate cooking methods that increase nutritional value, lower calorie and fat content, and utilize herbs and spices to enhance flavor.	FPP.02.03. Apply principles of human behavior to develop food products to provide a safe, wholesome and nutritious food supply for local and global food systems.	The focus of the laboratory activity is increasing muffin protein while accounting for muffin acceptability.
9.5.6 Conduct sensory evaluations of food products.	FPP.03.01. implement selection, evaluation and inspection techniques to ensure safe and quality food products.	Students evaluate all muffin variations using a muffin scorecard and reference photos
9.7.5 Relate the composition of lipids and proteins to their functions in foods and their impact on food preparation and nutrition.	FPP.02.01. Apply principles of nutrition and biology to develop food products that provide a safe, wholesome, and nutritious food supply for local and global food systems.	Students learn about the importance of protein and carbohydrates in a balanced diet in a lecture and during the food lab.

Recruitment

This study was reviewed and approved by the Institutional Review Board (IRB) at Kansas State University under proposal number 10709 (Appendix B6). In the Fall of 2021, Future Farmer's of America (FFA) and Family and Consumer Science (FCS) secondary education teachers in Kansas were invited to test the module. FFA teachers were contacted by email via a Kansas Agriculture Teachers email list and FCS teachers were contacted via a Facebook page for Kansas FACS teachers. A brief study description was provided along with a form link for teachers to indicate interest and provide contact information. Twenty-six teachers completed the initial form and were contacted by email to confirm interest and provide details about access to kitchen equipment and quantity of specialty ingredients requested. In the Spring of 2022, digital course materials were emailed to 25 interested teachers and specialty ingredients were mailed in the quantities requested. An anonymous module evaluation survey was emailed to the 25 teachers who received materials. The informed consent and debriefing statements are available in Appendix B7 and B8. The last question in the module evaluation survey invited interested participants to enter in a raffle for an Amazon gift card.

Survey Design

A survey was created and distributed to participating teachers using Qualtrics (Provo, UT; Seattle, WA). The full survey can be found in Appendix B9. The first section of the survey gathered information on the classes that participated in the module and what materials each teacher utilized. The second section used statements with a five-point Likert scale ranging from 1 (strongly agree) to 5 (strongly disagree) to gather feedback on the module's effectiveness and ease of use. After completing the survey, respondents interested in participating in a raffle for an Amazon gift card were directed to a second survey to preserve anonymity of responses.

Results

Survey Responses—General Information

Of the 25 teachers who received materials, four teachers completed the module evaluation survey for a response rate of 16%. One response was removed from the dataset because the responder indicated the module was not completed in the class due to a lack of extra ingredients. All three teachers used the module in a Nutrition and Wellness class. Two sections per teacher were taught for a total of six sections. Grades nine through 12 were represented with a total of 49 students.

Survey Responses–Evaluation of Module

The three remaining teacher responses to the Likert survey questions are summarized in Table 4.2. Questions addressed ease of use, difficulty of material, quality of instructional materials, and students’ interest. Overall, the respondents agreed or strongly agreed with every question, which indicates the module was positively received by respondents.

Table 4.2–Likert scale responses evaluating module effectiveness

Question	Strongly			Strongly	
	Disagree	Disagree	Neutral	Agree	Agree
Q1 I was able to use the module in my classroom with minimal additional preparation.	0	0	0	2	1
Q2 The provided laboratory instructions were clear and helpful for students performing the exercise.	0	0	0	2	1
Q3 The module was on-level for the students experience with lab/kitchen skills.	0	0	0	1	2
Q4 The muffin scorecard was a useful method for students to evaluate muffin quality.	0	0	0	1	2
Q5 The laboratory exercise increased students’ knowledge about soybean products.	0	0	0	1	2
Q6 The module engaged students.	0	0	0	2	1
Q7 The laboratory exercise was effective for teaching	0	0	0	1	2

students about the advantages
of and barriers to adding soy
ingredients to foods.

Discussion

Methods for improving study recruitment

A limitation of survey results collected is the small pool of participants (25) and low response rate (16%). Nonresponse bias is bias in survey responses due to the respondent samples differing in some way from nonrespondent samples (Berg, 2005). While survey responses about the module were positive overall, the module may not have been positively received by nonresponding teachers. Increasing both number of study participants and response rate to the module evaluation survey would provide more information about the module's effectiveness and modifications for improvement.

Low recruitment numbers can be attributed to ineffective recruitment methods and lack of interest and/or time from contacted teachers. Recruitment methods may be improved by diversifying distribution of study information. Only two methods were used to recruit teachers for the teacher-based trial: one post in a Kansas Family and Consumer Science Facebook group for teachers and one email to a Kansas Agriculture teacher email list. Both methods could be repeated with reminders about recruitment for the study. Additionally, other contact methods could be used. Kansas secondary teachers in all relevant subjects could be emailed recruitment information at email addresses listed on district websites.

Providing more attractive incentives for participation may also improve study participation. In a study on recruitment of educators for surveys panels, significantly more teachers were recruited with \$10 pre-incentives than with promised incentives. Pre-incentives were provided before recruitment regardless of the teacher's participation in the panel (Robbins et al., 2018). In our study, the only monetary incentive was the chance to win an Amazon gift card. No monetary incentive was guaranteed to any participant. Module materials including digitally available lesson materials and mailed ingredients were viewed as a benefit to reduce workload of teachers for developing class content, however, this may not be a strong enough incentive to increase participation. While pre-incentives are effective at increasing study enrollment, the cost of pre-incentives may be prohibitive. Promised incentives for recruitment as a teacher-based trial participant may increase study recruitment numbers.

Field specific factors may have affected response rates. Recruited teachers may have initially agreed to test the module but later decided to utilize a different lesson plan. Recruited teachers may have implemented the module but failed to complete the module evaluation survey. Educator response rates for surveys without pre-incentives are commonly between 20-30%. Possible reasons for low response rates from educators include disproportionally higher survey requests than other industries, increased workload and time demands, time of contact during school term, and length of time needed to complete surveys (Robbins et al., 2018). The module evaluation survey was designed to be short and easy to complete with seven Likert-type questions and three optional open-response questions. Estimated time to complete survey could be added to survey distribution emails to encourage participants to complete the survey. Multiple survey reminders could be sent at varying times during a school term to increase chances of emailing during a less busy time in the school term.

Conclusion

Overall, the module was well received by the teachers who participated in the survey, although response rate for the survey was low. The study could be repeated utilizing techniques for better recruitment and response rates to better evaluate the module. After evaluating the module and making desired changes, the module could be made available as supplemental material for teachers in relevant fields.

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Chapter 5 - Assessing Functionality of Enzyme-Modified Soy Protein Hydrolysates in Muffins

Introduction

Demand for plant-based protein foods is increasing due to an exponentially growing population and improved understanding of the nutritional value of protein (Alrosan et al., 2022; Henchion et al., 2017). A 2021 Food and Health survey from the International Food Information Council found that 62% of respondents try to consume protein, and 25% of respondents are consuming more plant-based proteins (International Food Information Council, 2021). Therefore, food companies are researching technologies that provide consumers with new high-protein products from plant-based sources. In particular, the market for high-protein baked goods is experiencing growth. Kodiak cakes, a high-protein baking mix company, grew from an annual revenue of \$6.7 million in 2014 to \$160 million in 2019 (Peckenpaugh, 2020).

Soy is a plant-based protein that is attractive for such product formulations because it is a complete protein with adequate amounts of all nine essential amino acids, unlike most plant-based sources of protein (Joye, 2019). Soy is often included in bakery formulations as soy flour (50-65% protein), soy protein concentrate (65-90% protein), or soy protein isolate (SPI) (>90% protein) (FAO & WHO, 2019). As an ingredient in baked goods, soy protein poses challenges with respect to taste and functionality (Beacom et al., 2021). Sung et al. (2006) tested addition of 3-24% soy protein concentrate to sponge cakes and found decreased batter viscosity, decreased cake volume, and increased cake hardness with increasing protein content. Majzoobi et al. (2014) tested addition of 5-30% soy protein isolate to sponge cake and found increased batter consistency, increased volume, and increased hardness with increasing protein levels. Addition of soy protein isolate up to 20% resulted in acceptable sensory scores (taste, color, texture, overall acceptability), however, addition of 30% soy protein isolate decreased sensory ratings ($p < 0.05$).

In general, the addition of protein to baked goods can cause undesirable changes to physical and organoleptic properties, so modifications are being explored to improve baking quality. Enzymatic hydrolysis is a method for breaking down proteins into smaller peptides and amino acids using proteases. Meinschmidt et al. tested the effects of hydrolysis by six proteases (Alcalase®, Corolase® 7089, Corolase® 2TS, Flavourzyme®, papain, and pepsin) on functional and sensory properties of soy protein hydrolysates. For all proteases tested, protein solubility increased ($p < 0.05$). Results varied depending on the specific enzyme, however, hydrolysis of SPI

in general resulted in increased emulsifying and foaming capacity but decreased foaming stability. In addition to changed functional properties, hydrolysis induces sensory changes. All hydrolysates were rated as having less of a “beany” aroma while Alcalase® and pepsin enzymes increased the bitter and sour tastes, respectively. Sensory evaluation of papain and Flavourzyme® hydrolysates indicate bitterness remained the same or decreased. Soy protein hydrolysates, especially papain and Flavourzyme®, show promising results for improving physical and organoleptic properties in model systems, however little research exists to explore functional properties of soy hydrolysates in food systems. The objective of this study was to evaluate the effect of soy protein hydrolysates on muffin physical properties.

Materials and Methods

Experimental Design

Two soy protein hydrolysates were prepared, using either Flavourzyme® or papain enzymes. Hydrolysates were analyzed for effect of hydrolysis using Sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) and Degree of Hydrolysis (DH). Seven muffin treatments were prepared in a 2×3 factorial design with protein level having two levels (10%, 20%) and protein type having three levels (Soy protein isolate, Flavourzyme® soy hydrolysate, papain soy hydrolysate) plus a control muffin with 0% protein (Figure 5.1). Batter and muffin physical properties were measured to compare the effect of treatments.

Muffin Treatments				
Protein Level	Protein Type			
	Soy Protein Isolate	Flavourzyme Soy Hydrolysate	Papain Soy Hydrolysate	
10	SPI10	FSH10	PSH10	C
20	SPI20	FSH20	PSH20	
				Control: 0% Protein

Figure 5.1–Seven treatments and factorial structure

Materials

Soy protein isolate (PurelyNature™ ProFam® 974) with a protein content of 93.5% was kindly donated by ADM (Chicago, IL, USA). Flavourzyme® was kindly donated by Novozymes (Franklinton, NC, USA; Bagsværd, Denmark). Papain (E.C. 3.4.22.2, Sigma No. P3375) was purchased from Sigma-Aldrich (St. Louis, MO, USA). All-purpose flour (Great Value), cornstarch (Great Value), granulated sugar (Great Value), double-acting baking powder (Clabbergirl), salt (Great Value), nonfat dry milk (Great Value), dry whole eggs (Judee's, Plain City, Ohio, USA), and shortening (Great Value) were purchased from a local grocery store (Manhattan, KS, USA). Water used to prepare muffins was city water from the tap (Manhattan, KS, USA).

Equipment to prepare muffins included disposable weigh boats, rubber spatulas, portable balances (Ohaus, Parsippany, NJ), stand mixers with flat paddle attachment (KitchenAid Artisan, Model No. KSM150PSWH, St. Joseph, MI, USA), 1.5 tablespoon cookie scoop, household baking oven, 12-cup non-stick muffin pans from Chicago Metallic (Bundy Baking Solutions, Urbana, Ohio), and cooling racks.

Preparation of Hydrolysates

Hydrolysis conditions were selected according to Meinlschmidt et al. (2015) and are listed in Table 5.1. To prepare the hydrolysates, 5% SPI suspension in distilled water was prepared (w/v, protein basis) and the pH adjusted to optimum level. The suspension was heated in a water bath shaker at optimal temperature for 15 min to allow the suspension to reach optimum temperature. Enzyme was added at the selected enzyme to substrate ratio and the mixture was returned to the water bath shaker for 30 minutes. At the end of the reaction the mixture was boiled for 15 min to deactivate the enzyme. After enzyme deactivation, the mixture was allowed to cool down to room temperature, lyophilized, and stored at room temperature for further use.

Table 5.1–Hydrolysis Conditions as given by Meinlschmidt et al. (2015)

Protease	E/S (%)	Temperature (°C)	pH value	Reaction Time (min)
Flavourzyme®	0.5	50	6	30
Papain	0.2	80	7	30

Degree of Hydrolysis

DH was determined based on the reaction of primary amino group with o-phthaldialdehyde (OPA) reagent according to the method of Nielsen and Damdbmann (2001). The OPA reagent was prepared as follows: (1) 3.81 g di-Na-tetraborate decahydrate ($\text{Na}_2\text{B}_4\text{O}_7$) and 100 mg Na-dodecyl-sulfate (SDS) were dissolved completely in 60 mL DI water (solvent A); (2) 80 mg OPA was dissolved in 2 mL ethanol and then added to the solvent A (solvent B); (3) 88 mg dithiothreitol (DTT) was added to the solvent B, and total volume was adjusted to 100 mL with DI water. Samples (1.2 mg/mL) were shaking for 2h and centrifuged before using. To measure the DH, 400 μL supernatant of each sample was mixed with 3 mL of OPA reagent and stored at room temperature for exactly 2 min. The absorbance of the mixture was then read immediately at 340 nm with a spectrophotometer (VWR UV-6300PC, Radnor, PA, USA). The DI water was used as blank, and serine standard solution (0.9516 mM) was prepared as standard. DH was calculated according to the method of Nielsen and Damdbmann (2001).

Sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE)

One hundred mg of each sample were suspended in 20 mL PBS (pH=6.8) buffer containing 2% w/v SDS. The mixtures were shaken for 1h (250 rpm) at room temperature and then centrifuged for 5 min at 8000 g. The supernatant was collected and subjected to SDS-PAGE analysis under reducing and non-reducing conditions.

For non-reducing conditions, an aliquot (60 μ L) of each extract was mixed with 20 μ L of 4 x Laemmli buffer (Bio-Rad Laboratories, Inc., Hercules, CA, USA) and heated for 5 min in boiling water. Then, 10 μ L of each mixture was loaded into wells of a 4-20% Mini-Protean TGX gel (Bio-Rad Laboratories, Inc., Hercules, CA, USA) and separated at 200 V for about 35 min at room temperature. A Precision Plus Protein Dual Color Standards (Bio-Rad Laboratories, Inc., Hercules, CA, USA) was loaded (5 μ L) parallelly to monitor the molecular weight. The gel was then stained with Brilliant Blue R Concentrate (Sigma, St. Louis, MO, USA) for 10 min with gentle shaking. Thereafter, the gel was de-stained with DI water repeatedly until the background was clear.

Muffin Batter Preparation

Seven muffin treatments were prepared (Figure 5.1). Muffin batter was prepared according to the formula in Table 5.2. All-purpose flour, cornstarch, non-fat dry milk dry whole eggs, shortening, baking powder, and salt were added to the bowl of a KitchenAid Mixer and mixed on speed 1 for 60s. Half of the water was added and mixed for 30s on speed 1. The sides of the bowl were scraped and the batter was mixed for 30s on speed 1. The remaining water and vanilla were added to the bowl and mixed for 120s on speed 3.

Batter was immediately deposited into a tared muffin tin using three level scoops in a 1.5 tablespoon cookie scoop. Muffin tin with batter was weighed to obtain an initial batter weight. Muffins were baked for 23 minutes in a 375F oven. After baking, muffins were allowed to cool for at least 23 minutes in the muffin tins before transferring to a cooling rack for at least 60 minutes. Muffins were analyzed no later than 3 hours after cooling. Three true muffin replications were prepared for each muffin treatment (n=3). Replications were blocked by day.

Table 5.2–Formulations for muffin treatments

Ingredient	Flour Weight Basis (%)		
	0% Protein	10% Protein	20% Protein

All-Purpose Flour	100	90	80
Cornstarch	5	5	5
Granulated sugar	50	50	50
Baking powder	3	3	3
Salt	2	2	2
Non-fat dry milk	12	12	12
Whole dry egg	8	8	8
Shortening	50	50	50
Water	160	160	160
Protein Variation ¹	0	10	20

¹Protein variations include unhydrolyzed soy protein isolate, papain soy hydrolysate, and Flavourzyme® soy hydrolysate

Batter Specific Gravity

A small cup was filled with water and the weight recorded. The same cup was filled with batter, leveled, and weighed. Specific gravity was taken as the weight of batter divided by the weight of the water (Equation 5.1). Specific gravity measurements were performed in duplicate and averaged for each replication.

$$\text{Specific Gravity} = \frac{(\text{Weight cup with batter})}{(\text{Weight cup with water})} \quad (5.1)$$

Batter pH

Batter pH was measured using a benchtop pH meter (Fisherbrand™ Accumet™ AP110, Fisher Scientific, Pittsburgh, PA, USA) calibrated with standard buffer solutions of pH 4 and 7. Two grams of muffin batter was dispersed in 20mL distilled water and the pH was recorded. Measurement of pH was performed in triplicate and averaged for each replication.

Muffin Height

Muffin height was measured using digital calipers (Tool Shop, Eau Claire WI) to measure the distance from the muffin bottom to the highest peak. Three muffins from were measured and averaged for each replication.

Muffin Imaging

Pictures of top, side, and cross sections of one muffin from each replication were taken with a Canon camera in a light box under 5500K LED lighting.

Muffin Color

A portable HunterLab MiniScan EZ colorimeter (Model No. 4500L, Reston, VA, USA) was used to measure L^* , a^* , and b^* values for muffin crust and crumb color. For the crust color measurement, readings were obtained by placing the colorimeter port on top of an uncut muffin in the center. For the crumb measurement, a muffin was sliced vertically with a serrated knife to obtain a cross-section. Crumb color readings were obtained by placing the colorimeter port in the center of the muffin cross section. Color readings were measured in triplicate and averaged for each replication.

Muffin Texture Analysis

A calibrated TA Texture Analyzer (TA-XT2, Scarsdale, NY) with a 5 kg load cell was used for texture profile analysis (TPA). Muffins were cut into 30mm cubes containing only crumb. Muffin cubes were compressed twice to 40% of the original height using a 3 inch, 10mm tall acrylic cylinder probe (TA-30A) with a pre-test speed of 1mm/s, test speed of 1 mm/s, post-test speed of 2 mm/s, and a 5 second interval between compressions. Three TPA measurements were taken and averaged for each replication.

Muffin Water Activity

Water activity was measured using a Rotronic water activity meter (HC2-AW-USB, Rotronic Instrument Corp., Huntington, NY, USA). Approximately two grams of muffin crumb was placed in 14mm Rotronic plastic sample containers and water activity measured with a dwell time of two minutes. Water activity measurements were performed in duplicate and averaged for each replication.

Statistical Analyses

Three muffin batters were prepared for each treatment ($n=3$). ANOVA was conducted followed by multiple contrasts tests described by Schaarschmidt and Vaas (2009). First, each treatment in the factorial structure was compared to the control to determine whether any of the applied treatments affects the response variables measured (hypothesis 1-6).

$$1. \mu_C - \mu_{SPI10} = 0$$

$$2. \mu_C - \mu_{SPI20} = 0$$

$$3. \mu_C - \mu_{FSH10} = 0$$

$$4. \mu_C - \mu_{FSH20} = 0$$

$$5. \mu_C - \mu_{PSH10} = 0$$

$$6. \mu_C - \mu_{PSH20} = 0$$

Additionally, each protein type was compared holding level constant. Hypotheses 7-9 compare protein types at the 10% level and hypotheses 10-12 compare protein types at the 20% level.

$$7. \mu_{FSH10} - \mu_{SPI10} = 0$$

$$8. \mu_{PSH10} - \mu_{SPI10} = 0$$

$$9. \mu_{PSH10} - \mu_{FSH10} = 0$$

$$10. \mu_{FSH20} - \mu_{SPI20} = 0$$

$$11. \mu_{PSH20} - \mu_{SPI20} = 0$$

$$12. \mu_{PSH20} - \mu_{FSH20} = 0$$

Finally, the effect of protein level was compared holding each protein type constant (hypothesis 13-15).

$$13. \mu_{SPI20} - \mu_{SPI10} = 0$$

$$14. \mu_{FSH20} - \mu_{FSH100} = 0$$

$$15. \mu_{PSH20} - \mu_{PSH10} = 0$$

Tukey's adjusted p-value was used to determine significance of comparisons at $\alpha=0.05$.

