# EVALUATION OF EXTRACTION METHODS AND GROAT TYPE WITH EFFECTS ON

## QUALITY ANALYSIS OF OAT BEVERAGE

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Brandon Robert Olson

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## Title

# EVALUATION OF EXTRACTION METHODS AND GROAT TYPE WITH EFFECTS ON QUALITY ANALYSIS OF OAT BEVERAGE

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Brandon Robert Olson

The Supervisory Committee certifies that this disquisition complies with North Dakota

State University's regulations and meets the accepted standards for the degree of

## MASTER OF SCIENCE

SUPERVISORY COMMITTEE:

Dr. Senay Simsek

Chair

Dr. Julie Garden-Robinson

Dr. Jae-Bom Ohm

Dr. Anuradha Vegi

Approved:

11-29-2021

Date

**Richard Horsley** 

Department Chair

## ABSTRACT

The recent uptick in consumption of plant-based beverages has forced companies to modify production methods to further optimize the process. The objective of this study was to evaluate how groat type, water to grain ratio, and extraction method affect the quality and composition of oat beverage. Wet milling and the use of an amylase treatment produced significantly (p<0.05) higher values of degree Brix, solids, and total starch within the beverage base. Stabilized groats produced the highest value of total starch of 61.97%. Microbial analysis revealed that amylase treated products had a much lower colony forming units per gram (CFU/g), when compared to dry and wet milling. To produce an oat beverage with ideal rheological and composition values, the beverage must be derived from heat treated groats, a grain to water ratio of 1:4, and must undergo an  $\alpha$ -amylase treatment.

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## **1. INTRODUCTION**

Dairy milk has been a staple in diets worldwide because it is an excellent source of calcium and vitamin D; it can also be used in cooking and baking. In the past decade or so, there has been a considerable uptick in plant-based beverage consumption. Plant-based beverages can be processed from hemp seeds, almonds, soy, and even flax. The most prominent options right now are almond and oat beverage. Oat beverage is popular because it emulates the flavor and versatility of bovine milk. A common slight against oat beverage is that it can sometimes have a "slimy" mouthfeel. The undesired slimy mouthfeel is caused by complex sugars that were not broken down entirely after enzyme treatment.

The amylase family consists of enzymes that breakdown complex starches into simple sugars, like glucose and maltose. A proper enzyme treatment is pivotal for a quality oat beverage. The production of oat beverage is substantially lower in emissions, land use, and water use when compared to dairy milk and other plant-based beverages (Poore, & Nemecek, 2018). Oats are high in  $\beta$ -glucan, which has many functions in food systems and health. The  $\beta$ -glucan found in the oats are considered dietary fiber and can yield many positive benefits upon consumption. These benefits include lowering cholesterol and maintain a healthy glycemic index. It was concluded that  $\beta$ -glucan can reduce risk of metabolic syndrome. Metabolic syndrome occurs when the body is affected by a cluster of conditions that increase the risk for heart attack, stroke, or type 2 diabetes (Wood, 2007; Bruce & Hansen, 2010).

The purpose of this experiment was to analyze how groat type, water to grain ratio, and extraction method affect the physical characteristics and composition of the oat beverage. The groat types examined were stabilized, non-stabilized, and rolled. Water to grain ratios of 1:4 and 1:6 was also assessed, as well as the extraction method: Dry milling, wet milling, and amylase

treatment. Once extracted, the beverage base was separated from the solid residues. Subsamples of the liquid beverage base was sent to Dr. Teresa Bergholz at Michigan State University. Both sample types underwent analysis of physical characteristics, such a moisture, ash, solids, and total starch. Evaluation of proximate composition of both sample types were also conducted (i.e., sugar composition, and  $\beta$ -glucan content). Physical characteristics of the raw groat type was also conducted, which included analysis of ash, moisture, dietary fiber and protein.

With a better understanding of how production methods and parameters affect processing, it will be easier to develop an oat beverage that so closely emulates dairy milk that the average consumer would be inclined to purchase. Applying modern food ingredient technologies, such as emulsifiers and texture stabilizers will be a focus in future research.

#### **2. LITERATURE REVIEW**

## 2.1. Oats & groats

Common oat, *Avena sativa*, is cultivated for its seed (Clemens, & Klinken, 2014). Oats are best grown in regions where summers are cool and wet. Unlike other cereal grains, the oat has a higher tolerance for cold weather and heavier rain. The oat plant is cultivated annually and can be planted in either fall or spring. Russia is the leading producer of oats globally, followed by Canada, then Poland (Clemens, & Klinken, 2014).

Oats have numerous uses; usually, they are rolled or crushed for oatmeal or milled into oat flour. More recently, oats have been used in plant-based milk substitutes. A serving of oats is high in carbohydrates and protein. Oat is also rich in soluble fiber, Vitamin B1, Iron, and Manganese (Clemens, & Klinken, 2014). Soluble fiber is essential for a healthy diet and can also lower your cholesterol. The predominant type of soluble fiber in oats is ß-glucans.

The oat is cultivated for its seed/kernel. The oat's seed comprises the hull, bran layer, endosperm, and germ. The seed's starch is found in the endosperm, and this is the fuel that helps the seed grow (Figure 1). In oat milk production, the endosperm is broken down into simple sugars, which add flavor and directly impact the product's texture.



Figure 1. Structural representation of the oat grain presenting different oat tissues Reprinted without changes from (Grundy et al., 2018)

For oat milk production, the endosperm is the essential part of the seed. The bran is high in fiber and rich in protein. The hull is often removed before production due to implications during processing and can be used as cattle feed, food fillers, and even fertilizer (Clemens, & Klinken, 2014). Like most cereal grains, all parts of the seed can be utilized in some way.

## 2.2. Oat beverage production

## 2.2.1. Industrial scale processing methods

The elementary process requires just water, oats or oat flour, and a blender. An initial blending step is done to mix the oats and water, next just strain to separate the insoluble fibers and chill the beverage. Although primitive, this process yields basic oat beverage with a mild sweetness, and moderate mouth feel (Sethi et al., 2016). Although the process of producing oat milk is relatively simple, the process is still evolving and being optimized.

It can be seen when evaluating today's methods of industrial production, that the basic process is the same but numerous steps have been added. The initial step is to soak the groats in water for at least 12 hours and blend the mixture to create a slurry. This step is also referred to as wet milling (Decker et al., 2014). Once the slurry is prepared, the mixture is heated to roughly 60°C and transferred to a saccharification tank. The heated slurry is then treated with enzymes to break down the oat starches into smaller components. The most common enzyme used in this step is  $\alpha$ -amylase. The mixture will undergo enzyme treatment for 1-2 hours and heated to at least 90°C for 1-3 minutes to deactivate the  $\alpha$ -amylase (Salama et al., 2011). The next step requires for the insoluble fibers to be separated from the oat beverage base. Many different methods can be utilized to accomplish this task, the most common include centrifugation and decanting. Straining can also be done instead but the process is quite inefficient on an industrial scale.

Homogenization needs to occur next. Recently, ultra-high-pressure homogenization (UHPH) has become a popular method. This method pressurizes the base and forces it through a small nozzle. The enzymes and microbes are terminated because of the shear force created from passing through the nozzle (Levy et al., 2020). Another benefit of this method is that it acts as a kill step for enzymes, microbes, and homogenizes the base. Having the ability to combine the two processing steps could save time and money during production. After separation and homogenization, the remaining liquid is considered the 'oat base' and is ready for formulation and/or fortification. In some instances, prior to formulation some companies will conduct heat processing or pasteurization to further ensure a safe product (Salama et al., 2011). During the formulation process ingredients are added to the base that increase the nutrition value of the product, while also giving the possibility of different flavors such as strawberry and chocolate.

Once the product is formulated the beverage is stored in a sterile tank and awaits packaging and shipping.

## **2.3.** Heat treatment of groats

The heat treating of groats is essential to the production of high-quality oat beverage. A reason heat treating is done is to inactivate endogenous enzymes, like lipase, to help ensure a longer shelf life and to prevent rancidity during storage (Zhang et al., 2021). Another reason is to produce a product that the consumers can prepare easily, such as ready to eat (RTE) products. A heat-treating step also partially gelatinizes the starch, thus making more starch available during enzyme treatment. It is important to monitor the temperature, moisture, and duration of heating during the heat treatment. Overheating can lead to reduced functionality and destruction of nutrition structure (Zhang et al., 2021).

The use of a kiln is a popular option for heat treatment. Using a kiln will inhibit lipase, prevent rancidity, and impart a pleasant caramel aroma. Dry kilning is executed at 100°C for at least two hours (Schlosser & Mitzkat, 2019). This production method closely resembles methods of malting barley to produce beer. Again, the temperature, moisture, and duration of kilning is pivotal to the quality of the product. The kiln is utilized by large scale productions, due to its ease of use and the ability to process large batches.

Other traditional heating methods include boiling, steaming, and autoclaving. All these methods are considered hydrothermal because it requires heat and moisture. The use of hydrothermal heat treatment also gelatinizes the starch at a higher rate (Chang et al., 2015). The use of these methods is common on small scale productions, due to running costs and resources.