Results and Discussion

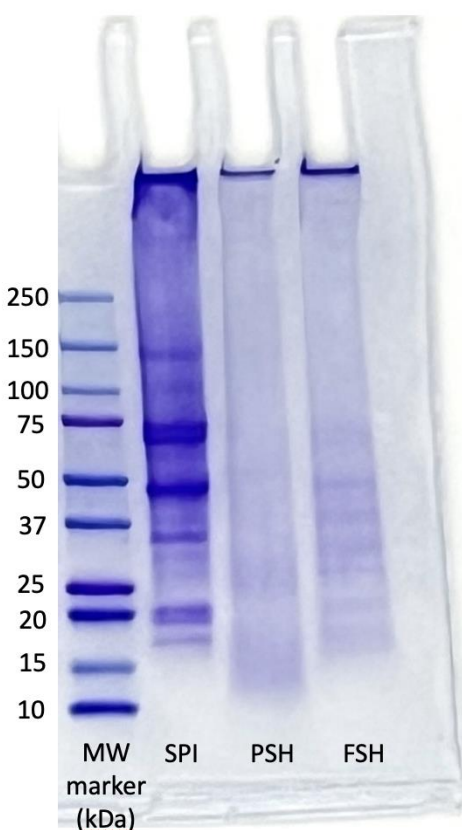


Figure 5.2–SDS-PAGE Profiles of SPI and hydrolysates

The effect of hydrolysis can be observed by gel electrophoresis to determine relative distribution of peptide size. The main protein subunits for soybean proteins are Glycinin with a molecular weight of approximately 360 kDa and β - Conglycinin with molecular weight of 150-180 kDa (Barac et al., 2004). For PSH and FSH hydrolysates, SDS-PAGE bands are very faint (Figure 5.2). No bands are visible at molecular weights above 50 kDa.

Degree of Hydrolysis (DH)

Table 5.3–Degree of Hydrolysis (DH) of soy protein hydrolysates

Protease	DH (%)
Flavourzyme®	4.81 ± 0.07

Another method for determining effect of hydrolysis is Degree of Hydrolysis (DH). Hydrolysates by papain had a higher DH than hydrolysates by Flavourzyme®. These results are consistent with SDS-PAGE results as well as results from Meinschmidt et al. (2015). Using the same reaction conditions, Meinschmidt et al. (2015) found Flavourzyme® hydrolysates reached 6.1% DH after 30 minutes, and papain hydrolysates reached 4.8% DH.

Batter Characteristics

Specific gravity and batter pH were measured as batter characteristics (Table 5.4). Batter pH was not significantly different ($p < 0.05$) for any of the protein treatments compared to the control, and no other comparisons resulted in a significant change in batter pH. Values for batter pH ranged from 6.53–6.72. Solubility of proteins depends on pH. Were et al. (1997) found that solubility of native SPI in model systems is lowest at the isoelectric point, 4.5, and increases from approximately 10% at a pH of 6.0 to approximately 80% at a pH of 8.0. Hydrolysis of soy protein with papain resulted in a significant ($p < 0.05$) increase in solubility from pH 6.0 to 8.0 and was 100% soluble above pH 7.0. Batter pH values were within the range of high solubility for both soy protein isolate and soy protein hydrolysates.

Table 5.4—Means and standard deviation for batter characteristics

Treatment¹	Specific Gravity	Batter pH
C	1.08 ± 0.02	6.72 ± 0.22
SPI10	1.00 ± 0.02	6.58 ± 0.14
SPI20	0.94 ± 0.04	6.58 ± 0.21
FSH10	1.04 ± 0.03	6.53 ± 0.35
FSH20	1.00 ± 0.03	6.53 ± 0.07
PSH10	1.06 ± 0.04	6.68 ± 0.19
PSH20	1.02 ± 0.01	6.69 ± 0.09

¹SPI10=10% Soy Protein Isolate, SPI20=20% Soy Protein Isolate, FSH10=10% Flavourzyme Soy Hydrolysate, FSH20=20%

Specific gravity values were more varied for muffin batter treatments. Specific gravity is a measure of the amount of air trapped by batter during mixing. Higher values indicate less trapped air while lower values indicate more trapped air, and optimal specific gravity for a given formulation is associated with larger baked product volume and better cell characteristics. Table 5.5 gives differences in means and adjusted p-values for the 15 defined contrasts. Addition of 10% SPI resulted in significantly ($p < 0.05$) lower batter specific gravity compared to control batter, and batter specific gravity also significantly decreased when SPI was increased from 10% to 20%. At the 10% level, addition of FSH and PSH did not change batter specific gravity compared to C, however, addition of 20% FSH and PSH did significantly decrease specific gravity compared C. Specific gravity was significantly increased when comparing PSH with native SPI at 10% and 20%. FSH did not significantly increase batter specific gravity compared to native SPI.

Batter specific gravity may be affected by foaming capacity of added protein ingredients. SPI is used in food formulations for its foaming properties to enhance egg white foaming or to replace eggs entirely (Wang and Wang, 2009; Lin et al., 2017; Hedayati and Mazaheri Tehrani, 2018). Studies differ on whether hydrolysis improves foaming capacity of SPI. Previous work has shown hydrolysis increases foaming capacity of SPI, (Puski, 1975; Were et al., 1997; Babiker, 2000; Molina Ortiz & Wagner, 2002); however, a decrease in foaming capacity was observed by Meinschmidt et al. (2015). Hydrolysates with Flavourzyme result in similar batter specific gravities compared to SPI, therefore, Flavourzyme hydrolysates may be used in soy protein batters without increasing specific gravity.

Table 5.5–Differences in specific gravity and custom contrast results

Contrast¹	Difference in Specific Gravity	SE	DF	t-value	p-value	Adjusted p-value²
1. C - SPI10	0.09	0.02	12	5.67	0.0001	0.0015
2. C - SPI20	0.14	0.02	12	9.37	<.0001	<.0001
3. C - FSH10	0.05	0.02	12	3.05	0.0101	0.1038

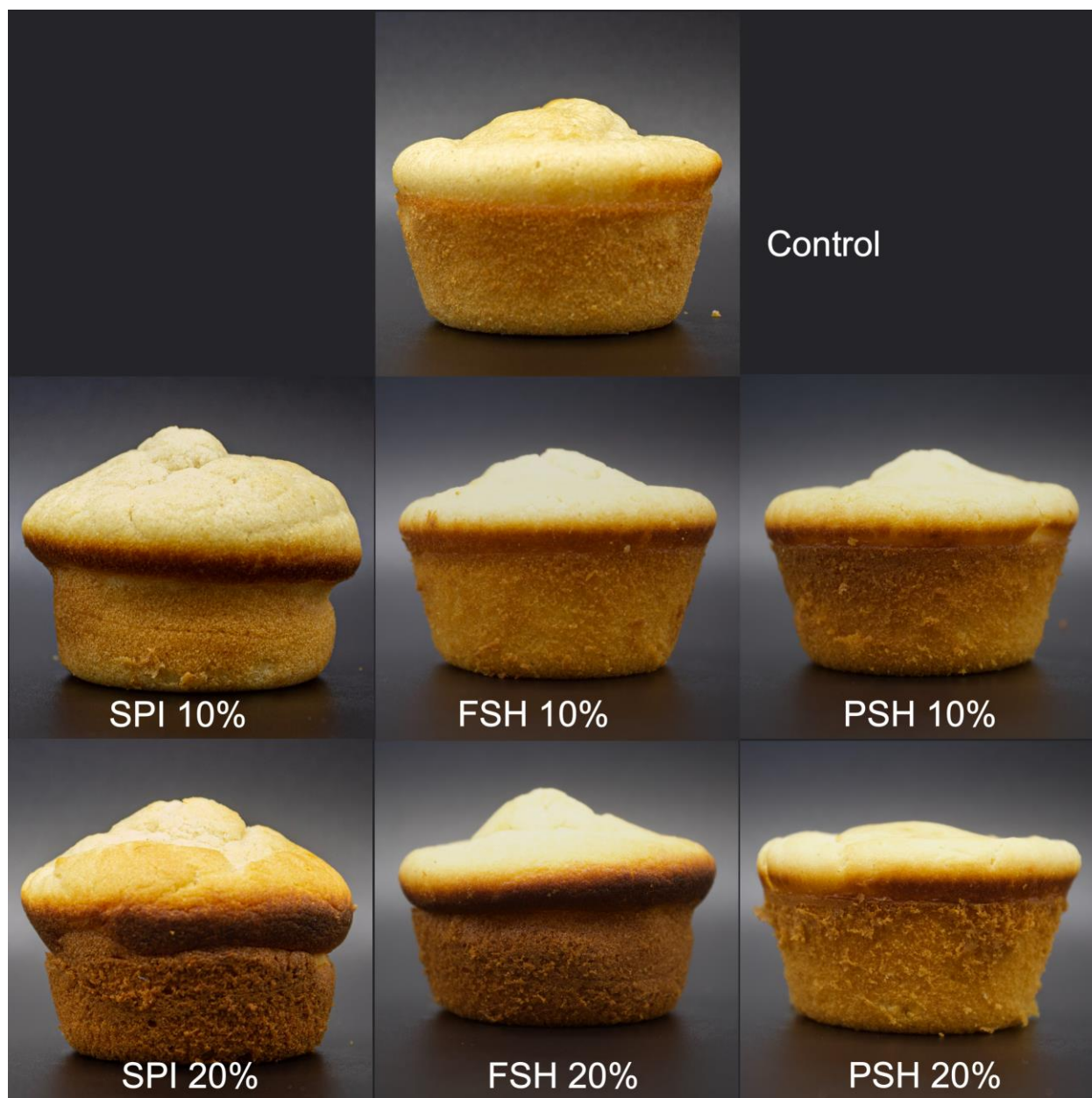
4. C - FSH20	0.09	0.02	12	5.89	<.0001	0.0011
5. C - PSH20	0.03	0.02	12	1.74	0.1067	0.6033
6. C - PSH20	0.06	0.02	12	4.14	0.0014	0.0172
7. FSH10 - SPI10	0.04	0.02	12	2.62	0.0226	0.2033
8. PSH10 - SPI10	0.06	0.02	12	3.92	0.0020	0.0247
9. FSH10 - PSH10	-0.02	0.02	12	-1.31	0.2154	0.8362
10. FSH20 - SPI20	0.05	0.02	12	3.49	0.0045	0.0510
11. PSH20 - SPI20	0.08	0.02	12	5.23	0.0002	0.0029
12. FSH20 - PSH20	-0.03	0.02	12	-1.74	0.1067	0.6033
13. SPI10 - SPI20	0.06	0.02	12	3.71	0.0030	0.0355
14. FSH10 - FSH20	0.04	0.02	12	2.83	0.0151	0.1462
15. PSH10 - PSH20	0.04	0.02	12	2.40	0.0337	0.2777

¹SPI10=10% Soy Protein Isolate, SPI20=20% Soy Protein Isolate, FSH10=10% Flavourzyme Soy Hydrolysate, FSH20=20%,

²Bolded adjusted p-values are considered statistically significant (p<0.05)

Muffin Visual Appearance

Muffin shape can be described as peak top, bell top, or flat top (Pyler 2009). Figure 5.3 shows muffin treatments from the side to highlight their shape. Crust color on the underside of the muffins was not measured, however, SPI20 and FSH20 have noticeably darker bottoms than other treatments. The protein hydrolysate treatments FSH10 and PSH10 look more similar in shape, size, and color to the control than SPI10. SPI10 muffins lacked symmetry as compared to the other 10% protein treatments. The PSH20 muffin was very flat compared to all other treatments with no obvious peak.

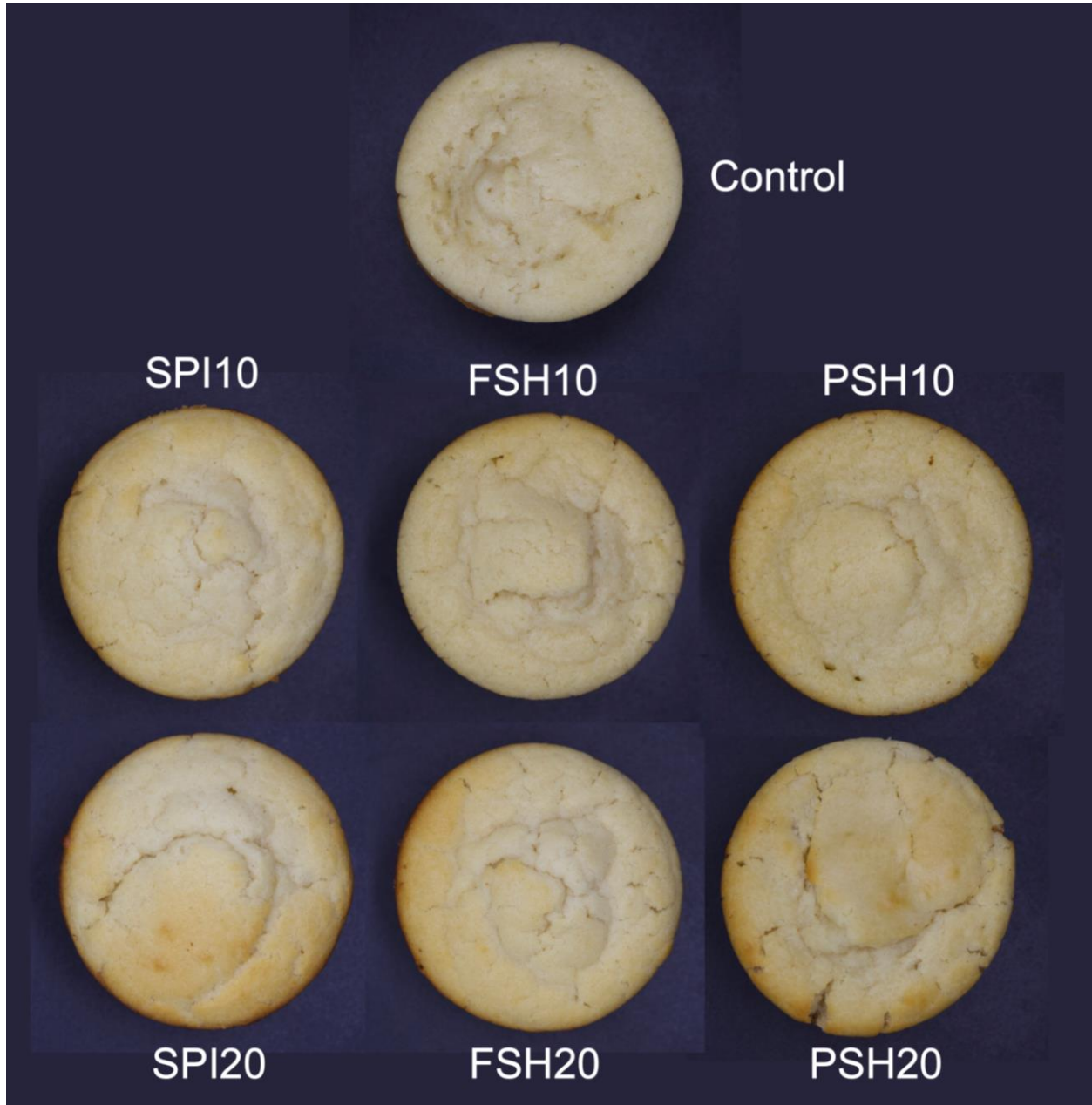


Note. SPI10=10% Soy Protein Isolate, SPI20=20% Soy Protein Isolate, FSH10=10% Flavourzyme Soy Hydrolysate, FSH20=20% Flavourzyme Soy Hydrolysate, PSH10=10% Papain Soy Hydrolysate, PSH20=20% Papain Soy Hydrolysate

Figure 5.3–Image of muffin treatments (side)

Muffin tops are pictured in figure 5.4. Because color measurements were taken in the center of each muffin top, data for differences in top edge color is not available. Color measurement using colorimeters is limited by location of color reading. The top edges of muffins are not flat and prevent scanning using colorimeters, therefore, visual observations are useful in determining differences in color. A visual observation of treatments shows all protein treatments

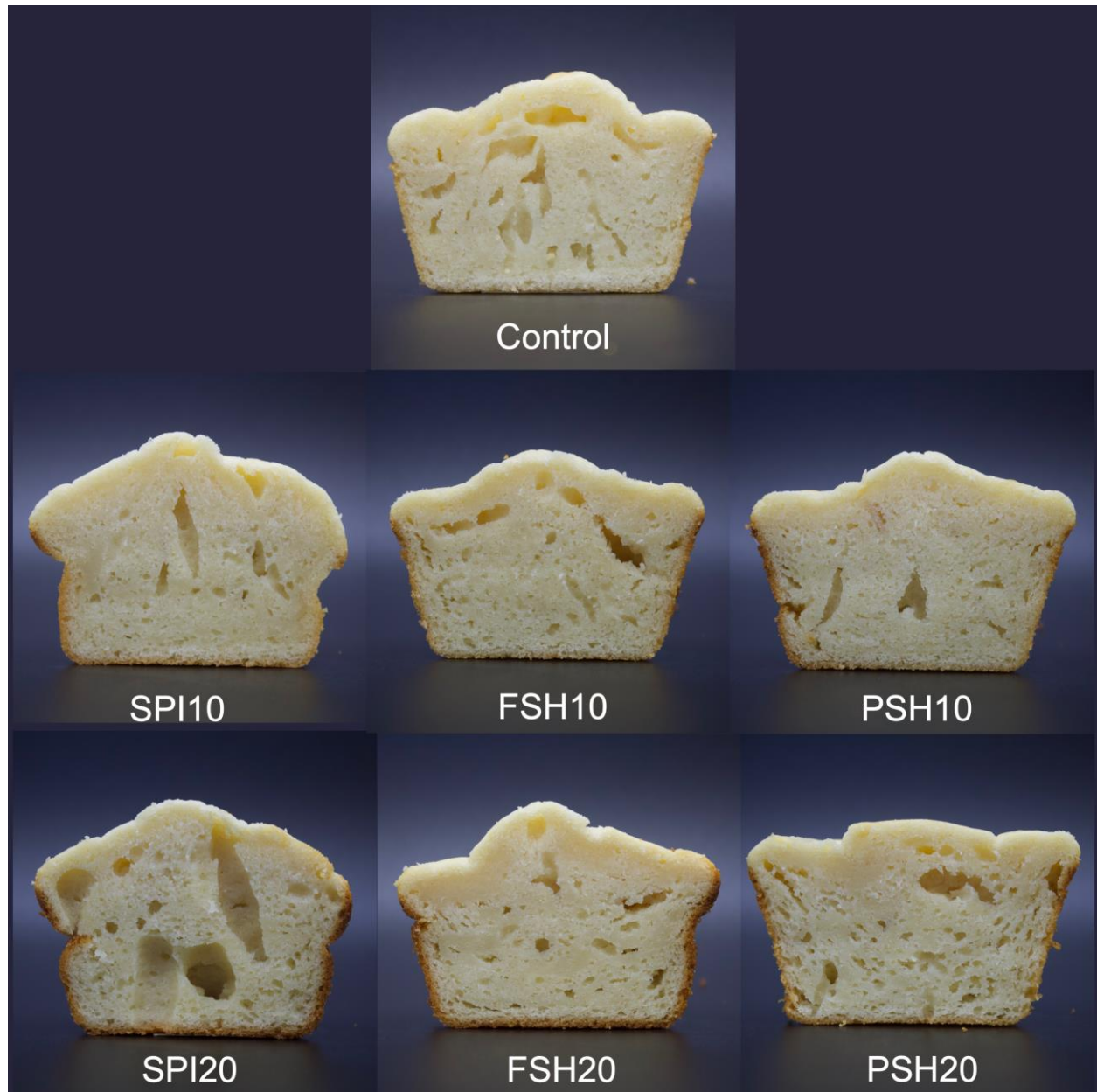
were darker than the control. The SPI20 treatment had a smoother surface with less cracks, while the PSH20 treatment had more cracks along the top and sides. Surface browning on the 20% protein treatments was uneven compared to the 10% protein treatments and control.



Note. SPI10=10% Soy Protein Isolate, SPI20=20% Soy Protein Isolate, FSH10=10% Flavourzyme Soy Hydrolysate, FSH20=20% Flavourzyme Soy Hydrolysate, PSH10=10% Papain Soy Hydrolysate, PSH20=20% Papain Soy Hydrolysate

Figure 5.4–Image of muffin treatments (top)

Figure 5.5 shows cross-sections of muffin treatments. Tunneling is seen in all treatments to some extent, with many tunnels in the control. Excessive tunneling is caused by overmixing, however, mix speed and time was held constant for all batters. SPI20 exhibits many large holes which may occur if air cells coalesce during baking.



Note. SPI10=10% Soy Protein Isolate, SPI20=20% Soy Protein Isolate, FSH10=10% Flavourzyme Soy Hydrolysate, FSH20=20% Flavourzyme Soy Hydrolysate, PSH10=10% Papain Soy Hydrolysate, PSH20=20% Papain Soy Hydrolysate

Figure 5.5–Image of muffin treatments (cross-section)

Muffin Crust and Crumb Color

Quantitative color data was obtained using a colorimeter to measure L*, a*, and b* values for crust and crumb (Table 5.7). L* values represent the light to dark parameter with 100 being the lightest and 0 being the darkest. Red to green is represented by the a* values with negative values representing green color and positive a* values representing red color. Blue to yellow is represented by the b* values with negative values representing blue color and positive a* values representing yellow color.

Overall, muffin crusts were moderately light with L* values between 70.86-74.59. Crust a* color values were all positive indicating more red than green color and ranged from 3.92-6.86. Crust b* color values were all positive indicating more yellow than blue color and ranged from 31.04-38.20.

Muffin crumb color was generally lighter than crumb color as indicated by lower L* values ranging from 72.73-74.77. Darker crust colors than crumb colors are expected because muffin crust dries during the final stage of baking and browns as a result of Maillard browning. Crumb a* values were again positive indicating more red than green; however, values ranged from 2.37-2.84 which is slightly less red than crust a* values. Muffin crumb b* values ranged from 26.52-27.81 indicating a less yellow crumb color compared to crust color.

Table 5.6– Means and standard deviations for muffin crust and crumb color

Treatment ¹	Crust Color			Crumb Color		
	L*	a*	b*	L*	a*	b*
C	73.69 ± 1.01	4.69 ± 0.36	32.87 ± 1.78	74.41 ± 0.84	2.37 ± 0.11	27.59 ± 0.17
SPI10	74.49 ± 0.61	3.92 ± 0.21	31.04 ± 0.96	74.77 ± 0.72	2.53 ± 0.11	27.36 ± 0.57
SPI20	73.46 ± 0.46	6.65 ± 1.21	36.61 ± 1.29	73.17 ± 1.71	2.78 ± 0.22	27.38 ± 0.56
FSH10	72.85 ± 1.40	4.05 ± 0.14	31.04 ± 1.37	74.34 ± 0.50	2.60 ± 0.16	27.81 ± 0.18
FSH20	72.97 ± 0.85	4.34 ± 0.97	31.93 ± 4.26	73.03 ± 1.42	2.84 ± 0.17	26.52 ± 0.59
PSH10	72.15 ± 0.76	5.14 ± 0.85	35.15 ± 2.48	73.93 ± 1.01	2.57 ± 0.10	27.57 ± 0.07
PSH20	70.86 ± 1.60	6.86 ± 1.33	38.20 ± 3.53	72.73 ± 0.10	2.82 ± 0.09	27.43 ± 0.10

¹SPI10=10% Soy Protein Isolate, SPI20=20% Soy Protein Isolate, FSH10=10% Flavourzyme Soy Hydrolysate, FSH20=20%,

Lightness values for SPI and FSH treatments were not different from the control, and at the 10% level FSH was also similar to the control (Table 5.8). PSH20 was lighter than the control ($P<0.05$). At the 20% level PSH was significantly lighter than SPI.

Table 5.7–Differences in crust L* values and contrast results

Contrast ¹	Difference in				Adjusted	
	Crust L*	SE	DF	t-value	p-value	p-value ²
1. C - SPI10	-0.804	0.733	12	-1.10	0.2941	0.9173
2. C - SPI20	0.226	0.733	12	0.31	0.7636	0.9999
3. C - FSH10	0.837	0.733	12	1.14	0.276	0.9028
4. C - FSH20	0.713	0.733	12	0.97	0.3498	0.9509
5. C - PSH20	1.721	0.733	12	2.35	0.0369	0.2974
6. C - PSH20	2.824	0.733	12	3.85	0.0023	0.0278
7. FSH10 - SPI10	-1.641	0.733	12	-2.24	0.0449	0.3439
8. PSH10 - SPI10	-2.526	0.733	12	-3.44	0.0049	0.0548
9. FSH10 - PSH10	0.884	0.733	12	1.21	0.2509	0.8789
10. FSH20 - SPI20	-0.488	0.733	12	-0.67	0.5184	0.9923
11. PSH20 - SPI20	-2.599	0.733	12	-3.54	0.004	0.0464
12. FSH20 - PSH20	2.111	0.733	12	2.88	0.0138	0.1361
13. SPI10 - SPI20	1.030	0.733	12	1.40	0.1854	0.7899
14. FSH10 - FSH20	-0.123	0.733	12	-0.17	0.8692	1.0000
15. PSH10 - PSH20	1.103	0.733	12	1.50	0.1582	0.7377

¹SPI10=10% Soy Protein Isolate, SPI20=20% Soy Protein Isolate, FSH10=10% Flavourzyme Soy Hydrolysate, FSH20=20%

²Bolded adjusted p-values are considered statistically significant ($p<0.05$)

Differences in crust a* values based on the defined contrasts are shown in Table 5.9. No significant change was observed when comparing added-protein treatments to C. At the 10% level, all protein types had similar crust a* values. At the 20% level, FSH was less red when compared to SPI and PSH. For SPI, increasing protein level from 10% to 20% resulted in lower a* values indicated a less red crust.

Table 5.8–Differences in crust a* values and contrast results

Contrast ¹	Difference			DF	t-value	p-value	Adjusted
	in Crust a*	SE					p-value ²
1. C - SPI10	0.770	0.654	12	1.18	0.2618	0.8899	
2. C - SPI20	-1.964	0.654	12	-3.00	0.0110	0.1119	
3. C - FSH10	0.638	0.654	12	0.98	0.3486	0.9503	
4. C - FSH20	0.352	0.654	12	0.54	0.6007	0.9976	
5. C - PSH20	-0.454	0.654	12	-0.69	0.5003	0.9904	
6. C - PSH20	-2.171	0.654	12	-3.32	0.0061	0.0672	
7. FSH10 - SPI10	0.132	0.654	12	0.20	0.8431	1.0000	
8. PSH10 - SPI10	1.224	0.654	12	1.87	0.0857	0.5303	
9. FSH10 - PSH10	-1.092	0.654	12	-1.67	0.1207	0.6453	
10. FSH20 - SPI20	-2.316	0.654	12	-3.54	0.0041	0.0466	
11. PSH20 - SPI20	0.207	0.654	12	0.32	0.7574	0.9999	
12. FSH20 - PSH20	-2.523	0.654	12	-3.86	0.0023	0.0276	
13. SPI10 - SPI20	-2.734	0.654	12	-4.18	0.0013	0.0161	
14. FSH10 - FSH20	-0.286	0.654	12	-0.44	0.6694	0.9992	
15. PSH10 - PSH20	-1.717	0.654	12	-2.63	0.0222	0.2004	

¹SPI10=10% Soy Protein Isolate, SPI20=20% Soy Protein Isolate, FSH10=10% Flavourzyme Soy Hydrolysate, FSH20=20%

²Bolded adjusted p-values are considered statistically significant (p<0.05)

Table 5.10 shows differences in crumb a* values for the 15 defined contrasts. At the 20% level, all protein types resulted in significantly less red crumb compared to C, while the 10% level did not significantly alter crumb a* values. When compared to SPI, no significant differences were observed in crumb a* values for enzyme-modified soy proteins compared to native SPI, so hydrolysis did not affect crumb redness.

Table 5.9–Differences in crumb a* values and contrast results

Contrast ¹	Difference in			DF	t-value	p-value	Adjusted
	Crumb a*	SE					p-value ²
1. C - SPI10	-0.158	0.094	12	-1.69	0.1173	0.6355	
2. C - SPI20	-0.407	0.094	12	-4.35	0.0009	0.0122	

3. C - FSH10	-0.223	0.094	12	-2.39	0.0342	0.2812
4. C - FSH20	-0.467	0.094	12	-4.99	0.0003	0.0043
5. C - PSH20	-0.201	0.094	12	-2.15	0.0526	0.3845
6. C - PSH20	-0.443	0.094	12	-4.74	0.0005	0.0064
7. FSH10 - SPI10	0.066	0.094	12	0.70	0.4966	0.9899
8. PSH10 - SPI10	0.043	0.094	12	0.46	0.6513	0.9989
9. FSH10 - PSH10	0.022	0.094	12	0.24	0.8161	1.0000
10. FSH20 - SPI20	0.060	0.094	12	0.64	0.5331	0.9937
11. PSH20 - SPI20	0.037	0.094	12	0.39	0.7018	0.9996
12. FSH20 - PSH20	0.023	0.094	12	0.25	0.8072	1.0000
13. SPI10 - SPI20	-0.249	0.094	12	-2.66	0.0207	0.1898
14. FSH10 - FSH20	-0.243	0.094	12	-2.60	0.0231	0.2072
15. PSH10 - PSH20	-0.242	0.094	12	-2.59	0.0236	0.2109

¹SPI10=10% Soy Protein Isolate, SPI20=20% Soy Protein Isolate, FSH10=10% Flavourzyme Soy Hydrolysate, FSH20=20%

²Bolded adjusted p-values are considered statistically significant (p<0.05)

Table 5.11 shows differences in crumb b* values for the 15 defined contrasts. FSH20 crumb color was significantly (p<0.05) less yellow than C and SPI20. Increasing FSH from 10% to 20% also resulted in less yellow crumb color. Lastly, at the 20% level, FSH was less yellow than PSH.

Table 5.10–Differences in crumb b* values and contrast results

Contrast ¹	Difference in			t-value	p-value	Adjusted p-value ²
	Crumb b*	SE	DF			
1. C - SPI10	0.229	0.211	12	1.09	0.2990	0.9209
2. C - SPI20	0.208	0.211	12	0.99	0.3438	0.9480
3. C - FSH10	-0.222	0.211	12	-1.05	0.3126	0.9302
4. C - FSH20	1.063	0.211	12	5.04	0.0003	0.0039

5. C - PSH20	0.017	0.211	12	0.08	0.9383	1.0000
6. C - PSH20	0.157	0.211	12	0.74	0.4717	0.9865
7. FSH10 - SPI10	0.451	0.211	12	2.14	0.0536	0.3899
8. PSH10 - SPI10	0.212	0.211	12	1.01	0.3340	0.9429
9. FSH10 - PSH10	0.239	0.211	12	1.13	0.2793	0.9056
10. FSH20 - SPI20	-0.856	0.211	12	-4.06	0.0016	0.0198
11. PSH20 - SPI20	0.051	0.211	12	0.24	0.8125	1.0000
12. FSH20 - PSH20	-0.907	0.211	12	-4.30	0.0010	0.0132
13. SPI10 - SPI20	-0.021	0.211	12	-0.10	0.9219	1.0000
14. FSH10 - FSH20	1.286	0.211	12	6.10	<.0001	0.0008
15. PSH10 - PSH20	0.140	0.211	12	0.66	0.5192	0.9924

¹SPI10=10% Soy Protein Isolate, SPI20=20% Soy Protein Isolate, FSH10=10% Flavourzyme Soy Hydrolysate, FSH20=20%

²Bolded adjusted p-values are considered statistically significant (p<0.05)

Muffin Height and Weight

Muffin height ranged from 41.7-46.7mm and no significant differences were observed among treatments (Table 5.6). Muffin height results are consistent with soy flour muffins produced by Banks et al. (2009), who found addition of defatted soy flour at 40-60% significantly (p<0.05) decreased muffin height. Muffins in the Banks et al. (2009) study had heights with added soy flour ranged from 40.83-43.00mm, while the control muffin had a height of 48.16mm. Muffin weight ranged from 68.2-72.1g (Table 5.6).

Table 5.11—Means and standard deviations for muffin height and weight

Treatment ¹	Weight (g)	Height (mm)
C	69.9 ± 0.90	43.9 ± 0.19
SPI10	70.8 ± 5.24	45.9 ± 1.09
SPI20	68.2 ± 7.20	46.7 ± 3.08
FSH10	71.9 ± 1.56	45.5 ± 0.71

FSH20	71.1 ± 4.98	43.5 ± 1.80
PSH10	69.1 ± 2.09	41.7 ± 1.55
PSH20	72.1 ± 2.10	46.2 ± 1.62

¹SPI10=10% Soy Protein Isolate, SPI20=20% Soy Protein Isolate, FSH10=10% Flavourzyme Soy Hydrolysate, FSH20=20%

Texture Profile Analysis

Texture Profile Analysis (TPA) was conducted to measure textural attributes of muffin crumb. Hardness is a common TPA parameter and measures the amount of force at first compression in Newtons. No significant differences were found for hardness, adhesiveness, resilience, cohesiveness, springiness, gumminess, or chewiness.

Table 5.12–Means and standard deviations for Texture Profile Analysis (TPA)

Treatment¹	Hardness (N)	Adhesiveness	Resilience	Cohesiveness	Springiness	Gumminess	Chewiness
C	10.342 ± 0.575	-0.038 ± 0.028	36.428 ± 0.661	0.645 ± 0.009	90.908 ± 1.014	6.671 ± 0.460	6.060 ± 0.357
SPI10	9.038 ± 0.605	-0.036 ± 0.018	33.425 ± 1.397	0.622 ± 0.019	89.292 ± 1.458	5.634 ± 0.293	5.031 ± 0.216
SPI20	6.853 ± 1.767	-0.005 ± 0.004	28.900 ± 1.845	0.570 ± 0.033	88.411 ± 1.003	3.962 ± 1.213	3.502 ± 1.116
FSH10	7.927 ± 1.673	-0.029 ± 0.017	36.759 ± 2.759	0.663 ± 0.017	82.298 ± 13.064	5.151 ± 1.129	4.384 ± 1.436
FSH20	11.174 ± 3.120	-0.034 ± 0.046	30.366 ± 6.240	0.597 ± 0.100	85.175 ± 1.184	6.778 ± 2.579	5.796 ± 2.285
PSH10	9.372 ± 2.229	-0.009 ± 0.010	33.213 ± 3.282	0.628 ± 0.054	87.677 ± 0.714	5.963 ± 1.871	5.239 ± 1.689
PSH20	8.777 ± 1.407	-0.016 ± 0.023	32.773 ± 2.746	0.643 ± 0.042	87.113 ± 2.788	5.700 ± 1.090	4.939 ± 0.794

¹SPI10=10% Soy Protein Isolate, SPI20=20% Soy Protein Isolate, FSH10=10% Flavourzyme Soy Hydrolysate, FSH20=20%

Water Activity

Table 5.13—Means and standard deviations for muffin water activity

Treatment ¹	Water Activity
C	0.5001 ± 0.0015
SPI10	0.5019 ± 0.0020
SPI20	0.5094 ± 0.0099
FSH10	0.5071 ± 0.0028
FSH20	0.5374 ± 0.0803
PSH10	0.5528 ± 0.0128
PSH20	0.5799 ± 0.0584

¹SPI10=10% Soy Protein Isolate, SPI20=20% Soy Protein Isolate, FSH10=10% Flavourzyme Soy Hydrolysate, FSH20=20%

Water activity was similar for all treatments and ranged from 0.5001-0.5799. Proteins have high water binding properties. For shelf-stable bakery products, water activity levels below 0.85 are preferred to reduce the risk of spoilage due to bacteria, yeast, or mold.

Conclusion

Muffins were prepared with added soy protein ingredients (soy protein isolate, Flavourzyme soy hydrolysate, papain soy hydrolysate) at two levels (10% and 20%) and compared to control muffins with no added protein. Physical properties measured included batter specific gravity and pH, crust and crumb color, texture, height, weight, and water activity. Of the properties measures, specific gravity and color were the only muffin attributes changed by addition of protein.

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Chapter 6 - Summary and Implications

An online undergraduate module featuring an at-home laboratory activity covering soybean products, processing, and end uses was developed and compared to an on-campus module and laboratory. Student grades for abstract writing were similar between online and on-campus courses and 72-93% of online students met or exceeded expectations on every rubric section. Students provided feedback on the module and activity via an online reflective survey using a Likert scale. Responses were largely positive and reflective survey results indicate that the module was effective in teaching students about soybean products processing and end uses. 80% or more of students in both course formats agreed or strongly agreed the module increased their food processing knowledge. Ninety-six percent of online students somewhat agreed or strongly agreed the provided instructions aided in their ability to complete the activity.

At the completion of the module, students were more aware of the advantages and disadvantages of adding soy ingredients to foods and were more open to trying soy products in the future. Students were exposed to the research process and communicated results in a scientific abstract. The laboratory exercise encouraged students to apply basic knowledge of food science principles to explore solutions to industry challenges. The soybean product processing module serves as a model for future online modules with hands-on activities to improve content knowledge and skills acquisition in food science and agriculture courses.

A baking with soy protein module with hands-on laboratory component for secondary education classes was developed and tested independently by teachers across Kansas. Overall, the module was well received by the teachers who participated in the survey. The study could be repeated utilizing techniques for better recruitment and response rates to better evaluate the module. After evaluating the module and making desired changes, the module could be made available as supplemental material for teachers in relevant fields. The module addresses national educational standards for both Family and Consumer Science and FFA Agricultural, Food, and Natural Resources to aid secondary teachers in providing students with novel learning experiences that supplement the existing curriculum. Both undergraduate and secondary modules were effective based on student performance data and reflective survey data.

In muffins prepared with two levels (10% and 20%) of soy protein types (soy protein isolate, Flavourzyme soy hydrolysate, papain soy hydrolysate), addition of protein resulted in no significant ($p < 0.05$) change in batter pH, textural attributes, height, or water activity. Specific gravity was most affected by addition of protein followed by color. Addition of Flavourzyme

hydrolysate did not significantly increase specific gravity compared to addition of native SPI. Addition of protein at 10% and 20% levels did not significantly alter most muffin physical properties, therefore, muffins with protein levels above 20% may retain acceptable physical qualities. Further research is needed to explore the effects of soy protein hydrolysate at levels greater than 20% on physical properties of muffins to aid in the development of nutritious, high-protein bakery foods. Additionally, while sensory analysis of soy hydrolysate protein slurries shows promising results for the use of enzymatic hydrolysis to reduce of beany flavor of soy protein isolate, sensory studies are needed to evaluate the sensory properties of hydrolysates in bakery foods.

Appendix A - Undergraduate Soy Module Supplemental Material

Supplemental Material A1–Undergraduate online soy muffin lab instructions

Pre-Lab Information: Before conducting this lab, you will need the following materials and equipment. A checklist is provided below.

- ___ Packet of ingredients from KSU with muffin mix for control, 50% soy flour 50% All-Purpose flour (SF 50) variation and 100% soy flour (SF100)
- ___ Vegetable oil
- ___ Eggs (6)
- ___ Muffin liners or no-stick cooking spray
- ___ Muffin tin or a disposable (one-time use) muffin tin
- ___ Large mixing bowl
- ___ Liquid measuring cup
- ___ Mixing spoon or spatula
- ___ Thermometer (good time to purchase one for safety)
- ___ Oven
- ___ Phone or camera for taking pictures

Please watch the video titled “Muffins: How Its Made” to learn about the commercial muffin processing procedures and to assist you in developing a flow diagram.

<https://www.youtube.com/watch?v=xFW5hJtkyQ&t=195s>

Lab instructions

Objectives: The objective of this lab is to observe the effect of different levels of soy flour in muffins while becoming familiar with the research process and the commercial muffin processing steps.

Variations:

- 1) Control - 100% Cake Flour
- 2) SF50 - 50% Cake Flour 50% Soy Flour
- 3) SF100 - 100% Soy Flour

Lab Test Formula:

Ingredient	Amount
Muffin Base	1 package (285.2g)
Oil	½ Cup
Egg	2 Whole Eggs
Water	1/3 Cup

Full Formulation:

Ingredients (g)	Weight (g)		
	Control	SF50	SF100
Soy Flour	0.0	50.6	101.2
AP Flour	101.2	50.6	0.0
Sugar	124.6	124.6	124.6
Mix	59.4	59.4	59.4
Oil	106.3	106.3	106.3
Eggs	112.0	112.0	112.0
Water	90.3	90.3	90.3

Baking Instructions:

- 1) Preheat oven to 375°F (190.6°C). Line a standard muffin tin with 6 paper muffin cups/liners or spray with Pam/no-stick cooking spray or grease with shortening.
- 2) In a large mixing bowl combine control muffin base, oil, and egg and mix for 1 minute. Below is an image of what the batter should look like at this stage.