Another method of heat treatment often used is extrusion. The use of an extruder, high heat, pressure, and shear forces enhances starch gelatinization and modifies starch structure

(Riaz, 2021). This method is commonly used to produce RTE foods. A study found that the use of an extruder at 18% moisture, at 155°C, with a screw speed of 150 revolutions per minute (rpm) produced flour with much higher degree gelatinization, which increases the release of glucose into the system (Wang et al., 2019).

#### 2.4. Enzymes used in oat beverage production

## 2.4.1. Amylase

The most critical ingredient in the production of oat beverage is the enzyme. Normally,  $\alpha$ -amylase, and/or  $\beta$ -amylase are the enzymes chosen for oat milk production. The  $\alpha$ -amylase catalyzes the hydrolysis of starch into simple sugars and limit-dextrins (Deswal et al., 2013). Without this treatment, the product would have poor emulsion stability, thus resulting in a stringy mouth feel (Deswal et al., 2013; Zhang et al., 2007). This is because the complex starches are unable to be broken down. The lack of sweetness can be attributed to the lack of maltose and glucose, which can be increased during the enzyme treatment (Zhang et al., 2007). There are three classifications of amylase: Alpha, beta, and gamma (Taniguchi, & Honnda, 2009) (Table 1). The  $\alpha$ -amylase enzyme activity of flour can be measured via falling number test, amylograph, or with a Rapid ViscoAnalyzer (RVA) (Tomić et al., 2018).

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				-	/								

	Amylase type					
	α-amylase	γ-amylase				
Source	Animals, plants, & microbes	Plants, and microbes	Animals, and microbes			
Reaction products	Maltose, α-limit dextrins, and small amount glucose	Maltose, and β-limit dextrins	Glucose			
Cleavage site	Random α-1,4 glycosidic bonds	Second α-1,4 glycosidic bonds	Last α-1,4, and α-1,6 glycosidic bonds			

## 2.4.2. β-glucanase

Indicated by Table 2, other enzymes can also be used during production, such as  $\beta$ glucanase, and protease. Glucanase is responsible for the breakdown of glucan, a polysaccharide made of multiple glucose molecules (Lafond et al., 2012). When no enzyme is present during oat milk production, the product tends to have an off mouth feel that makes the product seem runny, or slimy (Aastha et al., 2014). The oat flour does not have a high enough enzyme activity to break down enough of the complex sugars/starches. To achieve an ideal mouthfeel, additional enzymes are needed (Lafond et al., 2012).

Table 2.  $\beta$ -glucanase type and functionality

Туре	Functionality
β-1,3-glucanase	Breaks down $\beta$ -1,3-glucans
β-1,6-glucanase	Breaks down β-1,6-glucans
Cellulase	Hydrolysis of 1,4-β-D-glucosidic bonds

#### 2.4.3. Alternative enzyme options

Protease can also be used, but in addition to amylase and/or  $\beta$ -glucanase. Protease is typically used to break down protein via proteolysis (Razzaq et al., 2019). When used in oat beverage production, protease can break down protein within the oat base. The product of a protease treatment is predominately amino acids (Razzaq et al., 2019). Amino acids are the building blocks of proteins and play an important role in body function, body development, and digestion (Razzaq et al., 2019). Although there is still needed research, theoretically the presence of more amino acids could increase price and alter the nutrient composition of the product.

An exciting new enzyme treatment has been produced by Novozymes. The company has developed a processing method called that utilizes multiple enzyme treatments (Watson, 2017). The first treatment is done with  $\alpha$ -amylase after the slurry is heated. This liquification step allows for the breakdown of amylose and amylopectin into dextrins and starch fragments. The

mixture is then cooled and treated with glucoamylase, which further breaks down the starch and dextrins into glucose and isomaltose (Watson, 2017). The second enzyme treatment will increase the dextrose equivalent (DE) value, create better mouth feel, and help with adjusting sweetness of the beverage (Watson, 2017). When compared to the process methods described prior, an additional cooling step may be needed for optimization of the glucoamylase treatment (Watson, 2017). This step may not be cost effective in some situations. Novozymes is currently developing an all in-one enzyme treatment that yields similar results when compared to the two-enzyme treatment method.

### 2.4.4. Use of catalysts for optimization of enzyme treatment

To further ensure that the enzyme treatment is being optimized, a catalyst can be added during the treatment. A salt like calcium chloride (CaCl<sub>2</sub>) is often added to improve the enzyme activity of  $\alpha$ -amylase. The CaCl<sub>2</sub> acts as a stabilizer for the  $\alpha$ -amylase's chemical structure, as well as a thermodynamic stabilizer (Yadav & Prakash, 2011). The CaCl<sub>2</sub> and its functionality depends on the concentration being used. In a study conducted by (Yadav, 2012), the  $\alpha$ -amylase activity was recorded while the CaCl<sub>2</sub> concentration was increased from 0-50 millimolar (mM). Where one unit of enzyme activity is defined as the amount of enzyme required to produce 1 µmol of maltose from 1 ml of 1% starch solution in 5 minutes at 37°C at pH 5.9 (Figure 2).



Figure 2. Effect of CaCl<sub>2</sub> on  $\alpha$ -amylase activity Reprinted without changes from (Yadav, 2012)

The use of CaCl<sub>2</sub> as a catalyst for enzyme activity was optimized at around 2 mM. A lower concentration of CaCl<sub>2</sub> primarily maintains the integrity of the functional sites, while a higher concentration acts as an enzyme inhibitor (Yadav, 2012).

## **2.5.** Fortification of oat beverage

To ensure a high quality and stable oat beverage, fortification with other nutrients must be conducted (McClements, 2020). A common goal of oat beverage companies is to closely emulate dairy milk in its rheological and nutritional properties. Utilizing food ingredient technology and fortification helps achieve that goal. During fortification many different ingredients can be added to improve or alter the product. Important ingredients include oil, water, emulsifiers, micronutrients, and other additives.

# 2.5.1. Oil & water

One of the most important rheological properties, mouth feel, rely on the system's ability to create an emulsion. The water and oil within the system need to be similar in particle size to ensure a smooth mouth feel. Other plant oils are often added to supplement the nature oil content of the beverage base. Coconut oils are often used because they are composed of medium chain saturated fatty acids, which are highly stable to lipid oxidation but carries adverse health effects when compared to unsaturated fats (Neelakantan et al., 2020). Another popular plant oil used is flaxseed oil. Flaxseed oil contains high amounts of long chain polyunsaturated fatty acids, which are more susceptible to lipid oxidation but yield a much better health profile (Neelakantan et al., 2020).

The water chosen has an impact on product quality as well. Commercial water may contain unwanted minerals, as well as varying pH values. It is recommended that the water used be treated prior to fortification. Popular treatments include thermal processing, filtering, and reverse osmosis (Zhang et al., 2007). If needed, minerals and buffers can be added after water treatment to ensure the desired rheological profile, acidity, and taste (Zhang et al., 2007).

#### 2.5.2. Emulsifiers

The proper selection of an emulsifier is pivotal to produce high-quality oat beverage. The added emulsifier has a role in processing methods, shelf-life, appearance, mouthfeel, and flavor (McClements et al., 2019). Popular plant-based emulsifiers used include proteins, polysaccharides, phospholipids, and biosurfactants (McClements & Gumus, 2016). The two most important factors in creating an emulsion are to ensure the minimum amount of emulsifier is used, and to obtain the smallest possible mean droplet size (Guzey & McClements, 2006). Another contributing factor is the particle size distribution. Ideally, the particle size distribution should be narrow (McClements et al., 2019). This is to ensure a layer of cream does not form upon storage and to support a proper mouth feel. Phospholipids and plant saponins are the most effective in achieving a small particle size, while being size stable from pH 3-8 (McClements et al., 2019). To further ensure a quality emulsion, two or more emulsifiers can be used

synergistically. When using multiple emulsifiers, the means of delivery become more important. Commonly used delivery methods include formation of conjugates, formation of physical complexes, and simple mixing.

#### 2.5.3. Added nutrients

A goal of plant-based milk companies is to produce a beverage that closely emulates dairy milk in nutrition and functionality. An oat base that is not fortified is considered relatively low in nutrients. The base is mainly composed of soluble fibers, starches, and sugars (McClements, 2020). With the addition of macro/micro-nutrients, the composition of the beverage is changed, and becomes more nutritious. Commonly added nutrients include vitamins A, D, B2, B12, and minerals like calcium (McClements et al., 2019). When these ingredients are added, the oat beverage can resemble the nutrient profile of dairy milk.

## 2.5.4. Texture modifiers & stabilizers

Common issues that arise during the packaging and storage phases include creaming and concerns of sedimentation of dense insoluble matter. Creaming occurs when there is gravitational separation of the fats droplets, while sedimentation issues clumps, or aggregates, forming within the system (McClements, 2020). To counter these unwanted attributes numerous food additives can be introduced into the system, like texture modifiers and thickening agents. When assessing natural plant-based options, products such as starch, pectin, locust bean gum, or guar gum are often used. Carrageenan and alginate are also options that are derived from seaweed, while xanthan gum is a product of microbial fermentation (Håkansson, 2019).

The ability to manipulate these texture modifiers has become an area of emphasis in research and production. Electrically charged polymers can also be used to stabilize oil bodies and ensure fat droplets from aggregating by forming a protective barrier (McClements & Gumus,

2016). An example of this is anionic carrageenan, which can absorb to cationic patches on the surface of colloidal particles, thus preventing their tendency to aggregate near the isoelectric point (Håkansson, 2019). The addition of texture modifiers and stabilizers is essential to the production of a shelf stable, and a desired mouth feel.