- 3) Add cold tap water and mix for 1 minute. Below is an image of what the batter should look like at this stage. **Take pictures of each batter after mixing (3 pictures total) (6 pts.). Insert your pictures on the post-lab assignment.**



- 4) Spoon batter evenly into prepared pan filling cups $\frac{3}{4}$ full.
- 5) Bake the muffins for about 20 minutes. If you have a thermometer, you should test for doneness using the thermometer first. Muffins are done at around 200°F. From a food quality standpoint, muffins should be a pale golden brown and a toothpick inserted into the middle of a center muffin should come out clean. Commercial food facilities rely on both methods to ensure a safe and satisfactory product. From a food safety standpoint,

baking at 190.6°C for at least 17 minutes will reduce *Salmonella* by ≥ 5 logs (Channaiah et al. 2017).

- 6) Remove muffins from oven and let cool completely.
- 7) Repeat with variations SF50 and SF100.
- 8) Take pictures of the muffins for each variation (2 pictures total) (8 pts.). See example below. Insert your pictures on the post-lab assignment.

Side view



Top view



Control

SF50

SF100

Supplemental Material A2–Undergraduate online and on-campus soy module supporting material

MUFFIN INGREDIENTS AND PROCESSING STEPS

Author N. Cross*

*Adapted from Food Chemistry Workbook, edited by J.S. Smith and G. L. Christen, published
and copyright Science Technology System, West Sacramento, California, 2002.

Table 1. Steps in baking and application principles

Selection and scaling of ingredients	Application Principles
Flour	Gluten development, starch gelatinization, cell structure and volume, Maillard browning
Sugar	Flavor, tenderizer, crust quality, moisture retaining, reduction of water activity, Maillard browning
Fat	Flavor, tenderizer, moisturizing
Milk powder	Binding effect on flour protein, flavor, crust color, Maillard browning, moisture retention
Whole eggs	Protein coagulation, emulsification, flavor, color
Liquid	Hydration of flour proteins and starch, solvent for salt, sugar, leavening agent, moisture in final baked product
Leavening agent	Generation of carbon dioxide, volume, and cellular structure
Salt	Flavor enhancer
Additional ingredients	Variety in flavor, texture, and nutritive value
Processing steps	Application Principles
Mixing	Dispersion of ingredients, hydration of flour proteins and starch

Depositing	Scaling of muffin
Baking	Solubilization and activation of leavening agent, gelatinization of starch, coagulation of protein, caramelization of sugar, reduction of water activity, crumb development, color development, flavor development, crust formation, Maillard browning
Cooling	"Setting" of structure, water evaporation
Packaging	Retention of moisture, retention of flavor

Selection and scaling of ingredients

Muffins made by large commercial bakeries are cake type muffins while those made in the home or small institutions are bread muffins. A common problem encountered in bread type muffins is tunnel formation resulting from over development of gluten. However, since cake type formulas are higher in sugar, fat, and soft wheat flours, (all ingredients that interfere with gluten development) this problem is avoided. The discussion that follows is limited to cake muffins. The U.S. Dietary Guidelines¹⁴ promote healthy diets by increasing fiber while decreasing total fat, cholesterol, sugar, and sodium. Muffin formulas modified to meet the Guidelines utilize fat and sugar substitutes.

Flour

Flour is the primary ingredient in baked products. All-purpose flour is most commonly used in muffins. All-purpose flour is milled from hard or soft wheat or a mixture of hard and soft wheat.⁶ Flour contains starch and the proteins, glutenin and gliadin which hold other ingredients together to provide structure to the final baked product. Hydration and heat promote gelatinization of starch, a process which breaks hydrogen bonds, resulting in swelling of the starch granule which gives the batter a more rigid structure.¹² Hydration and mixing promote the development of a gluten matrix that provides structure.¹²

Substituting whole wheat flour, wheat germ, rolled oats or bran for part of the all-purpose flour is an excellent way to increase fiber. Other flours used in muffins include cornmeal, soy, oat, potato, and peanut. An acceptable product is possible when these flours are

substituted for one-third to one-half of all-purpose flour.¹² None of these flours contain glutenin or gliadin except whole wheat, and large pieces of bran in whole wheat flour cut and weaken gluten strands. Thus, there is minimal gluten development when these flours are used; the muffins tend to be crumbly and compact unless other modifications are made in the formula.

Sugar

Sugar contributes tenderness, crust color and moisture retention in addition to a sweet taste.³ Sucrose promotes tenderness by inhibiting hydration of flour proteins and starch gelatinization. Sugar is hygroscopic (attracts water) and maintains freshness. Corn syrup, molasses, maple sugar, fruit juice concentrates and honey are used as sweeteners for flavor variety. The quantity of liquid will need to be decreased if these sweeteners are used instead of sucrose because of the high-water content in these syrups.

Chemical changes in sugars during baking contribute characteristic flavors and browning. Caramelization of sugars involves inversion of sucrose, degradation of the ring structure, and creation of new polymers with distinctive flavors.¹² Maillard reactions (nonenzymatic browning) occur between the ketone or aldehyde group of reducing saccharides and amino acids in protein.⁶ Maillard reactions occur quickly at high temperatures and slowly at room temperature during periods of prolonged storage.¹² High fructose corn syrups and honey promote browning to a greater extent than sucrose due to the presence of high amounts of glucose and fructose.⁷ Maltose and lactose are reducing disaccharides and undergo browning but more slowly than monosaccharides.¹² Sucrose is a nonreducing disaccharide that requires hydrolysis and conversion to invert sugar (glucose plus fructose) for the Maillard reaction to occur. Water, an acidic pH, and heat facilitate sucrose hydrolysis. Heat also opens the ring structure to form an aldehyde or ketone.¹²

An acceptable muffin is possible with reduction of sugar by one-third.¹⁵ Non-nutritive sweeteners such as saccharin can be substituted for all or part of the sugar. Aspartame (Equal®) converts to diketopiperazine when heated. Diketopiperazine does not have the sweetness of aspartame.⁷ However, aspartame has been successfully substituted for all of the sugar in muffins, probably because the baking time is relatively short.⁶ Non-nutritive sweeteners however, do not contribute to tenderness, browning, or moisture retention and other adjustments in the muffin formulation may become necessary.

Fat

Fat contributes tenderness, flavor, and a characteristic mouthfeel to baked products.⁵ Fat keeps the crumb and crust soft and helps retain moisture, and thus contributes to texture and mouthfeel. Because fat dissolves flavor components, it enhances the flavor of baked products¹⁰. Both shortening and vegetable oils are used in muffins.

To meet the demands of the consumer, muffin formulas are being modified to reduce fat and cholesterol. Low fat and fat-free muffins are available ready-to-eat and as frozen batters or dry mixes for bake-off. Baked products are acceptable when fat is reduced by one-third.⁸ Bread muffins made with 25% less fat or 2 tablespoons per 1 cup of flour were comparable to standard muffins in shape, tenderness, flavor, and texture.⁵ Bread muffins were considered acceptable when safflower oil (high in polyunsaturated fatty acids) was used in amounts typically used in the formula, or reduced by one-fourth to one-third.⁴

Various types of fat replacers being used in baked products include emulsifiers, complex carbohydrates, corn syrups, sugar alcohols, gums, and fruit based fat replacers.^{11,16} Emulsifiers (esters of edible fatty acids and polyols) improve mouthfeel, texture and shelf-life of baked products by binding water and trapping air.¹¹ Common emulsifiers are monoglycerides and diglycerides. Monoglycerides retard starch retrogradation and extend shelf life by forming stable complexes with amylose.² Examples are Dimodan or Amidan by Danisco.² Polydextrose (LitesseTM) and sorbitol are examples of a complex carbohydrates and sugar alcohol respectively, that replace both fat and sugar. These help retain moisture and act as texturizers.¹⁶ Acceptable low fat cake muffins (5% fat) used 2% pregelatinized dull waxy starch (Amerimaize 2210, American Maize-Products) and corn syrup (3.6%) to replace fat.⁸ Examples of gums are guar, locust bean, xanthan and sugar beet fiber¹². Gums enhance mouthfeel and texture and extend the shelf life of baked products ¹².

Fruit purees or pastes of raisins, prunes, dates, and grapes as well as applesauce are being promoted as fat replacers. Just Like Shorten'TM is a mixture of dried prunes and apples.⁹ The fruit purees have humectant properties and promote tenderness and moistness and increase the shelf-life.¹¹ Sugar is reduced in formulas with fruit purees because of the natural sweetness of fruit.

Leavening Agents

Gases released by a leavening agent influence volume and cell structure. During baking,

heat increases gas volume and pressure to expand cell size until proteins are coagulated.¹² Stretching of the cell walls during baking improves texture and promotes tenderness.¹²

Baking powder contains an acid salt plus sodium bicarbonate and an inert ingredient such as starch to retard the reaction of the ingredients during storage. Double-acting baking powder (most commonly used in muffins) contains both slow and fast-acting acids. SAS-phosphate contains monocalcium phosphate as the fast-acting acid plus sodium aluminum sulfate (SAS), a slow acting acid. Fast acting acids are readily soluble at room temperature while slow acting acids are less soluble and require heat over extended time to release carbon dioxide.¹³

The quantity of leavening used in a baked product depends on the choice of leavening agent and other ingredients. For each cup of flour, 1 1/2 teaspoons of double-acting baking powder (SAS) are needed for leavening, or 5% based on flour at 100%. Baking soda may be used in addition to baking powder when muffins contain acidic ingredients such as sour cream, yogurt, buttermilk, light sour cream, molasses and some fruits and fruit juices.⁶ One cup of an acidic ingredient such as yogurt will contain sufficient hydrogen ions for 1/2 teaspoon of baking soda to leaven 1 cup of flour. Baking powder is required for leavening the remaining amount of flour.¹⁹ Sodium carbonate is a product of an incomplete reaction in formulas with excess sodium bicarbonate. Excess sodium carbonate results in a muffin with a soapy, bitter flavor, and a yellow color because of the effect of an alkaline medium on the anthoxanthin pigments of flour.¹⁹ Also, too much baking powder or soda results in a muffin with a coarse texture and low volume because of an over expansion of gas which causes the cell structure to weaken and collapse during baking. Inadequate amounts of baking powder will result in a compact muffin with low volume.

Whole eggs

Eggs provide flavor, color, and a source of liquid.³ Upon baking, the protein in egg white coagulates to provide structure. Lecithin in the yolk acts as an emulsifier and contributes to mouthfeel and keeping qualities. Modification of muffin formulas to reduce cholesterol may be accomplished by substituting an equivalent weight of egg whites or commercial egg substitute for whole eggs.

Nonfat dry milk powder

Milk powder is added to dry ingredients and water or fruit juice is used for liquid in muffin formulas.³ Milk powder binds flour protein to provide strength and body to muffins. In

addition, milk powder adds flavor and retains moisture.³ The aldehyde group from lactose in milk combines with the amino group from protein upon heating, contributing to Maillard browning.

Sodium chloride

The function of sodium chloride is to enhance the flavor of other ingredients.³ Sodium chloride may be omitted from the formula without compromising flavor if other ingredients such as dried fruit or spices are added for flavor.

Liquids

Liquids perform several functions in baked products. These include dissolving dry ingredients, hydration of gluten forming proteins, gelatinization of starch and moistness in the final baked product.¹² Insufficient water results in muffins low in volume with possible pockets of unmoistened dry ingredients.⁶

Additional Ingredients

Other ingredients are often added to muffins for variety in flavor, texture, and color and to increase the content of fiber, protein, vitamins, and minerals. Typical added ingredients are fresh and dried fruit, nuts, coconut, shredded carrots and zucchini, corn, grated cheese, chopped ham or bacon. Added flavorings include cinnamon, nutmeg, allspice, cloves, and orange or lemon peel.

Processing steps

Mixing

The muffin method of mixing involves adding liquid ingredients to a well in the dry ingredients and mixing using cutting or folding strokes until dry ingredients are moistened. Institutional or commercial bakeries use a mixer on slow speed for 3 to 5 minutes. Additional ingredients are added at the end of the mixing cycle or after depositing the muffin batter. Inadequate mixing results in a muffin with a low volume since some of the baking powder will be too dry to react completely.

Depositing

The standard size of baked muffins is two ounces although one and four ounces are also common. For institutions or bakeries, small batter depositors are available that will deposit four muffins at a time. Also available are large piston type depositors that maintain accurate flow of the batter.³

Baking

Many physical and chemical changes occur in the presence of heat to transform a liquid batter into a final baked muffin. Solubilization and activation of the leavening agent generates gases which expand to increase the volume of the muffin. Gelatinization of starch and coagulation of proteins provide permanent cell structure and crumb development. Carmelization of sugars and Maillard browning of proteins and reducing sugars promote browning of the crust. Reduced water activity facilitates Maillard Browning as well as crust hardening.

The choice of oven, baking pans and baking temperature influence the final baked product. A good flow of heat onto the bottom of the pan is necessary to produce a good product. Muffin tins are usually placed directly on the shelf or baking surface. The appropriate oven temperature is related to scaling and type of oven. Standard 2-ounce muffins are baked at 400° F (204°C) or slightly higher for a deck oven. Deck ovens may be stacked and are often used in small retail bakeries since these are cheaper and easier to maintain than reel or rotary ovens.¹⁰ Reel ovens consist of an insulated cubic compartment six or seven feet high. A ferris wheel type mechanism inside the chamber moves 4 to 8 shelves in a circle, allowing each shelf to be brought to the door for adding or removing muffin tins from the shelves.¹⁰ The reel oven is often preferred by retail bakers since several hundred to several thousand pounds of baked product can be baked each day.¹⁰ Rack ovens may be stationary or the racks may be rotated during baking.

Cooling

Products should be cooled prior to wrapping. This allows the structure to "set" and reduces the formation of moisture condensation within the package. Condensed moisture creates an excellent medium for yeast, mold, and bacterial growth and spoilage.

Wrapping

Muffins may be wrapped individually, in the tray in which they are baked, or transferred

into plastic form trays for merchandizing.³ The shelf life of muffins is 3 to 5 days for wrapped muffins, and 4 to 7 days for those packaged for wholesale and wrapped in foil or plastic wrap. The storage life of muffins is significantly influenced by the water activity. Cake muffins have a longer shelf life than bread muffins because of the high sugar content and lower water activity. Added ingredients such as cheese, ham or dried fruits are high in sodium or sugar content which reduces water activity¹² and increases the shelf life.

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USDA COMMERCIAL ITEM DESCRIPTION

Commercial Item Descriptions (CIDs) are documents that describe the most important characteristics of a commercial product, such as the types, and styles of products available. The CID may also contain information on analytical tests and requirements for food safety and quality for the product. CIDs are used by a purchaser during the procurement process to specify the product they wish to purchase.

CIDs may be used during the procurement process by anyone who does institutional feeding. This includes the school lunch program, the military, hospitals, day cares, and many other facilities. The CID is used by these institutions to specify exactly what they want in the commercial product that they are procuring.

The following supporting information for this lab is taken from the USDA CID for Muffins at: <https://www.ams.usda.gov/sites/default/files/media/CID%20Muffins%2C%20Fresh%20or%20Frozen.pdf>.

Processing: The fresh or frozen muffins shall be prepared in accordance with good manufacturing practice.

Ingredients: The fresh or frozen muffins shall include enriched flour; sweetening agents; eggs; peanut, corn, soybean, cottonseed, or canola vegetable oils, vegetable shortening, or butter; water; salt; flavorings; leavening agents; and emulsifiers or other stabilizers. Fruit puree or sauce may be used to replace oil in Style B Reduced fat, Style C Low fat, and Style D Fat free prepared fresh or frozen muffins. Fruit addition must not change characteristic flavor of the muffin. The fresh or frozen muffins may include nonfat milk and/or whey. The fresh or frozen muffins shall

include mold inhibitors of proper levels as allowed by the Federal Food, Drug, and Cosmetic Act.

Flour: The flour used for the muffins shall conform to the U.S. Standards for Enriched Flour (21 CFR 137.165).

Milk: The milk products used in the fresh or frozen muffins shall comply with all applicable requirements of the “Grade A Pasteurized Milk Ordinance - 1993 Recommendations of the United States Public Health Service” or latest revision thereof, or regulations substantially equivalent thereto.

Eggs: The eggs used in the fresh or frozen muffins shall conform to the applicable provisions of the Regulations Governing the Inspection of Eggs and Egg Products (7 CFR Part 59). The eggs shall be certified *Salmonella* free.

Finished product:

Appearance and color: The fresh or frozen muffin tops shall have a rounded pebbled surface and may be “sugar dusted”, “sugar crumb”, “sugar crumbled topping”, or “other appropriate topping”. The fresh or frozen muffin interiors shall have for example, a creamy white, slightly yellow, slightly blue, or caramel to moderate dark brown color, characteristic of the flavor of muffin. The fresh or frozen muffins shall be evenly baked without evidence of scorching or burning. Color of the fresh or frozen muffin surface shall be typical of the type of muffin. There shall be no foreign color to the product.

Odor and flavor: The fresh or frozen muffins shall have an odor and flavor characteristic of the particular type of muffin. There shall be no foreign odors or flavors such as, but not limited to, burnt, scorched, stale, rancid, or moldy.

Texture: The texture of the fresh or frozen muffins shall have a slightly moist, light, and tender crumb. When the fresh or frozen muffins include nuts or fruits, there shall be an even distribution of nuts or fruit throughout the crumb. The fresh or frozen muffins shall not contain spots of unbaked flour on the bottom of the muffin.

Foreign material: All ingredients shall be clean, sound, wholesome, and free from evidence of rodent or insect infestation.

Age requirement: Unless otherwise specified in the solicitation, contract, or purchase order, the fresh muffins shall be delivered within 48 hours after baking. When frozen muffins are specified, the fresh product shall be in a freezer within 6 hours after baking and frozen to a temperature of 0EF (-17.8EC), \pm 5EF and shall be at a temperature not higher than 10EF (-12.2EC) within 6

hours after being placed in the freezer. Unless otherwise specified in the solicitation, contract, or purchase order, the frozen muffins shall be manufactured not more than 90 days prior to delivery and shall not have exceeded 10EF (-12.2EC) at any time during storage and delivery.

21 CFR §101.62 Nutrient content claims for fat, fatty acid, and cholesterol content of foods.

Fat content claims. The terms “fat free,” “free of fat,” “no fat,” “zero fat,” “without fat,” “negligible source of fat,” or “dietarily insignificant source of fat” or, in the case of milk products, “skim” may be used on the label or in labeling of foods, provided that:

1. The food contains less than 0.5 gram (g) of fat per reference amount customarily consumed and per labeled serving or, in the case of a meal product or main dish product, less than 0.5 g of fat per labeled serving: and
2. The food contains no added ingredient that is a fat or is generally understood by consumers to contain fat unless the listing of the ingredient in the ingredient statement is followed by an asterisk that refers to the statement below the list of ingredients, which states “adds a trivial amount of fat,” “adds a negligible amount of fat,” or “adds a dietarily insignificant amount of fat;” and
3. As required in §101.13(e)(2), if the food meets these conditions without the benefit of special processing, alteration, formulation, or reformulation to lower fat content, it is labeled to disclose that fat is not usually present in the food (e.g., “broccoli, a fat free food”).

The terms “low fat,” “low in fat,” “contains a small amount of fat,” “low source of fat,” or “little fat” may be used on the label or in labeling of foods, provided that:

1. The food has a reference amount customarily consumed greater than 30 g or greater than 2 tablespoons and contains 3 g or less of fat per reference amount customarily consumed: or
2. The food has a reference amount customarily consumed of 30 g or less or 2 tablespoons or less and contains 3 g or less of fat per reference amount customarily consumed and per 50 g of food (for dehydrated foods that must be reconstituted before typical consumption with water or a diluent containing an insignificant amount, as defined in §101.9(f)(1), of all nutrients per reference amount customarily consumed, the per 50-g criterion refers to the “as prepared” form); and

3. If the food meets these conditions without the benefit of special processing, alteration, formulation, or reformulation to lower fat content, it is labeled to clearly refer to all foods of its type and not merely to the particular brand to which the label attaches (e.g., “frozen perch, a low-fat food”).

The terms “reduced fat,” “reduced in fat,” “fat reduced,” “less fat,” “lower fat,” or “lower in fat” may be used on the label or in the labeling of foods, provided that:

1. The food contains at least 25 percent less fat per reference amount customarily consumed than an appropriate reference food.

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LABORATORY INFORMATION

In this lab you will conduct a research experiment. Below are general guidelines for the process.