## **2.6.** Preferred packaging (methods and packaging)

Like dairy milk, the packaging used plays a role in shelf life and stability. Popular brands like Oatly<sup>™</sup> use a durable carboard paper carton. This carton doesn't allow light to enter the system, which aids in the fight against product degradation. When evaluating a dairy milk container, it's opaque nature was designed to retard degradation on riboflavin and lipids (Kontominas, 2010). The same can be inferred for oat beverage, cardboard cartons ensure that no light enters the system, thus retarding degradation and extending shelf life.

Industrial packaging methods utilize refrigeration, and aseptic conditions during packaging. This further ensures a safe product for consumers, while also improving shelf life and storability (Kontominas, 2010). After stored at room temperature for a couple weeks the oat beverage is ready for packaging in sterile and opaque cartons. Another benefit of sterile packaging and heat treatment of the beverage is the ability to store in room temperature or in refrigeration (Sanjana et al., 2016). This is a common trait of many plant-based milks and aids in the logistics and storage of beverage.

## 2.7. Oat β-glucan & benefits of consumption

## 2.7.1. Introduction & chemical structure

 $\beta$ -glucans are naturally occurring polysaccharides that are commonly found within the cell walls of bacteria, yeast, lichens, and cereal grains (Regand et al., 2011). Barley and oat are

the predominant source of  $\beta$ -glucan when assessing cereal grains. There are many functions of  $\beta$ -glucan, which allow for applications in food systems, cosmetics, and medicine.

The backbone of glucans is made of D-glucose rings, connected in a linear fashion, with some branching, and various linkage points. The structure of the glucan is an indicator of functionality and the source of which it is derived.  $\beta$ -D-glucan is polysaccharide commonly found in cereal grains, such as barley and oat (Regand et al., 2011). The backbone of  $\beta$ -D-glucan is derived of predominantly (1-4)-linked  $\beta$ -D-Glucopyranose (Glc*p*) units, interrupted by a single (1-3)-linked  $\beta$ -D-Glc*p*. Figure 3 represents the simplified backbone and linkages of  $\beta$ -glucan found in oat.



Figure 3. Diagram of  $\beta$ -glucan found in oat Reprinted without changes from (Channell et al., 2018)

Within oats, the  $\beta$ -glucan is mainly found in the endosperm of the oat, especially in the outer layers on the endosperm. When looking at  $\beta$ -glucan in barley, it is found uniformly throughout the endosperm (Regand et al., 2011).

#### 2.7.2. Cereal β-glucans & health claims

 $\beta$ -glucan has numerous applications within the health, wellness, and medicinal sectors.  $\beta$ glucan consumption has been used to aid in the fight against metabolic syndrome. Metabolic syndrome occurs when the body is affected by a cluster of conditions that increase the risk for heart attack, stroke, or type 2 diabetes (Wood, 2007; Bruce & Hanson, 2010). Side effects of this condition include high blood pressure, high blood sugar, excess stomach fat, and abnormal cholesterol.

Hypertension, or high blood pressure is a common side effect of metabolic syndrome. When hypertension goes unchecked, it can cause heart disease, stroke, and renal disease (Rasane et al., 2015). Although the relationship between hypertension and dietary fiber are studied less often, there has been promising finding that increased dietary fiber consumption provides a safe and acceptable means to lower blood pressure in patients that suffer from hypertension (Wood, 2007; Eckel et al., 2010; Ahmad et al., 2012). Another use within the medicinal sector would be its use as an insulin resistance material.  $\beta$ -glucan, as well as arabinoxylan, can be used with proper diet to ensure that the consumers insulin levels do not spike or drop after consuming food high in sugar. Another staple of metabolic syndrome is the accumulation of fat around the midsection. Professionals amongst many fields are trying to recommend foods that reduce energy intake by inducing satiation and increasing the feeling of satiety (El Khoury et al., 2012). Although there have been no findings that just an increase in dietary fiber consumption can affect body fat/weight. If the consumption is increased and the patient follows dietary guidelines and a proper exercise routine, it was shown to reduce weight. A similar study was conducted using glucomannans, which found that this soluble dietary fiber yields a larger decrease in weight, despite needing a smaller dose (Mathews et al., 2020; Alydar et al., 2020). When guar

gum is added to the matrix, it increased the perception of fullness, and decreased hunger after consumption (Mathews et al., 2020). Although there are some findings in the medical applications of  $\beta$ -glucan, there needs to be much more research done on the subject. With further understanding of how  $\beta$ -glucan affects the body, we can as food scientists and medical professionals recommend and produce healthier food/beverage for the consumer.

## 2.8. Comparing nutrient content of dairy milk & plant-based milks

A consumer may be interested in a plant-based substitute for many reasons, some of which include suffering from lactose intolerance, gluten sensitivity, and irritable bowel syndrome (IBS). Others are looking for a dairy free substitute out of personal choice, or the need of a low-calorie dairy substitute. Assessing plant-based substitutes regarding nutrient intake showed that oat beverage was most like bovine milk in calorie intake (Ferruzzi et al., 2019) (Table 3).

	Beverage Source*					
Nutrients	Almond	Bovine	Oat	Rice		
Carbohydrates (g)	14	13	26	22		
Protein (g)	1	8	4	0.7		
Fat (g)	2.5	1.3	2	2.3		
Fiber (g)	1	0	2	0.7		
Calories (kcal)	79	130	130	113		

Table 3. Nutrient contents of milk & plant-based substitutes

\*Each source was assessed given (8.0oz) of liquid beverage

\*Reprinted without change from (Ferruzzi et al., 2019)

This means that oat beverage would best suit those searching for dairy or gluten free options (Chalupa et al., 2018). If a consumer is searching for a low-calorie option, almond beverage showed to have less calories, carbohydrates, and protein, when compared to all options (Chalupa et al., 2018). Protein fortified beverages may be an area of future research due to plantbased options registering low in protein content. Increasing protein would further alter the nutrient profile of the beverage, while also increasing the price of the product.

## 2.9. Oat base & other uses

## 2.9.1. Oat base & baking

Like dairy milk, oat beverage provides numerous benefits to a baking system. When assessing baking moisture, oat beverage acts as a medium for the dissolution of salts and sugars, while also improving gluten development (Huang et al., 2019). The beverage also provides the sugar and proteins needed for Maillard reactions to occur. When baking, oat beverage can replace dairy milk on a 1:1 basis, meaning there is no need to compensate for amount used (Huang et al., 2019). Those who use oat beverage as a dairy substitute in baking compliment the unique toasted flavor profile. Oat beverage can be used as in products such as muffins, cakes, or moist breads like banana bread.

#### 2.9.2. Oat base & non-dairy creamers

Oat beverage is lactose-free and gluten-free. Having these attributes has driven oat beverage use as a dairy-free creamer. Chain coffee shops and craft coffee shops alike have been adding oat beverage to their list of possible ingredients. The ability to create a stable foam is important to making craft drinks such as cappuccinos, and lattes (Pilhofer et al., 1994). Oat beverage closely emulates the foaming and frothing of dairy milk, while offering a unique nutty flavor. Oat beverage would cater to vegan and non-dairy consumers but would not act as a lowcalorie substitute for those implementing a low-calorie diet.

## 2.10. Market opportunities

If you were to walk into a grocery store today, there would be numerous choices available for oat beverage. The number of options has dramatically increased when compared to

available options ten years ago. Potential reasons that the milk market has diversified include the looming global warming crisis and a growing movement for animals' ethical treatment. The estimated global market value of oat milk is nearly \$4,000,000,000 and is expected to grow at 9.8%/year (Sethi et al., 2016).

Today oat beverage has a variety of flavors that include vanilla, chocolate, strawberry, plain and reduced sweet. Oat beverage can also be used as a dairy substitute in baking and is often used in fermented dairy products like yogurt. An exciting sector seems to be the foodservice industry. Oat beverage is a substitute for coffee and espresso drinkers that want something with similar flavor and texture to milk/cream.

Another market opportunity would be with consumers who are lactose intolerant or have dietary restrictions. Lactose intolerance is a digestive disorder in which the body does not produce enough lactase. Lactase is the enzyme that breaks down lactose. The intact lactose causes cramping, bloating, and diarrhea (Szilagyi et al., 2019). Having a lactose-free milk option would accommodate those who suffer from lactose intolerance. Another dietary benefit would be the calories on a per-serving basis and a high amount of dietary fiber and healthy fats (Szilagyi et al., 2019). Oat beverage gives options to those who desire bovine milk but are incapable of consumption due to health and dietary restrictions.

## 2.11. Legal standing and regulation

Although the legal situation around how to refer to plant-based beverages is still murky, many companies within industry are preparing to change the technical name of their product. The traditional dairy milk industry hopes to rid the plant-based 'milk' name, claiming that real milk is obtained from the lacteal secretion, practically free from colostrum, obtained by the complete milking of one or more healthy cows. The opposition, plant-based food and beverage

companies, state that the standard of identity only applies to the unqualified term 'milk' and not compound names like 'almond milk'. The plant-based food and beverage sector also believes that displaying 'plant-based' on the label should allow for the term 'milk' to appear on the label. With plant-based meat becoming more popular, term like 'beyond' and 'impossible' meat have been implemented on products and advertisements. Although plant-based products are becoming much more popular, and production methods have been improved greatly over the past decade, the regulatory battle that ensues will cause headaches for both industries, while also demanding more resources and monetary capital.

As of 2021 the labeling of plant-based beverages as 'milk' still occurs in the United States. The Food and Drug Administration (FDA) states it will continue to prioritize issues that matter and affect consumers. With both dairy and plant-based industries growing we will see an uptick in lobbying from both fronts.

## 2.12. Sustainability and future research directions

## 2.12.1. Sustainability (emissions, methane prod, water & land usage)

With more of the public becoming aware of climate change, plant-based food substitutes have increased in popularity. One of the largest contributors to global warming is the emission of methane gas. Methane gas can be produced by cows and released by flatulence, as well as a byproduct of various production methods. The dairy industry needs substantial land and water reserves, to support the production of the milk and the housing and wellbeing of its cows. A study conducted by (Poore, & Nemecek, 2018) found that dairy milk when compared to plant-based beverages produces much higher emissions and requires more land and water use.

Oat beverage yields much lower emissions, with next to no methane produced. The land and water needed to yield the same amount of beverage is much lower than dairy milk as well.

Acquiring the ability to produce a beverage that has less of a carbon footprint, would not only be beneficial for the environment but also for those consumers who are searching for a sustainable alternative.

## **2.12.2.** Other uses for production byproducts (residues)

When finished producing oat beverage, there are solids or residues left over. These residues are rich in  $\beta$ -glucans, which has numerous uses and advantages of consumption.  $\beta$ -glucans are a source of soluble fiber, and research suggest that consumption of  $\beta$ -glucans can reduce high cholesterol and improve skin conditions such as bed wounds, and eczema (Wang et al., 2017). More times than not, the residues are sold to farms for animal feed. Using the byproducts this way utilizes other means of disposal, instead of incineration or disposal via land fill. Other uses are being discovered as well, these include being used in baking, cosmetics, and other food products. For baking, the residues can be used to enrich bread, or as a partially wheat flour substitute. Although not scene often in the industrial cosmetics sector, a new trend among consumers shows usage as a skin exfoliator (Wang et al., 2017).

## **3. OBJECTIVES AND NEEDS STATEMENT**

## 3.1. Needs statement

Milk has been a staple in diets worldwide because it is an excellent source of calcium and vitamin D; it can also be used in cooking and baking. In the past decade or so, there has been a considerable uptick in plant-based beverage consumption. The most prominent options right now are almond and oat beverage. Oat beverage is popular because it emulates the flavor and versatility of bovine milk. A common slight against oat milk is that it can sometimes have a "slimy" mouthfeel. The undesired slimy mouthfeel is caused by complex sugars that were not broken down entirely after enzyme treatment. With a better understanding on production methods of oat beverage, companies in industry can produce higher quality oat beverage that more closely resembles bovine milk. Thus, potentially opening the door for more people to consume plant-based beverages, especially an oat-based beverage.

## 3.2. Research objective

<u>Objective 1:</u> To evaluate how extraction methods affect the quality and composition of oat beverage.

<u>Objective 2:</u> To evaluate how utilizing different groats types affect the quality and composition of oat beverage.

<u>Objective 3:</u> To evaluate how the grain to water ratio affects the quality and composition of oat beverage.

## 4. MATERIALS AND METHODOLOGY

## 4.1. Materials

Materials needed for the study include various groats, which include heat treated, nonheat treated, and rolled groats. The heat-treated groats and non heat-treated groats were a gift from Dakota Specialty Milling (Fargo, ND, USA) and the rolled groats were purchased from a local grocery store. Calcium chloride,  $\alpha$ -amylase, and any other chemicals were of at least reagent grade and purchased from Sigma-Aldrich.

## 4.2. Methods

#### 4.2.1. Oat base production

The methods used were derived from a study with some modifications (Salama et al., 2011). The initial step of the experiment was to acquire different groat types for analysis. The three types of groats used include heat treated (Stabilized), non-heat treated (un-stabilized), and rolled groats that were pressed and steamed. The ratio of grain to water was also analyzed during the study. The sample either had a (1:4) or (1:6) ratio of grain to water. The (1:4) had 50g of grain/flour with 200ml of deionized water, while (1:6) had 33.33g of grain/flour with 200ml of deionized water. There were three extraction methods followed to produce the oat beverage. The first was dry milling with blending, centrifugation, and homogenization. The second method was wet milling with soaking, draining, blending, centrifugation, and homogenization. The final method was dry milling with an  $\alpha$ -amylase treatment, blending, centrifugation, and homogenization.

Industry standards were also compared against the various extraction methods described above. To accomplish this, samples of consumer ready oat beverage from Oatly<sup>TM</sup>, Planet Oat<sup>TM</sup>,

and Silk Oat<sup>™</sup> were acquired. Since the products were ready for consumption, the extraction steps were not necessary, and the samples were ready for analysis.

## 4.2.1.1. Groat milling

There were two milling methods used during the study. The first was a dry milling method using a Udy Cyclone Sample Mill with a 25µm filter (Udy Corporation, Fort Collins, CO, USA) (Figure 4). Roughly 1.5kg of groats were milled to flour using this method. The flour produced with this method was used for extraction methods 1 & 3.



## Figure 4. UDY cyclone mill prior to milling

Wet milling was used for extraction method 2. The process of wet milling included weighing out the required grain, either 50g or 33.33g, and adding 200ml of water to an airtight storage container for 18 hours. After soaking, the container was drained of water and the saturated groats are moved to a blender. Another 200ml of fresh deionized water was added to the groats in the blender. After blending for 60 seconds a slurry of water and groats remains.

#### **4.2.1.2.** Enzyme treatment

The only extraction method that underwent an enzyme treatment was method 3. Once the dry milling is conducted the flour is added to a blender with 200ml of deionized water and blended for 60 seconds. The slurry was then transferred into a glass beaker and depending on the grain to water ratio, either 0.0048g (1:4) or 0.0032g (1:6) of  $\alpha$ -amylase was added to the beaker. Each sample, regardless of grain to water ratio received 0.08g of calcium chloride, the addition of CaCl<sub>2</sub> acts as a catalyst during the enzyme treatment. The samples were then placed into a hot water bath set to 70°C for 40 minutes. After 40 minutes the temperature must be increased to 90°C, this is done to inactivate the  $\alpha$ -amylase and ensure the enzyme does not further breakdown the sample. The slurry was allowed to cool to room temperature and when ready the samples were transferred to plastic centrifugation containers.

#### 4.2.1.3. Centrifugation

Centrifugation was needed to separate the oat beverage base from the solids within the sample. This was done at 3000 rpm for 15 minutes. Every sample regardless of extraction method was centrifuged. The liquid oat beverage base was then separated into a clean beaker and awaits homogenization, while the solids were scraped out and stored in clear sample bags.

#### 4.2.1.4. Homogenization

Homogenization is a pivotal step that ensures the oat beverage base in mixed thoroughly and uniformly. This was done using a handheld homogenizer at speed half speed for 60 seconds. After homogenization the samples underwent physical analysis and both liquid and solids samples were freeze dried.

#### 4.3. Oat sample composition

The groat samples were first analyzed on a physical basis. To prepare the samples for analysis, milling was done using a UDY Cyclone mill. The samples analyzed included stabilized, non-stabilized, and heat treated rolled groats. The groats were inspected for moisture using the AACCI method 44-15.02, ash using AACCI method 08-01.0, and protein composition using the AACCI method 46-30.0 (Cereal and Grains Association, 2009). Analysis of total starch, and βglucan was executed by using the Megazyme/AACCI methods 76-13.01 and 32-23.01, respectively (Cereal and Grains Association, 2009). Finally dietary fiber analysis was conducted using the Ankom/AOAC Method 2011.25.

## 4.4. Oat base composition

The extracted oat beverage bases were analyzed as well. All samples were freeze dried before analysis. All dried beverage base samples underwent analysis for ash using AACCI method 08-01.01, protein composition using the AACCI method 46-30.01, and total starch while using Megazyme/AACCI method 76-13.01 (Cereal and Grains Association, 2009). Free glucose was assessed using the Megazyme free glucose kit, while β-glucan composition was analyzed using Megazyme/AACCI method 32-23.01 (Cereal and Grains Association, 2009). The Brix° was determined by using AACCI method 80.51.01 (Cereal and Grains Association, 2009). The solids and yield content were determined by weight, while yield refers to the liquid beverage and solids refer to the residues after centrifugation. Finally, sugar composition, and molecular weight analysis was carried out using GC (Blakeney et al 1983 & Mends and Simsek 2015), and HPSEC-MALS (Alahmed and Simsek 2020), respectively.

Microbial analysis was also done on oat milk bases. The samples were plated onto plate count agar (PCA) and plates were incubated at 30°C for 48 h. Aerobic colonies were counted on

a Q-count. Uncountable plates were marked with '\*', and for all other samples the log colony forming units (CFU)/ml are presented. We acknowledge Jessica Lauer (under Dr. Teresa Bergholz, Michigan State University) at North Dakota State University for conducting the microbiological analysis.