- 1) **Form your research question.** A research question is what you are looking to find answers to a problem or hypothesis. Sometimes the question comes from a company that is funding your research. Sometimes it is a topic you are genuinely passionate and

curious about (If you are lucky, it will be both!). In this lab, our research question is “does adding soy flour to muffins change the physical and sensory characteristics?”

2) **Conduct a literature review.** When you conduct a literature review, you are reading articles published by other scientists on your topic or related topics, to catch up on what is known already. You want your research to be novel and to contribute to the body of knowledge on your topic. In this lab, you will not need to conduct a literature review. You can consider your lecture slides as your literature review.

3) **Set-up the experiment.** Before you start baking, there are a lot of factors to consider. First, it is important to identify your variables. You are likely familiar with them, but in case you need a reminder, the mnemonic “**DRY MIX**” may be helpful.

Dependent	Manipulated
Responding	Independent
Y-Axis	X-axis

In this lab, the independent variable, or the variable manipulated, is the level of soy flour. The dependent variable, or the variable that responds, is the muffin characteristics. There are other variables in addition to the independent and dependent variables: confounding variables. These are variables that affect the data but, are not being studied in this experiment. Here are some examples:

- The oven does not hold a constant temperature during baking
- The humidity and temperature vary day by day
- Raw eggs vary in size

As scientists, we do our best to control these variables, but it is not always possible. One way to address confounding variables that we cannot control is to use replications. Replications are additional trials. The same experiment is repeated, usually three times. The data for the replications is averaged to spread out the effect of the confounding variables. Students typically confuse replications and variations, so make sure you understand the difference.

4) Conduct the experiment. This part accounts for a small portion of the research process. It is important to record your data carefully. For research that will be published, data should be recorded in pen in case the legitimacy of your data comes into question.

5) Analyze the data. Once the data has been collected it needs to be analyzed so we can draw conclusions accurately. The data for replications is averaged together and statistical analysis is performed. It is important to know if the differences in data are significant. For example, if the taste score for the control muffins was a 6 and the taste score for the SF50 muffins was an 8, it is obvious that the SF50 muffins scored higher for taste than control. What is not obvious however, is if the taste difference was large enough to be significant. Could it have been random chance that the SF50 muffins scored higher? Or did the SF50 muffins really taste better? Statistical analysis helps us decide. The statistical analysis software computes a P-value and compares it to a confidence level to decide, typically 0.05. The chart below explains the possible results.

P-Value	>	0.05	Not Statistically Significant
P-Value	<	0.05	Statistically Significant

6) Write the results. Scientists write the results of the experiment as a journal article and submit their results to relevant journals for other scientists to read. Other scientists can use the article in their own research. Scientists may also provide results to a supervisor in their own company. After the lab, the write-up will be discussed more.

Supplemental Material A3–Undergraduate online and on-campus soy module muffin scorecard and scoring information

MUFFIN EVALUATION

(Also refer to the Supporting Material for an official Common Item Description (CID) by USDA’s Agricultural Marketing Service)

Volume and cell structure are used for evaluating the quality of muffins. Cell structure can be evaluated by cutting the muffin in half and taking a picture of the cross-section. A desirable muffin should have a uniform cell structure without tunnels.

External Qualities

Color of crust

Color should be a pleasing golden brown, not pale or burnt.

Volume

The volume is determined indirectly by measuring the circumference of a cross section of the muffin ($\pi r^2 \times \text{height}$) in cubic centimeters and dividing by the weight in grams. A more objective method uses a volumeter to determine the volume of a baked product by measuring the volume of seeds in a closed system with and without the addition of the baked product.¹²

Internal Qualities

Grain

Uniform thick-walled cells are desirable. Coarseness, thin-cell walls, uneven cell size and tunnels indicate poor grain.

Aroma

Aroma is recognized by the sense of smell. The aroma may be sweet, rich, musty, or flat. The ideal aroma should be pleasant, fresh, sweet, and natural. Sharp, bitter, or foreign aromas are undesirable.

Taste

An acceptable muffin should have a pleasingly sweet flavor. Flat, foreign, salty, soda, sour or bitter tastes or unpleasant aftertastes are undesirable.

Texture

Texture is determined by the sense of touch. Texture depends on the physical condition of the crumb and is influenced by the grain. A desirable muffin should be easily broken, and slightly crumbly. Extreme crumbliness or toughness with lack of crumbling are undesirable characteristics.

Mouthfeel

Mouthfeel refers to the textural qualities perceived in the mouth. Characteristics can be described as gritty, hard, tough, tender, light and moist. A desirable muffin is tender, light, moist and requires minimal chewing.

Please use the following Scorecard for Muffins to evaluate the three muffin variations.

SCORECARD FOR MUFFINS¹

Name:	Sample and Score		
	Control	SF50	SF100
External Qualities			
Volume 1=low volume, compact cells; 5=light with moderate cells; 7=large volume, large cells and/or tunnels			
Contour of the surface 1=absolutely flat; 3=somewhat rounded; 5=pleasingly rounded; 7=somewhat pointed; 9=very pointed			
Crust color 1=much too pale; 3=somewhat pale; 5=pleasingly golden brown; 7=somewhat too brown; 9=much too brown			
Internal Qualities (cut the muffin in half from top to bottom to have a cross-section for evaluation)			

Name:	Sample and Score		
	Control	SF50	SF100
Interior color 1=much too white; 3=somewhat white; 5=pleasingly creamy; 7=somewhat too yellow; 9=much too yellow			
Cell uniformity and size 1=much too small; 3=somewhat thick; 5=moderate; 7=somewhat too large; 9= numerous large tunnels			
Thickness of cell walls 1=extremely thick; 3=somewhat thick; 5=normal thickness; 7=somewhat too thin; 9=much too thin			
Texture 1=extremely crumbly; 3=somewhat crumbly; 5=easily broken, 7=slightly crumbly; 9=tough, little tendency to crumble			
Flavor 1=absolutely not sweet enough; 3=not nearly sweet enough; 5=pleasingly sweet; 7=somewhat too sweet; 9=much too sweet			
Aftertaste 1=extremely distinct; 3=somewhat distinct; 5=none			
<u>Overall acceptability</u> 1=very unacceptable; 3=somewhat acceptable; 5=very acceptable			

¹Adapted from McWilliams 2001a.

Supplemental Material A4–Undergraduate online soy module post-lab assignment

Please insert pictures of your batter for Control, SF50 and SF100 below.

Please insert pictures of your side and top view of the three variations (control, SF50 and SF100)

Lab report:

As food scientists, it is important that you communicate your results in a variety of ways to suit your audience. For example, your writing would be different if you were submitting an article to a scientific journal versus presenting results from a study to the business executives of your company. Because we want to expose you to the research process, for this lab report you will write an abstract.

An abstract is a short, concise paragraph at the beginning of a journal article that quickly summarizes what the paper is about. It is useful for readers to decide if they would like to read the whole paper or if another source would better suit their needs. An abstract is simple to write if you know the parts. It includes a sentence or two for every section of the article: introduction, objectives, materials and methods, results. Below is an example, and then there is space for you to write your own. Your abstract should be 300 words or less but not less than 275 words.

Sample Abstract from “Physicochemical and nutritional properties of a healthy snack chip developed from germinated soybeans” (Maetens et al. 2017).

Introduction	[There is a growing interest in non-genetically modified, healthy snacks. Soy snacks are available on the market but snacks from germinated soybean are limited. The objectives of this research were to germinate non-genetically modified soybeans, analyze their physicochemical and nutritional characteristics, and develop a prototype snack chip. Moisture, protein, and lipid contents of flours from soybeans germinated for 1, 3, or 5 d ranged from 2.4 to 5.3, 42.7 to 43.3, and 21.4 to 25.5 g/100 g respectively ($P > 0.05$), while lipoxygenase-1 and lipoxygenase-3 activity, and trypsin inhibitor reductions ranged from 2.8 to 17.2, 16.2 to 26.1, and 16.6 to 31.1% and estimated glycemic indices ranged from 12.1 to 19.5. These reductions and estimated glycemic indices showed significant differences among the germinated soybeans ($P < 0.05$). The flour made from 5 d germinated
Objectives		
Materials and Methods		
Results		

soybean resulted in the highest reduction in lipoxygenase-1 and lipoxygenase-3 activities, and trypsin inhibitor content. Five-day germinated soybean chips were prepared with varying baking time, thickness, and amounts of baking soda. Based on fracturability, water activity, and color analysis, the optimal conditions were without baking soda, 1.25 mm thickness, and 10 min baking time. In conclusion, this prototype snack chip made from 5-day germinated soybean has the potential as a high nutritional, protein rich, low calorie healthy snack.

Write your abstract here (21 pts). Remember, it is one paragraph that is 300 words or less. A rubric is provided at the end of the lab instructions.

Post Lab Questions:

- 1) Explain the difference between a variation and a replication (2 pts.).
- 2) List at least three possible confounding variables in your experiment (3 pts.).
- 3) Create a flow diagram of how a muffin processing line might look in a commercial setting. This can be handwritten and attached as a picture, created in excel and attached as a separate document, or pasted below. Make sure you include receiving of ingredients AND packaging material and storage of ingredients AND packaging material (19 pts.).
- 4) List 2 advantages and 2 disadvantages of soy as an ingredient in food (4 pts.).

Use the sample abstract from “Physicochemical and nutritional properties of a healthy snack chip developed from germinated soybeans” above to answer the following questions:

- 5) What are the variations in this experiment? This will be different levels of the independent variable. (2 pts.)
(Would accept either stage as long as they match)
Stage 1: Moisture, protein, and lipid contents
- 6) What is being measured in this experiment? List at least 3 measurements. (3 pts.)

- 7) In the sample abstract taken from “Physicochemical and nutritional properties of a healthy snack chip developed from germinated soybeans” was the glycemic index statistically different between the germination variations? How do you know? (Hint: check the P-value!) (2 pts.)

Use the Pre-Lab Information and Supporting Material to answer the following questions. Circle the answer. (2 points each)

- 8) T or F Muffins made with bread flour are likely to have tunnels because of the high gluten potential.
- 9) T or F Excessive baking soda results in a muffin with a soapy, bitter flavor and a yellow color and coarse texture.
- 10) T or F Wrapping muffins before cooling will increase the shelf life by increasing the moisture content.
- 11) T or F According to the information and your observations, the protein content in soy flour causes more Maillard browning reactions to occur.
- 12) T or F Adding ham, cheese, or dried fruits to muffin batter will decrease the shelf life of muffins.
- 13) Undermixing muffins will result in a product with the following characteristics:
- a. Thick cell walls
 - b. Pockets of unmoistened dry ingredients
 - c. Low volume
 - d. Both b & c are correct
 - e. All of the above are correct
- 14) Emulsifiers such as monoglycerides and diglycerides function as fat substitutes by:

- a. Binding water
- b. Inhibiting hydration
- c. Promoting gelatinization
- d. Forming an outer crust that retains moisture

15) Maillard browning results from a reaction between the aldehyde group of glucose and other reducing sugars and:

- a. The hydroxyl group of water
- b. Sulfide groups in gluten
- c. The amino group of protein
- d. Carboxyl group of amino acids

16) The quantity of alternative flours that may be substituted for all-purpose flour for an acceptable muffin is:

- a. 10% to 20%
- b. 33% to 50%
- c. 75% to 100%
- d. 100%

17) An expected characteristic of muffins made by substituting all whole wheat flour for all-purpose flour is:

- a. Peaked top
- b. Tunnels
- c. Tough
- d. Crumbly

18) T or F The CID for muffins does not require the use of Grade A Pasteurized Milk.

19) T or F According to the picture provided for SF100, it would not be acceptable by CID due to evidence of scorching or burning.

20) T or F There are specific age requirements for both fresh and frozen muffins that are determined by time and temperature parameters.

21) T or F It is acceptable for a nuts or fruit be to unevenly distributed in the muffin.

22) From the video, describe how the blueberries were incorporated into the muffin batter.

**Supplemental Material A5–Undergraduate online and on-campus soy module
scientific abstract rubric**

Abstract Criteria	Exceeds Expectations	Meets Expectations	Below Expectations	No Expectations Met
Introduction (3 points)	Clear, concise, engaging; describes, connects the topic to literature and objectives (3 points)	Clear, but not engaging; Attempts to connect to literature (2 points)	Unclear; does not connect to literature (1 point)	Missing (0 points)
Objectives (3 points)	Clear, concise, and relevant; provides purpose of the study (3 points)	Clear but not concise; might contain irrelevant information; lacks specifics (2 points)	Unclear; contains irrelevant or unimportant information (1 point)	Missing (0 points)
Materials and Methods (4 points)	Identifies materials and methods used to answer the research question (3.5-4 points)	Somewhat identifies materials and methods used to answer the research question (3 points)	Minimally, identifies materials and methods used to answer the research question (2.5-1 points)	Missing (0 points)
Results (5 points)	Clear; provides explanation of what was expected, discovered, accomplished, collected, produced (4.5-5 points)	Attempts to present findings but might be unclear; some information missing (3-4 points)	Unclear or misinterpretation of the results (1-2.5 points)	Missing (0 points)

Professional Writing (5 points)	Few grammatical errors or typos; mixed verb tense (4.5-5 points)	Few grammatical errors or typos; mixed verb tense (3-4 points)	Many grammatical errors, typos but do not impeded understanding, inappropriate verb tense (1-2.5 points)	Grammatical errors, typos impede understanding, inappropriate verb tense (0 points)
Length (1 point)	275-300 words (1 point)	275-250 words (0.50 points)	Less than 250 words (0.25 words)	Only 100 words or less (0 points)

Supplemental Material A6–Module quiz, discussion, and exam questions

Post Laboratory Question Answers

1. Explain the difference between a variation and a replication.

Answer: A variation is a treatment designed measure a change in a variable of interest while a replication involved repeating the experiment multiple times to ensure any experimental similarities or differences were not due to random chance or other factors.

2. Create a flow diagram of how a muffin processing line might look in a commercial setting. This can be handwritten and attached as a picture, created in excel and attached as a separate document, or pasted below. Make sure you include receiving of ingredients AND packaging material and storage of ingredients AND packaging material.

3. List 2 advantages and 2 disadvantages of soy as an ingredient in food.

Advantages: High protein, low in saturated fats

Disadvantages: Beany flavor, may cause changes in physical quality

Use the sample abstract from “Physicochemical and nutritional properties of a healthy snack chip developed from germinated soybeans” above to answer the following questions:

What are the variations in this experiment? This will be different levels of the independent variable.

(Would have accepted either stage)

Stage 1: soybeans germinated for 1, 3, or 5 d

Stage 2: combinations of baking time, thickness, and amounts of baking soda.

What is being measured in this experiment? List at least 3 measurements.

Stage 1: moisture, lipid, protein amounts; lipoxygenase-1 and lipoxygenase-3 activity, and trypsin inhibitor reductions, glycemic index

Stage 2: fracturability, water activity, and color analysis

In the sample abstract taken from “Physicochemical and nutritional properties of a healthy snack chip developed from germinated soybeans” was the glycemic index statistically different between the germination variations? How do you know? (Hint: check the P-value!)

Yes, differences in glycemic index were statistically different because the p-value was < 0.05.

Use the Pre-Lab Information and Supporting Material to answer the following questions.

Circle the answer.

T or **F** Muffins made with bread flour are likely to have tunnels because of the high gluten potential.

T or **F** Excessive baking soda results in a muffin with a soapy, bitter flavor and a yellow color and coarse texture.

T or **F** Wrapping muffins before cooling will increase the shelf life by increasing the moisture content.

T or **F** According to the information and your observations, the protein content in soy flour causes more Maillard browning reactions to occur.

T or **F** Adding ham, cheese, or dried fruits to muffin batter will decrease the shelf life of muffins.

Undermixing muffins will result in a product with the following characteristics:

F. Thick cell walls

H. Low volume

G. Pockets of unmoistened dry ingredients

I. Both b & c are correct

J. All of the above are correct

Emulsifiers such as monoglycerides and diglycerides function as fat substitutes by:

E. Binding water

G. Promoting gelatinization

F. Inhibiting hydration

H. Forming an outer crust that retains moisture

Maillard browning results from a reaction between the aldehyde group of glucose and other reducing sugars and:

- E. The hydroxyl group of water
- F. Sulfide groups in gluten
- G. **The amino group of protein**
- H. Carboxyl group of amino acids

The quantity of alternative flours that may be substituted for all-purpose flour for an acceptable muffin is:

- E. 10% to 20%
- F. **33% to 50%**
- G. 75% to 100%
- H. 100%

An expected characteristic of muffins made by substituting all whole wheat flour for all-purpose flour is:

- E. Peaked Top
- F. Tunnels
- G. **Tough**
- H. Crumbly

T or F The CID for muffins does not require the use of Grade A Pasteurized Milk.

T or F According to the picture provided for SF100, it would not be acceptable by CID due to evidence of scorching or burning.

T or F There are specific age requirements for both fresh and frozen muffins that are determined by time and temperature parameters.

T or F It is acceptable for nuts or fruit to be unevenly distributed in the muffin.

Quiz Question Answers

A product extracted from soybeans similar to eggs that serves as a binding agent:

- E. Soybean oil
- F. Soy protein
- G. **Lecithin**
- H. Soybean meal

Fermentation assists with the following:

- E. Deactivating the antioxidant activity
- F. Decreasing the phenolic compound concentration
- G. **Improving nutrient digestibility**
- H. All of the above

True or False: Soybeans may turn a purple color due to drought and a specific fungus problem.

Match the soy product with a specific food application.

- | | |
|------------------------------|------------------------------|
| 5. Traditional Foods (F) | F. Miso |
| 6. Okara(G) | G. High fiber Breads |
| 7. Non-traditional foods (H) | H. Soynut butter |
| 8. Lecithin (I) | I. Emulsifying agents |
| | J. Isoflavones (Not matched) |

Match the term with the definition.

- | | |
|------------------------------|--|
| 5. Canning (E) | E. Commercially Sterile |
| 6. High-Pressure Cooking (F) | F. Hydrostatic Pressure |
| 7. Extrusion (G) | G. Twin-screw pushes product through a die |
| 8. Soaking (H) | H. Softens cotyledon |

Supplemental Material A7–Undergraduate on-campus soy muffin lab instructions

Objectives: The objective of this lab is to observe the effect of soy protein level on the physical and sensory characteristics of soy muffins while becoming familiar with the research process and commercial muffin processing steps.

Variations:

- 1) Control – No Soy Protein Isolate (SPI)
- 2) 10% Soy Protein Isolate
- 3) 20% Soy Protein Isolate
- 4) 30% Soy Protein Isolate

Formula:

Ingredient	Control		10% SPI		20% SPI		30% SPI	
	Baker %	g	Baker %	g	Baker %	g	Baker %	g
All-Purpose Flour	100%	140	90%	126	80%	112	70%	98
Cornstarch	10%	14	10%	14	10%	14	10%	14
Granulated sugar	63%	88	63%	88	63%	88	63%	88
Baking powder	4%	5.4	4%	5.4	4%	5.4	4%	5.4
Salt	2%	2.6	2%	2.6	2%	2.6	2%	2.6
Non-fat dry milk	15%	21.6	15%	21.6	15%	21.6	15%	21.6
Whole dry egg	8%	11	8%	11	8%	11	8%	11
Shortening	71%	100	71%	100	71%	100	71%	100
Water	165%	231	165%	231	165%	231	165%	231
Vanilla	1%	2	1%	2	1%	2	1%	2
Soy protein isolate	0%	0	10%	14	20%	28	30%	42

Baking Instructions:

- 9) Preheat oven to 375°F (190.6°C). Spray a muffin tin with nonstick cooking spray.
- 10) In a small mixing bowl add the soy protein isolate and water. Mix well by hand until no lumps remain. Allow to hydrate for at least 5 minutes.