## 4.5. Oat residue analysis

The oat beverage residue byproducts were also analyzed. The residue consists of the solids after centrifugation of the beverage base occurs. Similar to the oat base, the residue samples were also freeze dried prior to analysis. All dried beverage base samples underwent analysis for ash using AACCI method 08-01.01, protein composition using the AACCI method 46-30.01, and total starch while using Megazyme/AACCI method 76-13.01 (Cereal and Grains Association, 2009). Free glucose was assessed using the Megazyme free glucose kit, while β-glucan composition was analyzed using Megazyme/AACCI method 32-23.01 (Cereal and Grains Association, 2009). Finally, sugar composition, and molecular weight analysis was carried out using GC (Blakeney et al 1983 & Mends and Simsek 2015), and HPSEC-MALS (Alahmed and Simsek 2020), respectively.

## 4.6. Statistical analysis

All samples were analyzed in duplicate. Mean separation and least significant difference tests were utilized to indicate significant differences (p<0.05) between treatments using one-way Analysis of Variance (ANOVA) in SAS for Mac, Version 9.4 (TS Level IM4). A three-way ANOVA test was also conducted to analyze the relationship between groat type, water to grain ratio, and extraction method.
## 5. RESULTS & DISCUSSIONS

## 5.1. Groat samples

Once the groats were milled and freeze dried, all types underwent analysis for moisture, ash, protein, total starch, free glucose, and sugar composition. This analysis was done to further understand the physical characteristics of the groats, while also evaluating their chemical composition. All final calculations were determined on a dry weight basis. No commercial benchmark products (Oatly, etc.) were analyzed because the product was acquired already fully processed.

## 5.1.1. Compositional characteristics by groat type

The moisture and ash analysis were done in succeeding order. First, the samples underwent moisture analysis. Followed by ash analysis using the samples that had just be evaluated for moisture. Thus, the ash samples were already considered to be dry weight basis (Figure 5).



Figure 5. Samples in desiccator after moisture and ash analysis

The analysis found that stabilized groats yield significantly higher moisture and ash contents, when compared to non-stabilized, and rolled groats (p<0.05). The protein content was significantly higher regarding the rolled oats, while the non-stabilized groats yield the smallest amount of protein (p<0.05) (Table 4). These values closely resembled literature, with the average protein percent for oats ranging from 11-15% (Klose et al., 2009; Robert et al., 1985).

Table 4. Oat sample physical characteristics

Sample	Moisture	Ash	Protein
		(%)	
Stabilized Groats	15.89a	2.19a	14.44b
Non-Stabilized Groats	12.35b	2.12b	13.81c
Rolled Oats	10.61c	2.05c	14.96a

Mean values in a column followed by the same letter are not significantly different (p < 0.05).

#### 5.1.2. Carbohydrate composition by groat type

After the groats were milled, and freeze dried the proximate composition was determined (Table 5). Stabilized groats showed to have a significantly higher percent of total starch, roughly 62%, while non-stabilized and rolled oats did not significantly differ (p<0.05). All three types of groats were on par with other studies and literature. According to Berski et al., (2011) the average starch available in oats is roughly 60%.

The  $\beta$ -glucan content was significantly higher (p<0.05) in the rolled oats, while there was no statistical difference between the stabilized and non-stabilized groats (Table 5). According to Flander et al., (2007), the average  $\beta$ -glucan content of oats ranges from 2.3-8.5%, all analyzed samples fell within that range with rolled oats yielding the highest  $\beta$ -glucan content.

When looking at free glucose, the stabilized groats registered at 44.74 mg/g, which was significantly higher than other groat types. A study conducted on the composition of oats found that heat treated oats tend to yield higher glucose levels, than untreated, or steel cut oats (Varma et al., 2016). This is because when the oats were stabilized via heat treatment, and the availability

of starch for enzyme hydrolysis improves, and therefor increase the glucose within the sample (Varma et al., 2016).

Sample	Total Starch	β-glucan	Arabinoxylan	Arabinose	Xylose	Mannose	Galactose	Glucose
				(%)				mg/g
Stabilized Groats	61.97a	3.84b	1.80a	0.99b	1.06a	0.00a	1.51b	44.74a
Non- Stabilized Groats	58.84b	3.99b	1.57b	0.88c	0.91b	0.00a	2.02a	39.38b
Rolled Oats	58.00b	4.85a	1.28c	1.07a	0.39c	0.00a	1.02c	33.09c
1 .	1	C 11	11 /1	1		41 1.00	1 ( 10	0.5)

Table 5. Oat type and carbohydrate composition

Mean values in a column followed by the same letter are not significantly different (p < 0.05).

When assessing the proximate composition, mannose was not detected in any of the three groat types. In a similar study conducted on the sugar composition of oats, it was determined that oats do not contain any mannose (Welch, 1995; Berski et al., 2011). Arabinoxylan is a hemicellulose, similar to  $\beta$ -glucan, that upon consumption shows benefits for heart and gut health (Schupfer et al., 2021). Stabilized groats generated a significantly higher arabinoxylan content, followed by non-stabilized groats, and rolled oats, respectively (p<0.05). Regarding arabinose, rolled oats produced a significantly higher content, at roughly 1.1% (p<0.05). While stabilized groats had a significantly higher xylose content, and non-stable groats yielded significantly higher in galactose percent, 2.02%, when compared to other groat types.

#### 5.2. Oat beverage bases

The oat beverage base samples underwent numerous assays, which include analysis of physical characteristics, proximate composition, and microbial activity. Like the groat analysis, the beverage base samples where freeze dried prior to analysis. The physical and proximate composition were determined in the same manner as the groat samples discussed prior.

#### 5.2.1. Proximate composition and physical characteristics of oat beverage base

#### 5.2.1.1. Groat type & physical characteristics of oat beverage base

Indicated in Table 6, the rolled groats appeared to yield a significantly lower moisture and ash content, while stabilized and non-stabilized groats produced higher values but not significantly different amongst each other (p<0.05). According to Aydar et al., (2020) the ash content of oat beverage is 0.48%. The amylase treated samples were similar to the standard given by literature, but the commercial standards all yielded much higher ash contents 8.22-11.05%. The higher ash content could be attributed to the fact that the commercial standards underwent fortification, thus more inorganic ingredients would be added to the system.

The protein content of the samples made from non-stabilized groats was found to be significantly higher than both stabilized oats and rolled oats. This could be because both the rolled and stabilized oats underwent heat treatment prior to base extraction. The proteins within the sample begin to denature around 85-95°C, with significant denaturization at temperatures exceeding 110°C (Ma & Harwalkar, 1984). The amylase treated samples showed to have results similar too but still higher than literature states (Robert et al., 1985). Samples that underwent extraction with rolled oats produced significantly higher values for total starch (p<0.05), followed by non-stabilized and stabilized oats.

Treatment Type	Water/Flour Ratio	Extraction Method	Moisture	Ash	Protein	Solid	Yield	pН	rf(Brix)
- ) P •	10000				(%)				°Brix
Stabilized	1:4	Dry	7.69bc	7.64c	29.01c	1.69f	39.50egh	6.15fg	1.60g
Stabilized	1:6	Dry	9.41a	7.96abc	23.17de	1.07f	49.25cdeg	6.18fg	1.15g
Non-stabilized	1:4	Dry	8.45c	8.50ab	31.78b	2.21f	44.25degh	6.12g	2.25g
Non-stabilized	1:6	Dry	7.27c	6.65d	31.53b	1.86f	53.25cdeg	6.15fg	2.10g
Rolled	1:4	Dry	1.56g	2.33j	24.41d	10.26de	26.25h	6.33e	5.50f
Rolled	1:6	Dry	3.47e	3.82hi	34.41a	2.33f	38.05cgh	6.41d	2.40g
Stabilized	1:4	Wet	7.44c	4.91fg	20.25f	0.93f	67.00ab	6.18fg	1.30g
Stabilized	1:6	Wet	9.29bf	3.24ai	21.13ef	0.68f	69.50abc	6.19f	1.00g
Non-stabilized	1:4	Wet	7.81abc	5.25ef	34.97a	1.48f	67.00abc	6.01h	1.75g
Non-stabilized	1:6	Wet	9.54a	5.12aefg	35.56a	0.84f	67.25abc	6.00h	1.30g
Rolled	1:4	Wet	6.57d	5.83e	22.25ef	1.14f	79.75a	6.27e	0.65g
Rolled	1:6	Wet	4.97e	4.40gh	16.76g	0.73f	76.00ab	6.30e	0.60g
Stabilized	1:4	Amylase	2.18fg	1.11k	1.971	13.14bc	59.00cde	5.72ij	13.25bc
Stabilized	1:6	Amylase	2.65fg	0.87k	2.041	13.07bc	54.50cdeg	5.73ij	13.15bc
Non-stabilized	1:4	Amylase	3.44efg	0.84k	4.18jk	14.98ab	53.50cdeg	5.62kl	15.10ab
Non-stabilized	1:6	Amylase	3.67ef	0.65k	4.30j	11.08cd	59.50abcde	5.76i	11.25cd
Rolled	1:4	Amylase	1.79g	0.50k	2.20k	15.38a	53.50cdeg	5.68jk	15.65a
Rolled	1:6	Amylase	2.08fg	0.68k	1.991	10.65d	64.00abcd	5.551	10.95d
Silk	-	-	8.07abc	10.74a	10.35h	10.54d	NR	7.3c	8.8e
Planet Oat	-	-	2.86fg	8.20bc	8.06i	9.92de	NR	7.41b	9.9de
Oatly	-	-	7.11cd	11.05a	9.06hi	8.07e	NR	7.5a	8.8e

 Table 6. Physical characteristics of oat beverage base

Mean values in a column followed by the same letter are not significantly different (p<0.05).

The rolled oats produced significantly more solids than stabilized and non-stabilized oats (p>0.05). While there was no significant difference between the solid content of the stabilized and non-stabilized groats (p<0.05). The rolled oats produced samples with significantly higher pH values, followed by stabilized and non-stabilized. The pH for unfortified oat beverage should be between 5.5-7.5 (Syed et al., 2020; Aydar et al., 2020). All samples fell within the range given by literature, while commercial standards yielded values greater than 7.0. Lastly, there was no significant difference in the groat type used for extraction and Brix of the samples (p<0.05).

## 5.2.1.2. Water to grain ratio & physical characteristics of oat beverage base

The moisture content was significantly higher using a 1:6 water to grain ratio, when compared to 1:4 (p<0.05). This could be a result of more water within the system. The ash content and percent solids for the 1:4 ratio was significantly higher than the ash content and solids for the 1:6 ratio (p<0.05). This could potentially be attributed to the fact 1:4 ratio has more grain added than the 1:6. The 1:4 ratio also generated a significantly higher Brix value, when compared to the 1:6 ratio (p<0.05). Values for protein content and pH of the beverage bases did not show a significant difference among water to grain ratios (p<0.05). In commercial production, I would lean towards using the 1:4 water to flour ratio. This is because 1:4 produced a higher solids and Brix value, which is important to mouthfeel and texture of the product. Also, the 1:4 ratio produced values more similar to commercial benchmarks.

#### 5.2.1.3. Extraction method & physical characteristics of oat beverage base

Extraction methods were also analyzed during the study. The three types of extraction methods include dry milling, wet milling, and amylase treatment. All three methods produced significantly different average values for moisture and ash content, with wet milling having the highest value, followed by dry milling and then  $\alpha$ -amylase treatment, respectively (p<0.05). The

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protein content was highest among the samples that used the dry milling extraction method. Although significantly different, dry, and wet milling extraction methods produced similar protein values, 29.05% and 25.15%, respectively. While the amylase treated samples showed a value of just 2.78%.

All samples showed to have a higher protein percentage when compared to literature. Unfortified oat beverage yields roughly 0.75-0.1% protein (Syed et al., 2020; Aydar et al., 2020). The commercial standards all produced much higher protein contents, this could be because the commercial standards are fortified, thus increasing the nutrient content.

When assessing percent solids, it was noticed that the amylase treatment produced the largest average value, 13.05%, which was significantly more than both dry and wet milling, 3.23% and 0.97% (p<.05). The amylase treatment is responsible for the dramatic increase in percent solids, due to the enzymatic breakdown of complex starches. Once the enzyme treatment is finished, more simple sugars, and disaccharides are released into the beverage matrix, thus increasing the solids (Deswal et al., 2013).

The percent yield was significantly different amongst all three extraction methods (p<0.05). Wet milling had the highest percent yield, 71.08%, followed by amylase treatment, and dry milling. This could be because the wet milling samples were allowed to soak for 18 hours prior to extraction, while dry milling and amylase treatment samples were extracted from groats that had not been soaked. The soak time allows for further breakdown of its constituents, prior to extraction (Syed et al., 2020). Thus, solids and starch, are more readily available during extraction. Percent yield is a metric that is important to the production process. As a producer of oat beverage, the goal would be to have the highest percent yield. Optimization of this metric

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relies on proper production unit operations such as soak time, and the quality of ingredients like enzymes and oats.

Brix (°Brix) is the total amount of soluble solids within a solution (Bielmann et al., 2010). The average Brix value was significantly higher for samples that underwent amylase treatment, when compared to dry and wet milling (p<0.05) (Figure 6). These trends can be attributed to the amylase treatment, more specifically the breakdown of complex starch into simple sugars (Deswal et al., 2013).



Figure 6. t grouping of Brix<sup> $\circ$ </sup> averages and extraction method Mean values in a column followed by the same letter are not significantly different (p<0.05).

During enzyme treatment simple sugars that are soluble are produced, thus increasing the Brix value (Bielmann et al., 2010). If there was no amylase treatment, the starches will stay intact and insoluble. These insoluble long chain starches will play a negative role in mouth feel of the product, so it is important to break them down into their soluble constituents. Although much higher overall, the amylase treated samples closely resembled the commercial benchmarks. It is assumed that all commercial producers utilize an amylase treatment. The commercial samples produced between 8.8-9.9° Brix. The samples that underwent amylase treatment showed an average value of 13.22° Brix (Figure 7). Perhaps using less enzyme or reducing the time of enzyme treatment, could lower the Brix values by enough, to be like commercial options.

## 5.2.2. Composition of oat beverage base

## 5.2.2.1. Groat type & proximate composition of oat beverage base

When assessing groat type and total starch, it was evident that rolled oats produced a significantly higher average, when compared with stabilized and non-stabilized groats (p<0.05). While non-stabilized groats produced the smallest average value for total starch (Table 7). Similar to the total starch content, rolled oats produced a significantly higher (p<0.05) value of free glucose, followed by non-stabilized groats and stabilized groats, respectively. This trend makes sense because when the oat undergoes heat treatment, more starch is available to the process (Zhang et al., 2021).

Treatment Type	Water/Flour Ratio	Extraction Method	Total Starch	β-Glucan	Arabinoxylan	Arabinose	Xylose	Mannose	Galactose	Free Glucose
						·%				mg/g
Stabilized	1:4	Dry	2.50h	3.82d	1.95abc	1.78a	0.44defg	0.00d	0.65d	34.77f
Stabilized	1:6	Dry	2.72h	4.18c	2.21ab	1.69abc	0.83bc	1.17a	1.66d	32.35fg
Non-stabilized	1:4	Dry	1.20ij	2.59h	2.20ab	1.72ab	0.78bcd	1.00a	1.58d	64.61c
Non-stabilized	1:6	Dry	2.55h	3.06g	1.53cdefg	1.22def	0.52cde	0.66b	2.05d	50.33d
Rolled	1:4	Dry	5.55f	2.27i	2.44a	1.47abcde	1.30a	0.00d	1.68d	27.68g
Rolled	1:6	Dry	9.17d	3.43ef	1.89bc	1.27cdef	0.88b	0.56b	0.68d	43.74e
Stabilized	1:4	Wet	1.59i	4.34bc	1.34efgh	1.04e	0.49cdef	0.65b	1.91d	20.50h
Stabilized	1:6	Wet	0.96ijk	4.59b	1.60cdef	1.20def	0.61bcde	0.59b	2.22d	18.28hi
Non-stabilized	1:4	Wet	NR	3.71de	1.80bcd	1.45abcde	0.59bcde	0.45bc	2.26d	35.07f
Non-stabilized	1:6	Wet	0.081	3.86d	1.61cdef	1.29bcdef	0.54bcde	0.21cd	2.12d	32.56fg
Rolled	1:4	Wet	3.89g	3.78d	1.87bcd	1.46abcde	0.67bcd	0.00d	3.04cd	63.54c
Rolled	1:6	Wet	NR	3.72d	1.68cde	1.22def	0.69bcd	0.00d	1.46d	36.80f
Stabilized	1:4	Amylase	0.30kl	2.61h	1.15fgh	1.18def	0.12gh	0.00d	6.96ac	13.43ij
Stabilized	1:6	Amylase	0.46jkl	2.29i	1.17efgh	1.21def	0.11gh	0.00d	7.69a	49.61d
Non-stabilized	1:4	Amylase	6.69e	2.71h	0.89hi	0.89f	0.13gh	0.00d	3.01cd	6.53k
Non-stabilized	1:6	Amylase	6.42e	2.17fi	1.03gh	1.03ef	0.15fgh	0.00d	3.59bcd	8.44jk
Rolled	1:4	Amylase	9.48d	3.31g	1.68cde	1.34abcdef	0.57bcde	0.23cd	6.00ac	15.58hi
Rolled	1:6	Amylase	6.74e	2.57h	1.98abc	1.54abcd	0.72bcd	0.00d	3.12cd	15.42hi
Silk	-	-	51.95a	6.11a	0.49ij	0.26g	0.29efgh	0.13d	1.29d	93.74b
Planet Oat	-	-	36.88c	4.36bc	0.90hi	0.89f	0.13gh	0.18c	2.77cd	51.96d
Oatly	-	-	37.86b	4.55b	0.26j	0.30g	0.00h	0.00d	1.39d	214.18a

Table 7. Proximate carbohydrate composition of oat beverage base

Mean values in a column followed by the same letter are not significantly different (p < 0.05).

Evaluating the  $\beta$ -glucan composition showed that stabilized groats produced a significantly higher (p<0.05) value, followed by rolled, then non-stabilized. The  $\beta$ -glucan composition of the product is important for perceived health benefits, as well as the dietary fiber content. Having the ability to manipulate or optimize the  $\beta$ -glucan would be beneficial for commercial producers.