- 11) In a large mixing bowl combine all ingredients except water-protein slurry and vanilla and mix for 1 minute on speed 1. Scrape the sides of the bowl.
- 12) Add approximately half of the protein slurry and mix for 30 seconds on speed 1. Scrape the sides of the bowl.
- 13) Add the rest of the protein slurry and vanilla and mix for 30 seconds on speed 1. Scrape the sides of the bowl.
- 14) Mix on Speed 2 for 2 minutes.
- 15) Measure the specific gravity of the batter and record in **Table 3**. Discard batter used for specific gravity.
- 16) Weigh the muffin tin on a scale and record in **Table 1**. Deposit one level scoop in each cup.
- 17) Weigh the muffin tin filled with batter and record in **Table 1**. Calculate the average muffin batter weight.
- 18) Bake the muffins for about 23 minutes. Check internal temperature with a thermometer inserted into the center of a middle muffin. Muffins are done at around 200°F. The muffins may be undercooked even if they read 200°F, so also check for doneness by inserting a toothpick in the center.
 Note: *From a food quality standpoint*, muffins should be a pale golden brown and a toothpick inserted into the middle of a center muffin should come out clean. Commercial food facilities rely on both methods to ensure a safe and satisfactory product. *From a food safety standpoint*, baking at 190.6°C for at least 17 minutes will reduce *Salmonella* by ≥ 5 logs (Channaiah et al. 2017).
- 19) Remove muffins from oven and let cool completely.
- 20) Record the weight of the muffin tins with baked muffins. Calculate the average baked muffin weight, moisture loss during baking, and % moisture loss. Record in **Table 2**.
- 21) Exchange data with other groups to complete **Table 4**.
- 22) Score all 4 variations using the provided **Muffin Scorecard**.

Table 1: Muffin Batter Weight

Muffin Tin Weight (g)	Muffin Tin + Batter Weight (g)	Muffin Batter Weight (g)	Number of Muffins	Avg Muffin Batter Weight
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Table 2: Baked Muffin Weight

Muffin Tin + Baked Muffins (g)	Baked Muffin Weight (g)	Number of Muffins	Avg Muffin Weight (g)	Moisture Loss During Baking (g)	% Moisture Loss

Table 3: Batter Specific Gravity

Batter weight (g)	Water Weight	Specific Gravity

Table 4: Summary Data

	Control	SPI 10%	SPI 20%	SPI 30%
Specific Gravity				
Avg Muffin Weight (g)				
Avg % Moisture Loss				
Protein (g per muffin)				

Lab 12 Turn in Checklist

See word document template in Canvas

- | | |
|--|---|
| <input type="checkbox"/> Post-lab questions | Can be hand-written and scanned in or typed |
| <input type="checkbox"/> Flow Diagram (Q 22) | Typed |
| <input type="checkbox"/> Tables 1-4 | Typed |
| <input type="checkbox"/> Muffin scorecard | Typed |
| <input type="checkbox"/> Abstract | Typed |

Supplemental Material A8–Undergraduate on-campus soy module post-lab assignment

1. Post Lab Questions

Typed and pasted here or scanned and uploaded. If uploading handwritten answers, delete questions below.

- 1) Explain the difference between a variation and a replication (2 pts.).
- 2) List at least three possible extraneous variables in your experiment (3 pts.).
- 3) List 2 advantages and 2 disadvantages of soy as an ingredient in food (4 pts.).

Questions 5-7 are based on the sample abstract from “Physicochemical and nutritional properties of a healthy snack chip developed from germinated soybeans”

- 4) What are the variations in this experiment? Hint: This will be the independent variable. (2 pts.)
- 5) What are the dependent variables? Hint: What being measured in this experiment? List at least 3 measurements. (3 pts.)
- 6) In the sample abstract taken from “Physicochemical and nutritional properties of a healthy snack chip developed from germinated soybeans” was the glycemic index statistically different between the germination variations? How do you know? (Hint: check the P-value!) (2 pts.)

Use the Pre-Lab Information and Supporting Material to answer the following questions. Circle the answer. (2 points each)

- 7) T or F Muffins made with bread flour are likely to have tunnels because of the high gluten potential.
- 8) T or F Excessive baking soda results in a muffin with a soapy, bitter flavor and a yellow color and coarse texture.
- 9) T or F Wrapping muffins before cooling will increase the shelf life by increasing the moisture content.

- 10) T or F According to the information and your observations, the protein content in soy flour causes more Maillard browning reactions to occur.
- 11) T or F Adding ham, cheese, or dried fruits to muffin batter will decrease the shelf life of muffins.
- 12) Undermixing muffins will result in a product with the following characteristics:
- a. Thick cell walls
 - b. Pockets of unmoistened dry ingredients
 - c. Low volume
 - d. Both b & c are correct
 - e. All of the above are correct
- 13) Emulsifiers such as monoglycerides and diglycerides function as fat substitutes by:
- a. Binding water
 - b. Inhibiting hydration
 - c. Promoting gelatinization
 - d. Forming an outer crust that retains moisture
- 14) Maillard browning results from a reaction between the aldehyde group of glucose and other reducing sugars and:
- a. The hydroxyl group of water
 - b. Sulfide groups in gluten
 - c. The amino group of protein
 - d. Carboxyl group of amino acids
- 15) The quantity of alternative flours that may be substituted for all-purpose flour for an acceptable muffin is:
- a. 10% to 20%
 - b. 33% to 50%
 - c. 75% to 100%
 - d. 100%

- 16) An expected characteristic of muffins made by substituting all whole wheat flour for all-purpose flour is:
- a. Peaked top
 - b. Tunnels
 - c. Tough
 - d. Crumbly
- 17) T or F The CID for muffins does not require the use of Grade A Pasteurized Milk.
- 18) T or F According to the picture provided for SF100, it would not be acceptable by CID due to evidence of scorching or burning.
- 19) T or F There are specific age requirements for both fresh and frozen muffins that are determined by time and temperature parameters.
- 20) T or F It is acceptable for nuts or fruit to be unevenly distributed in the muffin.
- 21) A video is posted in Canvas on a muffin production process. From the video, describe how the blueberries were incorporated into the muffin batter. You should also use this video as a reference for making your flow diagram.

2. Flow Diagram

Create a flow diagram of how a muffin processing line might look in a commercial setting. Make sure you include receiving of ingredients AND packaging material and storage of ingredients AND packaging material (19 pts.).

3. Tables

Table 1: Muffin Batter Weight

Muffin Tin Weight (g)	Muffin Tin + Batter Weight (g)	Muffin Batter Weight (g)	Number of Muffins	Avg Muffin Batter Weight

Table 2: Baked Muffin Weight

Muffin Tin + Baked Muffins (g)	Baked Muffin Weight (g)	Number of Muffins	Avg Muffin Weight (g)	Moisture Loss During Baking (g)	% Moisture Loss

Table 3: Batter Specific Gravity

Batter weight (g)	Water Weight	Specific Gravity

Table 4: Summary Data

	Control	SPI 10%	SPI 20%	SPI 30%
Specific Gravity				
Avg Muffin Weight (g)				

Avg % Moisture Loss				
Protein (g per muffin)				

4. Muffin Scorecard

Typed here

	Control	10% SPI	20% SPI	30% SPI
External Qualities				
Volume 1=low volume, compact cells; 5=light with moderate cells; 7=large volume, large cells and/or tunnels				
Contour of the surface 1=absolutely flat; 3=somewhat rounded; 5=pleasingly rounded; 7=somewhat pointed; 9=very pointed				
Crust color 1=much too pale; 3=somewhat pale; 5=pleasingly golden brown; 7=somewhat too brown; 9=much too brown				
Internal Qualities (cut the muffin in half from top to bottom to have a cross-section for evaluation)				
Interior color 1=much too white; 3=somewhat white; 5=pleasingly creamy; 7=somewhat too yellow; 9=much too yellow				
Cell uniformity and size 1=much too small; 3=somewhat thick; 5=moderate; 7=somewhat too large; 9= numerous large tunnels				
Thickness of cell walls 1=extremely thick; 3=somewhat thick; 5=normal thickness; 7=somewhat too thin; 9=much too thin				
Texture 1=extremely crumbly; 3=somewhat crumbly; 5=easily broken, 7=slightly crumbly; 9=tough, little tendency to crumble				
Flavor 1=absolutely not sweet enough; 3=not nearly sweet enough; 5=pleasingly sweet; 7=somewhat too sweet; 9=much too sweet				
Aftertaste 1=extremely distinct; 3=somewhat distinct; 5=none				

<u>Overall acceptability</u> 1=very unacceptable; 3=somewhat acceptable; 5=very acceptable				
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Adapted from McWilliams 2001a.

5. **Abstract**

Typed here.

Supplemental Material A9–Undergraduate soy module IRB exempt letter



University Research
Compliance Office

TO: Dr. Kelly Getty
Food Science Institute
Call Hall

Proposal Number: 10248

A handwritten signature in black ink, appearing to be "Rick Scheidt", written over a horizontal line.

FROM: Rick Scheidt, Chair
Committee on Research Involving Human Subjects

DATE: 11/13/2020

RE: Proposal Entitled, "Development of Food Science and Baking Science Laboratory Exercises Utilizing Soybean Protein Extracts"

The Committee on Research Involving Human Subjects / Institutional Review Board (IRB) for Kansas State University has reviewed the proposal identified above and has determined that it is EXEMPT from further IRB review. This exemption applies only to the proposal - as written – and currently on file with the IRB. Any change potentially affecting human subjects must be approved by the IRB prior to implementation and may disqualify the proposal from exemption.

Based upon information provided to the IRB, this activity is exempt under the criteria set forth in the Federal Policy for the Protection of Human Subjects, **45 CFR §104(d), category: 6, subsection: i.**

Certain research is exempt from the requirements of HHS/OHRP regulations. A determination that research is exempt does not imply that investigators have no ethical responsibilities to subjects in such research; it means only that the regulatory requirements related to IRB review, informed consent, and assurance of compliance do not apply to the research.

Any unanticipated problems involving risk to subjects or to others must be reported immediately to the Chair of the Committee on Research Involving Human Subjects, the University Research Compliance Office, and if the subjects are KSU students, to the Director of the Student Health Center.

Supplemental Material A10–Undergraduate online and on-campus soy module IRB informed consent form



Institutional Review Board (IRB) Informed Consent Template Form

comply@k-state.edu | 785-532-3224

PROJECT TITLE:

Development of Food Science and Baking Science Laboratory Exercises Utilizing Soybean Protein Extracts

PROJECT APPROVAL DATE:

11/13/2020

PROJECT EXPIRATION DATE:

N/A

LENGTH OF STUDY:

15
minutes

PRINCIPAL INVESTIGATOR: Dr. Kelly Getty

CO-INVESTIGATOR(S): Dr. Yonghui Li, Dr. Umut Yucel, Aaron Clanton, Janae Brown

CONTACT DETAILS FOR PROBLEMS/QUESTIONS:

Dr. Kelly Getty (785) 532-2203
Janae Brown (479) 685-8252

IRB CHAIR CONTACT INFORMATION:

For the subject should he/she have questions or wish to discuss on any aspect of the research with an official of the university or the IRB. These are: Rick Scheidt, Chair, Committee on Research Involving Human Subjects, 203 Fairchild Hall, Kansas State University, Manhattan, KS 66506, (785) 532-3224; Cheryl Doerr, Associate Vice President for Research Compliance, 203 Fairchild Hall, Kansas State University, Manhattan, KS 66506, (785) 532-3224

PROJECT SPONSOR:

Kansas Soybean Commission

PURPOSE OF THE RESEARCH:

Undergraduate Students enrolled in the online Fundamentals of Food Processing course will conduct laboratory exercises from home or laboratory setting to evaluate the effect of soybean protein extract on various baked grain products, including pancakes and muffins. This study is part of a thesis research project. The objective of this study is to develop educational modules about soybean processing and assess how they affect students' perception and understanding of soybean processing and soybean utilization in human foods.

PROCEDURES OR METHODS TO BE USED:

Students that participated in the laboratory exercise will receive an online message asking for their willingness to participate in this study. Should they choose to participate they will be asked to answer survey questions online.

BIOLOGICAL SAMPLES COLLECTED (Describe procedure, storage, etc.):

N/A

ALTERNATIVE PROCEDURES OR TREATMENTS, IF ANY, THAT MIGHT BE ADVANTAGEOUS TO SUBJECT:

N/A

RISKS OR DISCOMFORTS ANTICIPATED:

There are no known risks of participation.

BENEFITS ANTICIPATED:

Participants will be acknowledged of their time via an online message upon completing the survey. Additionally, participants who complete the survey will receive bonus points toward their laboratory grade.

EXTENT OF CONFIDENTIALITY:

Quantitative responses will be aggregated. Subjects will provide written consent if direct quotes are to be included in the project outcome.

IS COMPENSATION OR MEDICAL TREATMENT AVAILABLE IF INJURY OCCURS? N/A

Terms of participation: I understand this project is research, and that my participation is voluntary. I also understand that if I decide to participate in this study, I may withdraw my consent at any time, and stop participating at any time without explanation, penalty, or loss of benefits, or academic standing to which I may otherwise be entitled.

By clicking the button below, you acknowledge that your participation is voluntary, you are 18 years of age, and that you are aware that you may choose to terminate your participation in the study at any time and for any reason.

☐ I consent, begin the study

☐ I do not consent, I do not wish to participate

Supplemental Material A11–Undergraduate online and on-campus soy module IRB debriefing statement

IRB Debriefing Statement

Thank you for participating in our survey. Your responses are critical to improving this laboratory exercise and learning experience for future students enrolled in FDSCI 305: Fundamentals of Food Processing. All data collected in this survey will remain confidential and anonymous when discussed in peer reviewed journal publications.

If you are interested in receiving a short report of the results, please email jeb98@ksu.edu and provide an email address where the report may be sent. Again, thank you for your time and assistance in completing this research.

Sincerely,

Janae Brown
M.S. Graduate Student
Food Science Institute
Kansas State University
jeb98@ksu.edu

Kelly Getty, Ph.D.
Associate Professor
Food Science Institute
Kansas State University
kgetty@ksu.edu

☐ Yes, I would like to receive a short report of the results collected from this survey.

Email address: _____

Supplemental Material A12–Undergraduate online soy module IRB survey questions

Muffin Physical and Sensory Properties

1. My understanding of the effect of soy ingredients on sensory properties (appearance, flavor, texture, etc.) of muffins was improved.
☐ Strongly Disagree ☐ Somewhat Disagree ☐ Neither Agree nor Disagree ☐ Somewhat Agree ☐ Strongly Agree
2. My understanding of the function of soy ingredients on physical properties (volume, color, cell structure, etc.) of muffins was improved.
☐ Strongly Disagree ☐ Somewhat Disagree ☐ Neither Agree nor Disagree ☐ Somewhat Agree ☐ Strongly Agree
3. his laboratory exercise introduced me to standard preparation procedures (mixing times, baking times, etc.) for muffins.
☐ Strongly Disagree ☐ Somewhat Disagree ☐ Neither Agree nor Disagree ☐ Somewhat Agree ☐ Strongly Agree

Research and Writing Skills

4. My ability to accurately communicate scientific data was reinforced.
☐ Strongly Disagree ☐ Somewhat Disagree ☐ Neither Agree nor Disagree ☐ Somewhat Agree ☐ Strongly Agree
5. My abstract writing skills were improved.
☐ Strongly Disagree ☐ Somewhat Disagree ☐ Neither Agree nor Disagree ☐ Somewhat Agree ☐ Strongly Agree
6. My knowledge of experimental design was improved.

☐ Strongly Disagree ☐ Somewhat Disagree ☐ Neither Agree nor Disagree ☐
Somewhat Agree ☐ Strongly Agree

7. The importance of using scientific and/or government resources was reinforced.

☐ Strongly Disagree ☐ Somewhat Disagree ☐ Neither Agree nor Disagree ☐
Somewhat Agree ☐ Strongly Agree

8. My ability to analyze research data was reinforced.

☐ Strongly Disagree ☐ Somewhat Disagree ☐ Neither Agree nor Disagree ☐
Somewhat Agree ☐ Strongly Agree

9. The importance of performing multiple replications in a research project was introduced.

☐ Strongly Disagree ☐ Somewhat Disagree ☐ Neither Agree nor Disagree ☐
Somewhat Agree ☐ Strongly Agree

10. My independence to complete laboratory procedures on my own was reinforced.

☐ Strongly Disagree ☐ Somewhat Disagree ☐ Neither Agree nor Disagree
☐ Somewhat Agree ☐ Strongly Agree

11. The provided laboratory instructions aided in my ability to adequately perform the exercise.

☐ Strongly Disagree ☐ Somewhat Disagree ☐ Neither Agree nor Disagree
☐ Somewhat Agree ☐ Strongly Agree

12. Participation in the laboratory exercises has improved my understanding of the overall process of conducting scientific research.

☐ Strongly Disagree ☐ Somewhat Disagree ☐ Neither Agree nor Disagree
☐ Somewhat Agree ☐ Strongly Agree

13. Participation in the laboratory exercises has increased my interest in a career in research (product development, academic research, etc.)

☐ Strongly Disagree ☐ Somewhat Disagree ☐ Neither Agree nor Disagree

☐ Somewhat Agree ☐ Strongly Agree

Food Processing Knowledge

14. Participating in the muffin laboratory exercise helped me to apply principles of food science to practical issues associated with food processing.

☐ Strongly Disagree ☐ Somewhat Disagree ☐ Neither Agree nor Disagree ☐

Somewhat Agree ☐ Strongly Agree

15. The effects of processing parameters (mixing time, baking time, baking temperature, etc.) on muffin quality were introduced.

☐ Strongly Disagree ☐ Somewhat Disagree ☐ Neither Agree nor Disagree

☐ Somewhat Agree ☐ Strongly Agree

16. My understanding of soybean processing (preparation, extraction, etc.) was improved.

☐ Strongly Disagree ☐ Somewhat Disagree ☐ Neither Agree nor Disagree

☐ Somewhat Agree ☐ Strongly Agree

17. My understanding of the advantages and disadvantages of adding soy ingredients to human foods was improved.

☐ Strongly Disagree ☐ Somewhat Disagree ☐ Neither Agree nor Disagree

☐ Somewhat Agree ☐ Strongly Agree

Appendix B - High School Baking with Protein Module Materials

Supplemental Material B1–High school baking with protein supporting material given to teachers

SUPPORTING INFORMATION

MUFFIN EVALUATION

External Qualities

Crust Color

Color should be a pleasing golden brown, not pale or burnt.

Volume

The volume is determined indirectly by measuring the circumference of a cross section of the muffin ($\pi r^2 \times \text{height}$) in cubic centimeters and dividing by the weight in grams. A more objective method uses a volumeter to determine the volume of a baked product by measuring the volume of seeds in a closed system with and without the addition of the baked product.¹²

Internal Qualities

Grain

Uniform thick-walled cells are desirable. Coarseness, thin-cell walls, uneven cell size and tunnels indicate poor grain.

Aroma

Aroma is recognized by the sense of smell. The aroma may be sweet, rich, musty, or flat. The ideal aroma should be pleasant, fresh, sweet, and natural. Sharp, bitter, or foreign aromas are undesirable.

Taste

An acceptable muffin should have a pleasingly sweet flavor. Flat, foreign, salty, soda, sour or bitter tastes or unpleasant aftertastes are undesirable.

Texture

Texture is determined by the sense of touch. Texture depends on the physical condition of the

crumb and is influenced by the grain. A desirable muffin should be easily broken, and slightly crumbly. Extreme crumbliness or toughness with lack of crumbling are undesirable characteristics.

Mouthfeel

Mouthfeel refers to the textural qualities perceived in the mouth. Characteristics can be described as gritty, hard, tough, tender, light and moist. A desirable muffin is tender, light, moist and requires minimal

MUFFIN INGREDIENTS AND PROCESSING STEPS

Author N. Cross*

*Adapted from Food Chemistry Workbook, edited by J.S. Smith and G. L. Christen, published and copyright Science Technology System, West Sacramento, California, 2002.

Table 1. Steps in baking and application principles

Selection and scaling of ingredients	Application Principles
Flour	gluten development, starch gelatinization, cell structure and volume, Maillard browning
Sugar	flavor, tenderizer, crust quality, moisture retaining, reduction of water activity, Maillard browning
Fat	flavor, tenderizer, moisturizing
Milk powder	binding effect on flour protein, flavor, crust color, Maillard browning, moisture retention
Whole eggs	protein coagulation, emulsification, flavor, color
Liquid	hydration of flour proteins and starch, solvent for salt, sugar, leavening agent, moisture in final baked product

Leavening agent	generation of carbon dioxide, volume, and cellular structure
Salt	flavor enhancer
Additional ingredients	variety in flavor, texture, and nutritive value
Processing steps	
Mixing	dispersion of ingredients, hydration of flour proteins and starch
Depositing	scaling of muffin
Baking	solubilization and activation of leavening agent, gelatinization of starch, coagulation of protein, caramelization of sugar, reduction of water activity, crumb development, color development, flavor development, crust formation, Maillard browning
Cooling	"setting" of structure, water evaporation
Packaging	retention of moisture, retention of flavor

SELECTION AND SCALING OF INGREDIENTS

Muffins made by large commercial bakeries are cake type muffins while those made in the home or small institutions are bread muffins. A common problem encountered in bread type muffins is tunnel formation resulting from over development of gluten. However, since cake type formulas are higher in sugar, fat, and soft wheat flours, (all ingredients that interfere with gluten development) this problem is avoided. The discussion that follows is limited to cake muffins.

The U.S. Dietary Guidelines¹⁴ promote healthy diets by increasing fiber while decreasing total fat, cholesterol, sugar, and sodium. Muffin formulas modified to meet the Guidelines utilize fat and sugar substitutes.

Flour

Flour is the primary ingredient in baked products. All-purpose flour is most commonly used in muffins. All-purpose flour is milled from hard or soft wheat or a mixture of hard and

soft wheat.⁶ Flour contains starch and the proteins, glutenin and gliadin which hold other ingredients together to provide structure to the final baked product. Hydration and heat promote gelatinization of starch, a process which breaks hydrogen bonds, resulting in swelling of the starch granule which gives the batter a more rigid structure.¹² Hydration and mixing promote the development of a gluten matrix that provides structure.¹²

Substituting whole wheat flour, wheat germ, rolled oats or bran for part of the all-purpose flour is an excellent way to increase fiber. Other flours used in muffins include cornmeal, soy, oat, potato, and peanut. An acceptable product is possible when these flours are substituted for one-third to one-half of all-purpose flour.¹² None of these flours contain glutenin or gliadin except whole wheat, and large pieces of bran in whole wheat flour cut and weaken gluten strands. Thus, there is minimal gluten development when these flours are used; the muffins tend to be crumbly and compact unless other modifications are made in the formula.

Sugar

Sugar contributes tenderness, crust color and moisture retention in addition to a sweet taste.³ Sucrose promotes tenderness by inhibiting hydration of flour proteins and starch gelatinization. Sugar is hygroscopic (attracts water) and maintains freshness. Corn syrup, molasses, maple sugar, fruit juice concentrates and honey are used as sweeteners for flavor variety. The quantity of liquid will need to be decreased if these sweeteners are used instead of sucrose because of the high-water content in these syrups.

Chemical changes in sugars during baking contribute characteristic flavors and browning. Carmelization of sugars involves inversion of sucrose, degradation of the ring structure, and creation of new polymers with distinctive flavors.¹² Maillard reactions (nonenzymatic browning) occur between the ketone or aldehyde group of reducing saccharides and amino acids in protein.⁶ Maillard reactions occur quickly at high temperatures and slowly at room temperature during periods of prolonged storage.¹² High fructose corn syrups and honey promote browning to a greater extent than sucrose due to the presence of high amounts of glucose and fructose.⁷ Maltose and lactose are reducing disaccharides and undergo browning but more slowly than monosaccharides.¹² Sucrose is a nonreducing disaccharide that requires hydrolysis and conversion to invert sugar (glucose plus fructose) for the Maillard reaction to occur. Water, an acidic pH, and heat facilitate sucrose hydrolysis. Heat also opens the ring structure to form an

aldehyde or ketone.¹²

An acceptable muffin is possible with reduction of sugar by one-third.¹⁵ Non-nutritive sweeteners such as saccharin can be substituted for all or part of the sugar. Aspartame (Equal®) converts to diketopiperzine when heated. Diketopiperzine does not have the sweetness of aspartame.⁷ However, aspartame has been successfully substituted for all of the sugar in muffins, probably because the baking time is relatively short.⁶ Non-nutritive sweeteners however, do not contribute to tenderness, browning, or moisture retention and other adjustments in the muffin formulation may become necessary.

Fat

Fat contributes tenderness, flavor, and a characteristic mouthfeel to baked products.⁵ Fat keeps the crumb and crust soft and helps retain moisture, and thus contributes to texture and mouthfeel. Because fat dissolves flavor components, it enhances the flavor of baked products¹⁰. Both shortening and vegetable oils are used in muffins.

To meet the demands of the consumer, muffin formulas are being modified to reduce fat and cholesterol. Low fat and fat-free muffins are available ready-to-eat and as frozen batters or dry mixes for bake-off. Baked products are acceptable when fat is reduced by one-third.⁸ Bread muffins made with 25% less fat or 2 tablespoons per 1 cup of flour were comparable to standard muffins in shape, tenderness, flavor, and texture.⁵ Bread muffins were considered acceptable when safflower oil (high in polyunsaturated fatty acids) was used in amounts typically used in the formula, or reduced by one-fourth to one-third.⁴

Various types of fat replacers being used in baked products include emulsifiers, complex carbohydrates, corn syrups, sugar alcohols, gums, and fruit based fat replacers.^{11,16} Emulsifiers (esters of edible fatty acids and polyols) improve mouthfeel, texture and shelf-life of baked products by binding water and trapping air.¹¹ Common emulsifiers are monoglycerides and diglycerides. Monoglycerides retard starch retrogradation and extend shelf life by forming stable complexes with amylose.² Examples are Dimodan or Amidan by Danisco.² Polydextrose (Litesse™) and sorbitol are examples of a complex carbohydrates and sugar alcohol respectively, that replace both fat and sugar. These help retain moisture and act as texturizers.¹⁶ Acceptable low fat cake muffins (5% fat) used 2% pregelatinized dull waxy starch (Amerimaize 2210,

American Maize-Products) and corn syrup (3.6%) to replace fat.⁸ Examples of gums are guar, locust bean, xanthan and sugar beet fiber¹². Gums enhance mouthfeel and texture and extend the shelf life of baked products ¹².

Fruit purees or pastes of raisins, prunes, dates, and grapes as well as applesauce are being promoted as fat replacers. Just Like ShortenTM is a mixture of dried prunes and apples.⁹ The fruit purees have humectant properties and promote tenderness and moistness and increase the shelf-life.¹¹ Sugar is reduced in formulas with fruit purees because of the natural sweetness of fruit.

Leavening Agents

Gases released by a leavening agent influence volume and cell structure. During baking, heat increases gas volume and pressure to expand cell size until proteins are coagulated.¹² Stretching of the cell walls during baking improves texture and promotes tenderness.¹²

Baking powder contains an acid salt plus sodium bicarbonate and an inert ingredient such as starch to retard the reaction of the ingredients during storage. Double-acting baking powder (most commonly used in muffins) contains both slow and fast-acting acids. SAS-phosphate contains monocalcium phosphate as the fast-acting acid plus sodium aluminum sulfate (SAS), a slow acting acid. Fast acting acids are readily soluble at room temperature while slow acting acids are less soluble and require heat over extended time to release carbon dioxide.¹³

The quantity of leavening used in a baked product depends on the choice of leavening agent and other ingredients. For each cup of flour, 1 1/2 teaspoons of double-acting baking powder (SAS) are needed for leavening, or 5% based on flour at 100%. Baking soda may be used in addition to baking powder when muffins contain acidic ingredients such as sour cream, yogurt, buttermilk, light sour cream, molasses and some fruits and fruit juices.⁶ One cup of an acidic ingredient such as yogurt will contain sufficient hydrogen ions for 1/2 teaspoon of baking soda to leaven 1 cup of flour. Baking powder is required for leavening the remaining amount of flour.¹⁹ Sodium carbonate is a product of an incomplete reaction in formulas with excess sodium bicarbonate. Excess sodium carbonate results in a muffin with a soapy, bitter flavor, and a yellow color because of the effect of an alkaline medium on the anthoxanthin pigments of flour.¹⁹ Also, too much baking powder or soda results in a muffin with a coarse texture and low volume because of

an over expansion of gas which causes the cell structure to weaken and collapse during baking. Inadequate amounts of baking powder will result in a compact muffin with low volume.

Whole eggs

Eggs provide flavor, color, and a source of liquid.³ Upon baking, the protein in egg white coagulates to provide structure. Lecithin in the yolk acts as an emulsifier and contributes to mouthfeel and keeping qualities. Modification of muffin formulas to reduce cholesterol may be accomplished by substituting an equivalent weight of egg whites or commercial egg substitute for whole eggs.

Nonfat dry milk powder

Milk powder is added to dry ingredients and water or fruit juice is used for liquid in muffin formulas.³ Milk powder binds flour protein to provide strength and body to muffins. In addition, milk powder adds flavor and retains moisture.³ The aldehyde group from lactose in milk combines with the amino group from protein upon heating, contributing to Maillard browning.

Sodium chloride

The function of sodium chloride is to enhance the flavor of other ingredients.³ Sodium chloride may be omitted from the formula without compromising flavor if other ingredients such as dried fruit or spices are added for flavor.

Liquids

Liquids perform several functions in baked products. These include dissolving dry ingredients, hydration of gluten forming proteins, gelatinization of starch and moistness in the final baked product.¹² Insufficient water results in muffins low in volume with possible pockets of unmoistened dry ingredients.⁶

Additional Ingredients

Other ingredients are often added to muffins for variety in flavor, texture, and color and to increase the content of fiber, protein, vitamins, and minerals. Typical added ingredients are fresh and dried fruit, nuts, coconut, shredded carrots and zucchini, corn, grated cheese, chopped ham or bacon. Added flavorings include cinnamon, nutmeg, allspice, cloves, and orange or lemon peel.

MIXING

The muffin method of mixing involves adding liquid ingredients to a well in the dry

ingredients and mixing using cutting or folding strokes until dry ingredients are moistened. Institutional or commercial bakeries use a mixer on slow speed for 3 to 5 minutes. Additional ingredients are added at the end of the mixing cycle or after depositing the muffin batter. Inadequate mixing results in a muffin with a low volume since some of the baking powder will be too dry to react completely.

DEPOSITING

The standard size of baked muffins is two ounces although one and four ounces are also common. For institutions or bakeries, small batter depositors are available that will deposit four muffins at a time. Also available are large piston type depositors that maintain accurate flow of the batter.³

BAKING

Many physical and chemical changes occur in the presence of heat to transform a liquid batter into a final baked muffin. Solubilization and activation of the leavening agent generates gases which expand to increase the volume of the muffin. Gelatinization of starch and coagulation of proteins provide permanent cell structure and crumb development. Caramelization of sugars and Maillard browning of proteins and reducing sugars promote browning of the crust. Reduced water activity facilitates Maillard Browning as well as crust hardening.

The choice of oven, baking pans and baking temperature influence the final baked product. A good flow of heat onto the bottom of the pan is necessary to produce a good product. Muffin tins are usually placed directly on the shelf or baking surface. The appropriate oven temperature is related to scaling and type of oven. Standard 2-ounce muffins are baked at 400° F (204°C) or slightly higher for a deck oven. Deck ovens may be stacked and are often used in small retail bakeries since these are cheaper and easier to maintain than reel or rotary ovens.¹⁰ Reel ovens consist of an insulated cubic compartment six or seven feet high. A Ferris wheel type mechanism inside the chamber moves 4 to 8 shelves in a circle, allowing each shelf to be brought to the door for adding or removing muffin tins from the shelves.¹⁰ The reel oven is often preferred by retail bakers since several hundred to several thousand pounds of baked product can be baked each day.¹⁰ Rack ovens may be stationary or the racks may be rotated during baking.

COOLING

Products should be cooled prior to wrapping. This allows the structure to "set" and reduces the formation of moisture condensation within the package. Condensed moisture creates an excellent medium for yeast, mold, and bacterial growth and spoilage.

WRAPPING

Muffins may be wrapped individually, in the tray in which they are baked, or transferred into plastic form trays for merchandizing.³ The shelf life of muffins is 3 to 5 days for wrapped muffins, and 4 to 7 days for those packaged for wholesale and wrapped in foil or plastic wrap. The storage life of muffins is significantly influenced by the water activity. Cake muffins have a longer shelf life than bread muffins because of the high sugar content and lower water activity. Added ingredients such as cheese, ham or dried fruits are high in sodium or sugar content which reduces water activity¹² and increases the shelf life.

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- ## COMMERCIAL ITEM DESCRIPTIONS

CIDs may be used during the procurement process by anyone who does institutional feeding. This includes the school lunch program, the military, hospitals, day cares, and many other facilities. The CID is used by these institutions to specify exactly what they want in the commercial product that they are procuring.

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Processing: The fresh or frozen muffins shall be prepared in accordance with good manufacturing practice.

Ingredients: The fresh or frozen muffins shall include enriched flour; sweetening agents; eggs; peanut, corn, soybean, cottonseed, or canola vegetable oils, vegetable shortening, or butter; water; salt; flavorings; leavening agents; and emulsifiers or other stabilizers. Fruit puree or sauce may be used to replace oil in Style B Reduced fat, Style C Low fat, and Style D Fat free prepared fresh or frozen muffins. Fruit addition must not change characteristic flavor of the muffin. The fresh or frozen muffins may include nonfat milk and/or whey. The fresh or frozen muffins shall include mold inhibitors of proper levels as allowed by the Federal Food, Drug, and Cosmetic Act.

Flour: The flour used for the muffins shall conform to the U.S. Standards for Enriched Flour (21 CFR 137.165).

Milk: The milk products used in the fresh or frozen muffins shall comply with all applicable requirements of the “Grade A Pasteurized Milk Ordinance - 1993 Recommendations of the United States Public Health Service” or latest revision thereof, or regulations substantially equivalent thereto.

Eggs: The eggs used in the fresh or frozen muffins shall conform to the applicable provisions of the Regulations Governing the Inspection of Eggs and Egg Products (7 CFR Part 59). The eggs shall be certified Salmonella free.

Finished product:

Appearance and color: The fresh or frozen muffin tops shall have a rounded pebbled surface and may be “sugar dusted”, “sugar crumb”, “sugar crumbled topping”, or “other appropriate topping”. The fresh or frozen muffin interiors shall have for example, a creamy white, slightly yellow, slightly blue, or caramel to moderate dark brown color, characteristic of the flavor of muffin. The fresh or frozen muffins shall be evenly baked without evidence of scorching

or burning. Color of the fresh or frozen muffin surface shall be typical of the type of muffin. There shall be no foreign color to the product.

Odor and flavor: The fresh or frozen muffins shall have an odor and flavor characteristic of the particular type of muffin. There shall be no foreign odors or flavors such as, but not limited to, burnt, scorched, stale, rancid, or moldy.

Texture: The texture of the fresh or frozen muffins shall have a slightly moist, light, and tender crumb. When the fresh or frozen muffins include nuts or fruits, there shall be an even distribution of nuts or fruit throughout the crumb. The fresh or frozen muffins shall not contain spots of unbaked flour on the bottom of the muffin.

Foreign material: All ingredients shall be clean, sound, wholesome, and free from evidence of rodent or insect infestation.

Age requirement: Unless otherwise specified in the solicitation, contract, or purchase order, the fresh muffins shall be delivered within 48 hours after baking. When frozen muffins are specified, the fresh product shall be in a freezer within 6 hours after baking and frozen to a temperature of 0EF (-17.8EC), \pm 5EF and shall be at a temperature not higher than 10EF (-12.2EC) within 6 hours after being placed in the freezer. Unless otherwise specified in the solicitation, contract, or purchase order, the frozen muffins shall be manufactured not more than 90 days prior to delivery and shall not have exceeded 10EF (-12.2EC) at any time during storage and delivery.

21 CFR §101.62 Nutrient content claims for fat, fatty acid, and cholesterol content of foods.

Fat content claims. The terms “fat free,” “free of fat,” “no fat,” “zero fat,” “without fat,” “negligible source of fat,” or “dietarily insignificant source of fat” or, in the case of milk products, “skim” may be used on the label or in labeling of foods, provided that:

1. The food contains less than 0.5 gram (g) of fat per reference amount customarily consumed and per labeled serving or, in the case of a meal product or main dish product, less than 0.5 g of fat per labeled serving; and

2. The food contains no added ingredient that is a fat or is generally understood by consumers to contain fat unless the listing of the ingredient in the ingredient statement is followed by an asterisk that refers to the statement below the list of ingredients, which states “adds a trivial amount of fat,” “adds a negligible amount of fat,” or “adds a dietarily insignificant amount of fat;” and
3. As required in §101.13(e)(2), if the food meets these conditions without the benefit of special processing, alteration, formulation, or reformulation to lower fat content, it is labeled to disclose that fat is not usually present in the food (e.g., “broccoli, a fat free food”).

The terms “low fat,” “low in fat,” “contains a small amount of fat,” “low source of fat,” or “little fat” may be used on the label or in labeling of foods, provided that:

1. The food has a reference amount customarily consumed greater than 30 g or greater than 2 tablespoons and contains 3 g or less of fat per reference amount customarily consumed; or
2. The food has a reference amount customarily consumed of 30 g or less or 2 tablespoons or less and contains 3 g or less of fat per reference amount customarily consumed and per 50 g of food (for dehydrated foods that must be reconstituted before typical consumption with water or a diluent containing an insignificant amount, as defined in §101.9(f)(1), of all nutrients per reference amount customarily consumed, the per 50-g criterion refers to the “as prepared” form); and
3. If the food meets these conditions without the benefit of special processing, alteration, formulation, or reformulation to lower fat content, it is labeled to clearly refer to all foods of its type and not merely to the particular brand to which the label attaches (e.g., “frozen perch, a low fat food”).

The terms “reduced fat,” “reduced in fat,” “fat reduced,” “less fat,” “lower fat,” or “lower in fat” may be used on the label or in the labeling of foods, provided that:

1. The food contains at least 25 percent less fat per reference amount customarily consumed than an appropriate reference food.

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Supplemental Material B2–High School baking with protein lecture slides

Baking with Added Protein

Developed by: Jarrise Brown, Melly Gentry, Ph.D.

KANSAS STATE UNIVERSITY | Food Science Institute

1

Challenge: How many bakery items can you name?



2

What are some reasons we eat grain-based foods?

3

What are some reasons we eat grain-based foods?

- Provide energy
- Provide nutrition
- Enjoy a tasty snack
- Celebrate a special occasion

4

What are some reasons we eat grain-based foods?

- Provide energy
- Provide nutrition
- Enjoy a tasty snack
- Celebrate a special occasion

Did you know?
You have been eating grains all day long. Grains make up 48% of the food we eat. Grains are a source of energy and are a healthy part of your diet.

5

Grain-based foods are part of a well-balanced diet...BUT we need to eat other foods to meet our full nutrition needs.



High In:	Low In:
• Calories	• Protein
• Fiber (whole grains only)	• Other vitamins (A, C, D, E, K)
• B Vitamins	• Other minerals
• Iron	

6

Which snack would you prefer?



The problem:
Healthy versions of foods sometimes don't taste as good.

The solution:
Food Scientists are working to create Ingredients that are healthy AND tasty.

Have you tried the Impossible Burger?



Have you tried bacon bits?
(The crunchy kind)




10

Both of these foods are made from soybeans!



11

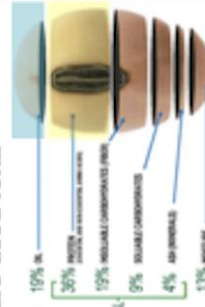
Soybeans are made of...



Component	Percentage
Oil	19%
Protein	36%
Meal	19%
Insoluble Carbohydrates	9%
Soluble Carbohydrates	4%
Cellulose	13%

12

Soybeans are used for...



- Soybean oil
- Soy protein

13

Soy Protein Powder → Extract Protein

- Grind soybeans
- 90% Protein
- Flavor compounds left behind
- Can add to:
 - Protein shake mixes
 - Protein bars
 - Breakfast cereal



14

Protein in our diet

- Build and repair bones, muscle, cartilage, skin, and blood
- Help our body make enzymes and hormones
- Helps with satiation (feeling full)
- Males ages 14-18: 52g
- Females ages 14-18: 46g



15

Lab Activity: Protein Muffins



- Typical protein sources in muffins:
 - Milk
 - Eggs
 - Flour
- We will experiment with adding soy protein powder to increase the protein

16

Protein Muffins

- How much protein powder can we add?
- How will the added protein affect the muffins:
 - Taste?
 - Size?
 - Color?
- What compromises are people willing to make for healthier food?

17

Baking with Added Protein Lab Slides

Developed by: Jesse Brown, Kelly Getty, Ph.D.