The arabinoxylan and xylose content of the oat beverage base was significantly higher (p<0.05) when processed with rolled oats. Regarding free glucose, rolled oats produced the highest average value, 33.79 mg/g, followed by non-stabilized and stabilized groats, respectively. The arabinose and galactose values were not significantly different (p<0.05) among groat type. Mannose was not expected to be a constituent of the oat beverage base. The presence of mannose could be because of the breaking down of galactomannans and the fact that they share similar structure.

#### 5.2.2.2. Water to grain ratio & proximate composition of oat beverage base

The only significant difference among the water to grain ratio was observed during analysis of free glucose. The water to grain ratio of 1:6 produced significantly (p<0.05) higher average values of free glucose when compared to the 1:4 (Figure 7).



Figure 7. t grouping of free glucose (mg/g) and water to grain ratio Mean values in a column followed by the same letter are not significantly different (p<0.05).

Total starch,  $\beta$ -glucan, arabinoxylan, arabinose, xylose, mannose, and galactose

composition were all not significantly different (p<0.05) across water to grain ratio.

# 5.2.2.3. Extraction method & proximate composition of oat beverage base

The total starch of samples that underwent an amylase treatment were significantly higher

(p<0.05) than both dry and wet milling. Amylase treatment produced an average of 5.01% starch,

while dry and wet milling produced values of, 3.95% and 1.09%, respectively (Figure 8).



Figure 8. t grouping of total starch (%) averages and extraction method Mean values in a column followed by the same letter are not significantly different (p<0.05).

The commercial standards produced much higher values of total starch, within the range of 36.88-51.95%. When starch is heated in the presence of water, the starch granules begin to swell. If the starch is continuously heated, the granules burst and dissolve into the matrix (Sethi et al., 2016). The temperature range of the gelatinization of oat starch is between 43.7-74.7°C (Sethi et al., 2016). There could be a few reasons that the starch content is relatively low for all samples, when compared to the commercial standards. For the dry and wet milling, the bases did not undergo amylase treatment, thus enzymatic hydrolysis could not occur. Regarding the amylase treated samples, the base had to be heated for enzymatic hydrolysis and for enzyme inactivation. During enzyme deactivation the samples were heated to 90°C for 3-4 minutes. After amylase treatment some of the samples were very viscous and resembled a gel. This could have been due to heating at a high temperature for too long. There is also the possibility that the enhanced starch content of the commercial standards is due to the fortification of the beverage.

All extraction methods produced significantly different (p<0.05) values for  $\beta$ -glucan composition. Wet milling was observed to have the highest value, followed by dry milling and

amylase treatment. According to Syed et al., (2020), the  $\beta$ -glucan content for unfortified oat beverage should be roughly 0.54%, which is much lower than all samples. The commercial standards all yield much higher values of  $\beta$ -glucan, which could be attributed to the fortification process.

The arabinoxylan and arabinose content both showed no significant difference (p<0.05) among extraction type. The galactose content samples that underwent amylase treatment were found to be significantly higher (p<0.05) then those samples that we processed with dry or wet milling alone. The presence of galactose will help ensure flavor quality, because it has about the same sweetness as table sugar.

When assessing the free glucose content of the base samples, the amylase treated samples produced a significantly (p<0.05) higher average value, 49.98 mg/g, when compared to other methods of production. Dry milling produced the second highest value, 20.91 mg/g, followed by wet milling, 11.92 mg/g (Figure 9).



Figure 9. t grouping of free glucose (mg/g) and extraction methods Mean values in a column followed by the same letter are not significantly different (p<0.05).

# 5.2.2.4. Molecular weight analysis of oat beverage base

Molecular weight and polydispersity (PDI), which is defined as the weight average and number average molecule weight (Mw/Mn) were also analyzed during this assay (Rane & Choi, 2007) (Table 8). Groat type and grain to water ratio, produced average values that were not significantly (p<0.05) differen

Treatment Type	Water/Flour Ratio	Extraction Method	HMW*	LMW*	HMW*	LMW*	HMW*	LMW*
			9	%	M	W	Mw	/Mn
Stabilized	1:4	Dry	24.86f	75.14g	325600k	28520h	1.19hi	3.52b
Stabilized	1:6	Dry	21.23h	78.77e	313150k	38535f	1.12hi	3.87a
Non-stabilized	1:4	Dry	47.64b	52.36k	647400jk	11325mn	2.26e	2.47ef
Non-stabilized	1:6	Dry	46.69b	53.31k	1045000ij	13690klm	1.72f	1.33h
Rolled	1:4	Dry	46.52b	53.48k	693700jk	63585d	1.14hi	2.72cde
Rolled	1:6	Dry	47.11b	52.89k	692200jk	67590c	1.11h	3.40b
Stabilized	1:4	Wet	36.15d	63.85i	874750jk	61255d	2.12ei	3.35b
Stabilized	1:6	Wet	31.22e	68.78h	270250k	21985	1.80f	2.68cde
Non-stabilized	1:4	Wet	47.07b	52.93k	829350jk	121351mn	4.31b	1.77g
Non-stabilized	1:6	Wet	43.47c	56.53j	1602500hi	18960j	4.52a	1.84g
Rolled	1:4	Wet	59.37a	40.631	566500jk	108450b	1.47g	1.65g
Rolled	1:6	Wet	59.95a	40.051	2100000gh	471700a	1.02i	3.37b
Stabilized	1:4	Amylase	18.16ij	81.84cd	8453000d	15675k	4.47ab	2.61cde
Stabilized	1:6	Amylase	18.96i	81.04d	6009500e	18865jl	4.02c	2.63cde
Non-stabilized	1:4	Amylase	22.60gh	77.40ef	931400jk	14735kl	1.28h	2.83b
Non-stabilized	1:6	Amylase	18.39ij	81.61cd	818250jk	14295kl	2.49d	2.76bd
Rolled	1:4	Amylase	17.72ij	82.28cd	3440500f	10515n	4.10c	2.82b
Rolled	1:6	Amylase	16.82j	83.18c	2355000g	21800ij	4.51a	2.58cdef
Silk	-	-	23.89fg	76.11fg	10246500c	32775g	1.02i	2.30f
Planet Oat	-	-	11.29k	88.71b	15110000a	34775g	1.04i	2.33f
Oatly	-	-	9.331	90.67a	14405000b	43770e	1.08i	2.52def

Table 8. Molecular weight and polydispersity index of oat beverage base

Mean values in a column followed by the same letter are not significantly different (p<0.05). \*High molecular weight (HMW), Low molecular weight (LMW)

When evaluating the high molecular weight portion (HMW) of the PDI it was evident that the amylase treatment produced the highest values of high molecular weight polymers when compared to dry milling. There was no significant (p<0.05) difference between samples that underwent amylase treatment and wet milling extraction. Regarding the low molecular weight (LMW) portion of the PDI, there was no significant (p<0.05) difference between extraction methods.

The commercial samples produced on average a higher value for HMW. With a higher HMW value the product rheological properties are affected as well. The mouth feel and viscosity will be favorable with higher molecular weight polymers (Stokes et al., 2013)

#### 5.3. Microbial activity analysis of oat beverage base

The microbial activity was assessed by Dr. Teresa Bergholz and staff at Michigan State University (East Lansing, Michigan). Once the samples underwent centrifugation, the solid residues and liquid base were separated and sub-sampled to allow for analysis of microbial activity. The metric of log CFU/g was determined during the assay. This term demonstrates the colony forming units per gram, which ideally should be as low as possible. Microbe activity is an important to the storability and shipping logistics of oat beverage. Acquiring the ability to minimize microbial activity will ensure quality products, as well as extending the shelf life of the product. Only extracted beverage bases underwent microbe activity analysis. Groats, oat beverage residues, and commercial benchmarks were not analyzed. Table 9 demonstrates the results of the microbial activity assay.

Rolled oats demonstrated the lowest value for CFU/g, 3.81. While stabilized and nonstabilized yields average values of 5.17 and 5.04, respectively (Figure 10).

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Treatment Type	water/flour ratio	Extraction Method	log CFU/g
Stabilized	1:4	Dry	7.23a
Stabilized	1:6	Dry	6.58ab
Non-stabilized	1:4	Dry	6.21b
Non-stabilized	1:6	Dry	6.21b
Rolled	1:4	Dry	NR*
Rolled	1:6	Dry	4.55c
Stabilized	1:4	Wet	6.55ab
Stabilized	1:6	Wet	6.64ab
Non-stabilized	1:4	Wet	6.51ab
Non-stabilized	1:6	Wet	6.32ab
Rolled	1:4	Wet	6.40ab
Rolled	1:6	Wet	6.84ab
Stabilized	1:4	Amylase	2.04ef
Stabilized	1:6	Amylase	2.00ef
Non-stabilized	1:4	Amylase	2.09ef
Non-stabilized	1:6	Amylase	2.90de
Rolled	1:4	Amylase	3.19d
Rolled	1:6	Amylase	1.88f
Silk	-	-	NR*
Planet Oat	-	-	NR*
Oatly	-	-	NR*

Table 9. Microbial activity of oat beverage base

Mean values in a column followed by the same letter are not significantly different (p<0.05). \*NR indicates value 'not recorded'.