KANSAS STATE UNIVERSITY | Food Science Institute

18

Crumb Evaluation Examples



Crust Evaluation Examples



**Supplemental Material B3–High school baking with protein laboratory
handout, gravimetric measurements**

Muffins with Added Protein

Muffin Types:

- 5) Control: No soy protein
- 6) Low Soy: 10% of the flour replaced with soy protein powder
- 7) High Soy: 20% of the flour replaced with soy protein powder

Circle your group's muffin type: Control Low Soy High Soy

Formulas:

	Control	Low Soy	High Soy
Ingredient	Amount (g)	Amount (g)	Amount (g)
AP Flour	140	126	112
Cornstarch	14	14	14
Granulated sugar	88	88	88
Baking powder	5.4	5.4	5.4
Salt	2.6	2.6	2.6
Non-fat dry milk	21.6	21.6	21.6
Whole dry egg	11	11	11
Shortening	100	100	100
Water	231	231	231
Vanilla	2	2	2
Soy protein isolate	0	14	28

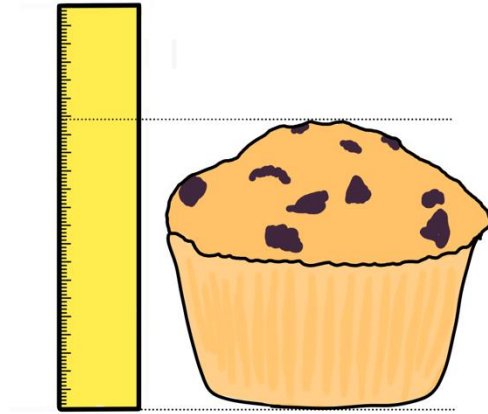
Baking Instructions:

- 23) Preheat oven to 375°F (190.6°C). Line a muffin tin with 12 paper muffin cups.
- 24) In a large mixing bowl combine all ingredients except water and vanilla and mix for 1 minute on speed 1. Scrape the sides of the bowl.
- 25) Add approximately half of the water and mix for 30 seconds on speed 1 on a KitchenAid mixer (low speed if handheld mixer). Scrape the sides of the bowl.
- 26) Add the rest of the water and vanilla and mix for 30 seconds on speed 1 on a KitchenAid mixer (low speed if handheld mixer). Scrape the sides of the bowl.
- 27) Mix on Speed 3 on a KitchenAid mixer (medium speed if handheld mixer) for 2 minutes.
- 28) Fill the muffin tins with a large scoop, or about $\frac{3}{4}$ full (see example below).



- 29) Bake the muffins for about 23 minutes, or until a toothpick inserted in the center comes out clean (from a food safety standpoint, baking at 190.6°C for at least 17 minutes will kill 99.999% of *Salmonella* bacteria).
- 30) Remove muffins from oven and let cool completely (approximately 10 minutes).
- 31) Record the weight of each muffin in **Table 1**.

- 32) Use a ruler to measure the height from the bottom to the tallest point on the muffin and record in **Table 2**. Lay an index card or other straight edge on top of



the muffin to help measure the height.

- 33) Exchange data with other groups to complete **Table 3**.
- 34) Evaluate all 3 muffin types using the provided muffin scorecard.

Circle your group's muffin type: Control Low Soy High Soy

Table 1: Your group's muffin weight

Muffin #	Weight Units: _____
1	
2	
3	
4	
5	
6	
7	
8	
9	
10	
11	
12	
Average	

To find the average muffin weight:

Add all the muffin weights together: _____

Count how many muffins you made: _____

Divide the sum of all muffin weights by how many muffins you made: _____

$$\text{Average Muffin Height} = \frac{\text{Sum of all muffin weights}}{\text{Number of muffins made}}$$

Table 2: Your group's muffin height

Muffin #	Height Units: _____
1	
2	
3	
4	
5	
6	
7	
8	
9	
10	
11	
12	
Average	

Table 3: Class muffin data (get from other groups)

Variation	Average Muffin Weight	Average Muffin Height
Control		
Low Soy		
High Soy		

SCORECARD FOR MUFFINS¹

	Control	Low Soy	High Soy
External Qualities			
<u>Volume</u> 1=low volume 2= slightly too small 3=just the right size 4=slightly too large 5=much too large			
<u>Shape</u> 1=absolutely flat 2=somewhat rounded 3=pleasingly rounded 4=somewhat pointed 5=very pointed			
<u>Crust color</u> 1=much too pale 2=somewhat pale 3=pleasingly golden brown 4=somewhat too brown 5=much too brown			
Internal Qualities (cut the muffin in half from top to bottom to have a cross-section for evaluation)			
<u>Interior color</u> 1=much too white 2=somewhat white 3=pleasingly creamy 4= somewhat too yellow 5=much too yellow			
<u>Texture</u> 1=extremely crumbly, falls apart 2=somewhat crumbly 3=fluffy, moist, does not fall apart 4=slightly tough 5=tough, chewy			
<u>Flavor</u> 1=absolutely not sweet enough 2=not nearly sweet enough 3=pleasingly sweet 4=somewhat too sweet 5=much too sweet			
<u>Aftertaste</u> 1=extremely distinct 2=very noticeable 3=somewhat noticeable			

4=slight aftertaste 5=no aftertaste			
<u>Overall acceptability</u> 1=very unacceptable 2=somewhat unacceptable 3=somewhat acceptable 4=acceptable 5=very acceptable			

Adapted from McWilliams 2001a.

Post Lab Questions:

- 1) Which muffin variation had the highest score for appearance?

- 2) Which muffin variation has the highest score for taste?

- 3) Which muffin variation had the highest score for texture?

- 4) Are you willing to give up any quality attributes in exchange for a more nutritious muffin? Who might be willing to give up some quality attributes to get a healthier product?

- 5) Imagine you are working for a bakery that is looking to expand their product line to include a protein muffin. Which of the soy muffin types would you recommend and why? What type of customer should the business market this new product to?

Supplemental Material B4—High school baking with protein laboratory
handout, volumetric measurements
Muffins with Added Protein

Muffin types:

- 8) Control: No soy protein
- 9) Low Soy: 10% of the flour replaced with soy protein powder
- 10) High Soy: 20% of the flour replaced with soy protein powder

Circle your group's muffin type: Control Low Soy High Soy

Formulas:

	Control	Low Soy	High Soy
Ingredient	Amount	Amount	Amount
AP Flour	1 cup	1 cup	1 cup
Cornstarch	2 Tablespoons	2 Tablespoons	2 Tablespoons
Granulated sugar	1/2 Cup	1/2 Cup	1/2 Cup
Baking powder	1 1/2 teaspoons	1 1/2 teaspoons	1 1/2 teaspoons
Salt	1/2 teaspoon	1/2 teaspoon	1/2 teaspoon
Non-fat dry milk	3 Tablespoons	3 Tablespoons	3 Tablespoons
Whole dry egg	2 Tablespoons	2 Tablespoons	2 Tablespoons
Shortening	1/2 Cup	1/2 Cup	1/2 Cup
Water	1 Cup	1 Cup	1 Cup
Vanilla	1 teaspoon	1 teaspoon	1 teaspoon

Soy protein isolate	-	3 Tablespoons	5 Tablespoons
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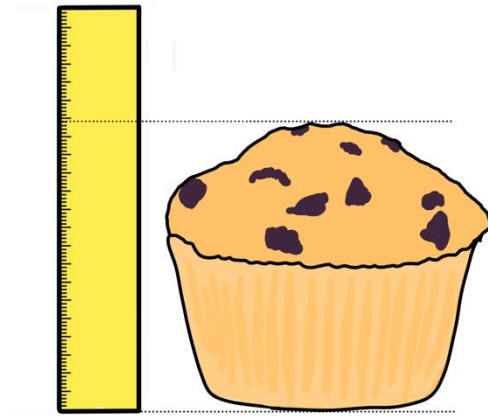
Baking Instructions:

- 35) Preheat oven to 375°F (190.6°C). Line a muffin tin with 12 paper muffin cups.
- 36) In a large mixing bowl combine all ingredients except water and vanilla and mix for 1 minute on speed 1. Scrape the sides of the bowl.
- 37) Add approximately half of the water and mix for 30 seconds on speed 1 on a KitchenAid mixer (low speed if handheld mixer). Scrape the sides of the bowl.
- 38) Add the rest of the water and vanilla and mix for 30 seconds on speed 1 on a KitchenAid mixer (low speed if handheld mixer). Scrape the sides of the bowl.
- 39) Mix on Speed 3 on a KitchenAid mixer (medium speed if handheld mixer) for 2 minutes.
- 40) Fill the muffin tins with a large scoop, or about $\frac{3}{4}$ full (see example below).



- 41) Bake the muffins for about 23 minutes, or until a toothpick inserted in the center comes out clean (from a food safety standpoint, baking at 190.6°C for at least 17 minutes will kill 99.999% of *Salmonella* bacteria).
- 42) Remove muffins from oven and let cool completely (approximately 10 minutes).

- 43) Use a ruler to measure the height from the bottom to the tallest point on the muffin and record in **Table 1**. Lay an index card or other straight edge on top of



the muffin to help measure the height.

- 44) Exchange data with other groups to complete **Table 2**.
- 45) Evaluate all 3 muffin types using the provided muffin scorecard.

Circle your group's muffin type: Control

Low Soy

High Soy

Table 1: Your Group's Muffin Data

Muffin #	Height Units: _____
1	
2	
3	
4	
5	
6	
7	
8	
9	
10	
11	
12	
Average	

To find the average muffin height:

Add all the muffin heights together: _____

Count how many muffins you made: _____

Divide the sum of all muffin heights by how many muffins you made: _____

$$\text{Average Muffin Height} = \frac{\text{Sum of all muffin heights}}{\text{Number of muffins made}}$$

Table 2: Class Muffin Data (get from other groups)

Variation	Average Muffin Height
Control	
Low Soy	
High Soy	

Muffin Scorecard¹

	Control	Low Soy	High Soy
External Qualities			
<u>Volume</u> 1=low volume 2= slightly too small 3=just the right size 4=slightly too large 5=much too large			
<u>Shape</u> 1=absolutely flat 2=somewhat rounded 3=pleasingly rounded 4=somewhat pointed 5=very pointed			
<u>Crust color</u> 1=much too pale 2=somewhat pale 3=pleasingly golden brown 4=somewhat too brown 5=much too brown			
Internal Qualities (cut the muffin in half from top to bottom to have a cross-section for evaluation)			
<u>Interior color</u> 1=much too white 2=somewhat white 3=pleasingly creamy 4= somewhat too yellow 5=much too yellow			
<u>Texture</u> 1=extremely crumbly, falls apart 2=somewhat crumbly 3=fluffy, moist, does not fall apart 4=slightly tough 5=tough, chewy			
<u>Flavor</u> 1=absolutely not sweet enough 2=not nearly sweet enough 3=pleasingly sweet 4=somewhat too sweet 5=much too sweet			
<u>Aftertaste</u> 1=extremely distinct 2=very noticeable 3=somewhat noticeable			

4=slight aftertaste 5=no aftertaste			
<u>Overall acceptability</u> 1=very unacceptable 2=somewhat unacceptable 3=somewhat acceptable 4=acceptable 5=very acceptable			

¹Adapted from McWilliams 2001a.

Post Lab Questions:

- 1) Which muffin variation had the highest score for appearance?

- 2) Which muffin variation has the highest score for taste?

- 3) Which muffin variation had the highest score for texture?

- 4) Are you willing to give up any quality attributes in exchange for a more nutritious muffin? Who might be willing to give up some quality attributes to get a healthier product?

- 5) Imagine you are working for a bakery that is looking to expand their product line to include a protein muffin. Which of the soy muffin types would you recommend and why? What type of customer should the business market this new product to?

**Supplemental Material B5–High school baking with protein quiz and short
essay questions**

Baking with Protein Quiz Questions

1. Soybeans are processed for what ingredient?
 - a. Oil
 - b. Protein
 - c. Both oil and protein**
 - d. Carbohydrates

2. What role does protein serve in our bodies?
 - a. Build muscle
 - b. Signals fullness
 - c. Make digestive enzymes
 - d. All of the above**

3. **True** or False: Milk and eggs are a good source of protein.

4. Which of the following is NOT a grain-based food?
 - a. French fries**
 - b. Sandwich bread
 - c. Rice Krispies Treat
 - d. Bagel

5. Whole-grain foods are low in _____ compared to other food groups.
 - a. Calories
 - b. Protein**
 - c. B Vitamins
 - d. Fiber

6. Imitation meat products like the Impossible Whopper from Burger King can be made from what ingredient:

- a. Carrots
 - b. Potatoes
 - c. Soybeans**
 - d. Eggplant
7. True or **False**: When soy protein powder is produced, the powder contains many flavor compounds.
8. What oil is actually made of soybeans?
- a. Canola oil
 - b. Vegetable Oil**
 - c. Lard
 - d. Palm Oil
9. About how much protein should the average person eat in a day?
- a. 5g
 - b. 10g
 - c. 50g**
 - d. 100g
10. What products can protein powder be added to in order to boost the protein content?
- a. Energy bars
 - b. Breakfast cereal
 - c. Post-workout shakes
 - d. All of the above**

Open-Response Question:

What are the benefits of foods with added protein? Are there any downsides? Are you personally interested in consuming products with added protein?

Supplemental Material B6–High school baking with protein IRB exempt letter



TO: Kelly Getty
Animal Sciences & Industry
Manhattan, KS 66506

Proposal Number: IRB-10709

FROM: Rick Scheidt, Chair
Committee on Research Involving Human Subjects

DATE: 02/08/2022

RE: Proposal Entitled, "Development of Food Science Laboratory Exercises Utilizing Soybean Protein Extracts for Food Science Secondary Education Classrooms."

The Committee on Research Involving Human Subjects / Institutional Review Board (IRB) for Kansas State University has reviewed the proposal identified above and has determined that it is EXEMPT from further IRB review. This exemption applies only to the proposal - as written – and currently on file with the IRB. Any change potentially affecting human subjects must be approved by the IRB prior to implementation and may disqualify the proposal from exemption.

Based upon information provided to the IRB, this activity is exempt under the criteria set forth in the Federal Policy for the Protection of Human Subjects, **45 CFR §104(d), category:Exempt Category 1.**

Certain research is exempt from the requirements of HHS/OHRP regulations. A determination that research is exempt does not imply that investigators have no ethical responsibilities to subjects in such research; it means only that the regulatory requirements related to IRB review, informed consent, and assurance of compliance do not apply to the research.

Any unanticipated problems involving risk to subjects or to others must be reported immediately to the Chair of the Committee on Research Involving Human Subjects, the University Research Compliance Office, and if the subjects are KSU students, to the Director of the Student Health Center.

Electronically signed by Rick Scheidt on 02/09/2022 11:09 AM ET

Supplemental Material B7–High school baking with protein IRB Informed consent



Institutional Review Board (IRB) Informed Consent Template Form

comply@k-state.edu | 785-532-3224

PROJECT TITLE:

Development of Food Science and Baking Science Laboratory Exercises Utilizing Soybean Protein Extracts

PROJECT APPROVAL DATE:

02/08/2022

PROJECT EXPIRATION DATE:

N/A

LENGTH OF STUDY:

15
minutes

PRINCIPAL INVESTIGATOR: Dr. Kelly Getty

CO-INVESTIGATOR(S): Janae Brown

CONTACT DETAILS FOR PROBLEMS/QUESTIONS: Dr. Kelly Getty (785) 532-2203
Janae Brown (479) 685- 8252

IRB CHAIR CONTACT INFORMATION:

For the subject should he/she have questions or wish to discuss on any aspect of the research with an official of the university or the IRB. These are: Rick Scheidt, Chair, Committee on Research Involving Human Subjects, 203 Fairchild Hall, Kansas State University, Manhattan, KS 66506, (785) 532-3224; Cheryl Doerr, Associate Vice President for Research Compliance, 203 Fairchild Hall, Kansas State University, Manhattan, KS 66506, (785) 532-3224

PROJECT SPONSOR: Kansas Soybean Commission

PURPOSE OF THE RESEARCH:

The objective of this study is to develop educational modules about soybean products and assess the effect on Secondary Education students' perception and understanding of soybean utilization in human foods.

PROCEDURES OR METHODS TO BE USED:

Kansas Secondary Education teachers will indicate interest in receiving materials for teaching a module about baking with soy protein. Materials will be distributed to interested teachers via email. A voluntary, anonymous survey to assess the module will be distributed to teachers that received module materials.

RISKS OR DISCOMFORTS ANTICIPATED:

There are no known risks of participation.

BENEFITS ANTICIPATED:

Participants will be acknowledged of their time via an online message upon completing the survey. Additionally, participants who complete the survey will be entered into a drawing to win a gift card.

EXTENT OF CONFIDENTIALITY:

Quantitative responses will be aggregated. Direct quotes may be used; however, no identifying information will be used.

IS COMPENSATION OR MEDICAL TREATMENT AVAILABLE IF INJURY OCCURS? N/A

Terms of participation: I understand this project is research, and that my participation is voluntary. I also understand that if I decide to participate in this study, I may withdraw my consent at any time, and stop participating at any time without explanation, penalty, or loss of benefits, or academic standing to which I may otherwise be entitled.

By clicking the button below, you acknowledge that your participate is voluntary, you are 18 years of age, and that you are aware that you may choose to terminate your participation in the study at any time and for any reason.

☐ I consent, begin the study

☐ I do not consent, I do not wish to participate

Supplemental Material B8—High school baking with protein IRB debriefing statement

IRB Debriefing Statement

Thank you for participating in our survey. Your responses are critical to improving this laboratory exercise and learning experience for future students enrolled in Food Science and Baking Science courses. All data collected in this survey will remain confidential and anonymous when discussed in peer reviewed journal publications.

If you are interested in receiving a short report of the results, please email jeb98@ksu.edu and provide an email address where the report may be sent. Again, thank you for your time and assistance in completing this research.

Sincerely,
Janae Brown
M.S. Graduate Student
Food Science Institute Kansas State University
jeb98@ksu.edu

Kelly Getty, Ph.D.
Associate Professor
Food Science Institute Kansas State University
kgetty@ksu.edu

☐ Yes, I would like to receive a short report of the results collected from this survey. Email address:

Supplemental Material B9–High school baking with protein survey

Q2 Approximately how many students participated in the module?

Q3 How many class sections did you use the module in?

Q4 What class did you teach the module in?

Q5 What grade level(s) is the class? Check all that apply.

☐

6th grade (4)

☐

7th grade (5)

☐

8th grade (6)

☐

9th grade (7)

☐

10th grade (8)

☐

11th grade (9)

☐

12th grade (10)

Q6 Which of the following materials did you use in your class?

- ☐ Powerpoint Slides
- ☐ Google Slides with Slido Interactions
- ☐ Laboratory Instructions Handout
- ☐ Muffin Scorecard
- ☐ Quiz Questions
- ☐ Supplemental Information for Teachers

Q7 I was able to use the module in my classroom with minimal additional preparation.

- ☐ Strongly disagree (1)
- ☐ Somewhat disagree (2)
- ☐ Neither agree nor disagree (3)
- ☐ Somewhat agree (4)
- ☐ Strongly agree (5)

Q8 The provided laboratory instructions were clear and helpful for students performing the exercise.

- ☐ Strongly disagree (1)
- ☐ Somewhat disagree (2)
- ☐ Neither agree nor disagree (3)
- ☐ Somewhat agree (4)
- ☐ Strongly agree (5)

Q9 The module was on-level for the students experience with lab/kitchen skills.

- ☐ Strongly disagree (1)
- ☐ Somewhat disagree (2)
- ☐ Neither agree nor disagree (3)
- ☐ Somewhat agree (4)
- ☐ Strongly agree (5)

Q10 The muffin scorecard was a useful method for students to evaluate muffin quality.

- ☐ Strongly disagree (1)
- ☐ Somewhat disagree (2)
- ☐ Neither agree nor disagree (3)
- ☐ Somewhat agree (4)
- ☐ Strongly agree (5)

Q11 The laboratory exercise increased students' knowledge about soybean products.

- ☐ Strongly disagree (1)
- ☐ Somewhat disagree (2)
- ☐ Neither agree nor disagree (3)
- ☐ Somewhat agree (4)
- ☐ Strongly agree (5)

Q12 The module engaged students.

- ☐ Strongly disagree (1)
- ☐ Somewhat disagree (2)
- ☐ Neither agree nor disagree (3)
- ☐ Somewhat agree (4)
- ☐ Strongly agree (5)

Q13 The laboratory exercise was effective for teaching students about the advantages of and barriers to adding soy ingredients to foods.

- ☐ Strongly disagree (1)
- ☐ Somewhat disagree (2)
- ☐ Neither agree nor disagree (3)
- ☐ Somewhat agree (4)
- ☐ Strongly agree (5)

Q14 Given the opportunity would you teach this module or a similar module again?

- ☐ Yes
- ☐ Yes, with changes (please elaborate in Q15)
- ☐ No

Q15 Do you have any comments to share about the module or any suggestions for improvement?

Q16 Were any particularly notable comments made by students during the laboratory?

Q17 Would you like to be entered in a drawing to win an Amazon gift card?

☐ Yes

☐ No