It was evident that the extraction method played an important role in limiting microbe activity. Wet milling produced an average of 6.54 CFU/g, while dry milling had a value of 5.13 CFU/g. The amylase treated samples yield an average value of 2.35 CFU/g, which is significantly (p<0.05) lower than all other treatments (Figure 11). This could be because the  $\alpha$ -amylase treatment required heating the slurry to at least 90°C for enzyme inactivation. It appears the heating of the slurry was a multi-purpose step that both inactivated enzymes and lowered microbe activity.



Figure 11. t grouping of colony forming units per gram averages and extraction method Mean values in a column followed by the same letter are not significantly different (p<0.05).

#### 5.4. Oat beverage residues

Upon centrifugation, solids from the extraction were packed into the bottom of the samples tube. The tubes were placed standing upside down, which allowed for any residual water to leak out. The solids were then scraped into samples bags and freeze dried prior to physical characteristic and proximate composition analysis (Table 10).

Groat type	water/flour ratio	Extraction Method	Moisture	Ash	Protein	Total Starch
					%	
Stabilized	1:4	Dry	0.06f	1.49gh	13.35g	49.11e
Stabilized	1:6	Dry	0.15def	1.44ghi	13.48g	51.42d
Non-stabilized	1:4	Dry	0.36cdef	1.29ij	11.37hi	54.35c
Non-stabilized	1:6	Dry	0.61cdef	1.27j	11.62h	48.86e
Rolled	1:4	Dry	0.08ef	1.51fgh	9.86i	60.51a
Rolled	1:6	Dry	0.22cdef	1.66f	12.36gh	58.84ab
Stabilized	1:4	Wet	0.10ef	1.42hij	13.70fg	60.95a
Stabilized	1:6	Wet	0.51cdef	1.42hij	13.36g	57.98b
Non-stabilized	1:4	Wet	0.78c	1.38hij	11.38hi	57.85b
Non-stabilized	1:6	Wet	0.67cde	1.39hij	12.58gh	60.46a
Rolled	1:4	Wet	0.73cd	1.58fg	15.25f	59.02ab
Rolled	1:6	Wet	0.56cdef	1.52fg	15.26f	38.41f
Stabilized	1:4	Amylase	1.38b	3.26d	32.50d	35.48g
Stabilized	1:6	Amylase	1.86ab	3.48c	36.38b	24.84i
Non-stabilized	1:4	Amylase	1.71ab	2.99e	27.75e	33.09h
Non-stabilized	1:6	Amylase	2.01a	3.66b	34.55c	21.54j
Rolled	1:4	Amylase	1.47ab	3.62bc	32.61d	32.46h
Rolled	1:6	Amylase	1.61ab	4.10a	38.75a	24.03i

Table 10. Proximate composition of oat beverage residue

Mean values in a column followed by the same letter are not significantly different (p<0.05).

#### **5.4.1.** Characteristics of oat beverage residues

## 5.4.1.1. Groat type & characteristics of oat beverage residues

Non-stabilized groats produced a significantly higher percent moisture when compared to stabilized groats but was not significantly different from rolled groats (Figure 12). All three groat types produced significantly (p<0.05) different values of percent ash. Rolled oats yielded the highest percent ash at 2.33%, followed by stabilized and non-stabilized, 2.08 and 1.99, respectively. The protein content analysis showed that rolled groats and stabilized groats produced significantly (p<0.05) higher protein content but were not statistically (p<0.05) different from each other.



Figure 12. Moisture (%) t grouping of means and groat type Mean values in a column followed by the same letter are not significantly different (p<0.05).

Total starch analysis showed that stabilized groats produced the highest percent total

starch, followed by non-stabilized groats, and rolled groats (Figure 13).



Figure 13. Total starch (%) t grouping of means and groat type Mean values in a column followed by the same letter are not significantly different (p<0.05).

#### 5.4.1.2. Water to grain ratio & physical characteristics of oat beverage residues

The average values produced for the 1:4 and 1:6 ratio were 0.74% and 0.91%, respectively. The given values for moisture were not significantly (p<0.05) different. The 1:6 ratio demonstrated a significantly (p<0.05) higher average ash content and protein content when compared the 1:4 ratio. The total starch content for 1:4 and 1:6 was 49.3% and 42.93%, respectively. The percent total starch for 1:4 ratio was significantly (p<0.05) higher than the 1:6 ratio.

## 5.4.1.3. Extraction methods & physical characteristics of oat beverage residues

The moisture content of the amylase treated samples was 1.67%, while wet milling and dry milling followed with values of 0.56% and 0.25%, respectively (Figure 14). The amylase treated samples were significantly (p<0.05) higher than wet and dry milling.





A similar pattern can be seen when assessing the ash content and extraction method. The amylase treated samples produced the highest ash content at 3.51%, followed by wet and dry milling values of 1.45% and 1.44%, respectively. The protein content was significantly (p<0.05)

different among all three extraction methods. The amylase treated samples yielded the highest average protein content at 33.75%, followed by wet and dry milling average values of 13.59% and 12.01%, respectively. The same can be said about the starch content. All extraction methods produced significantly (p<0.05) different average values for total starch. Wet milling extraction produced an average of 55.78% total starch, followed by dry milling and amylase treatment at, 53.85% and 26.57%.

## 5.4.2. Proximate composition of oat beverage residues

#### 5.4.2.1. Groat type & proximate composition of oat beverage residues

Total starch analysis revealed that stabilized groats produced the highest average value of 46.63%, followed by non-stabilized groats and rolled groats. Both stabilized and non-stabilized groats were not significantly (p<0.05) different, while rolled groats produced samples with significantly (p<0.05) less total starch.

The  $\beta$ -glucan content of samples that used rolled groats were significantly (p<0.05) higher then both stabilized and non-stabilized. Rolled groats produced a value of 7.05%, followed by stabilized and non-stabilized groats, 6.24% and 5.75%, respectively (Figure 15).



Figure 15. Free glucose (mg/g) t grouping of means and extraction method Mean values in a column followed by the same letter are not significantly different (p<0.05).

Arabinoxylan, arabinose, xylose, mannose, galactose, and free glucose average values were not significantly (p<0.05) different.

## 5.4.2.2. Water to grain ratio & proximate composition of oat beverage residues

Total starch analysis showed that the 1:4 water to grain ratio yields significantly (p<0.05) higher average values, when compared to the 1:6 ratio. The same could be said of the  $\beta$ -glucan content, with 1:4 ratio producing an average value of 6.66%, followed by 1:6 ratio at 6.05%. The arabinoxylan, arabinose, xylose, mannose, and free glucose average values were not significantly (p<0.05) different.

The galactose composition of the 1:6 ratio was significantly higher than the 1:4 ratio with average values of 4.39% and 2.98%, respectively.

# 5.4.2.3. Extraction method & proximate composition of oat beverage residues

The  $\beta$ -glucan content was assessed, and dry milling produced the highest average value with 7.50%, followed by wet milling and amylase treatment, 5.98% and 5.55%. The dry milling samples average value was significantly (p<0.05) higher than wet milling and amylase treatment.

The samples that underwent amylase treatment produced a significantly higher average value for arabinoxylan, arabinose, xylose content (Table 11). Wet milling produced a significantly (p<0.05) higher galactose content, when compared to dry milling and amylase treatment.

Treatment Type	Water/Flour Ratio	Extraction Method	β- glucan	Arabinoxylan	Arabinose	Xylose	Mannose	Galactose	Glucose
					(%)	)			mg/g
Stabilized	1:4	Dry	7.07ef	1.00e	0.52de	0.61d	0a	1.47ef	41.84abcde
Stabilized	1:6	Dry	9.27b	0.97e	0.52de	0.58d	0a	3.97cdf	32.74cdefg
Non-stabilized	1:4	Dry	8.36c	1.02e	0.53de	0.63d	0a	3.05def	46.75abcd
Non-stabilized	1:6	Dry	3.451	0.96e	0.48de	0.61d	0a	4.58cd	40.15bcde
Rolled	1:4	Dry	10.66a	1.00e	0.54de	0.59d	0a	6.27bc	39.96bcdeg
Rolled	1:6	Dry	6.19gh	0.91e	0.46e	0.57d	0a	4.82cd	34.52cdef
Stabilized	1:4	Wet	6.22gh	1.75bce	0.93bcde	1.06bcd	0a	3.18def	57.53abc
Stabilized	1:6	Wet	5.03j	1.40ce	0.78bcde	0.80cd	0a	10.91a	51.46ab
Non-stabilized	1:4	Wet	5.22ij	1.21ce	0.60de	0.77cd	0a	4.49cd	42.43abcde
Non-stabilized	1:6	Wet	7.76d	2.27abce	1.16abcde	1.42abcd	0a	8.22ab	60.43a
Rolled	1:4	Wet	5.56i	1.60bce	0.82bcde	1.00bcd	0a	2.99def	42.55abcdef
Rolled	1:6	Wet	6.14h	1.31e	0.72cde	0.77cd	0a	4.93cd	37.08cde
Stabilized	1:4	Amylase	4.73k	2.99ab	1.56abc	1.84ab	0a	1.01f	27.92defg
Stabilized	1:6	Amylase	5.14ijk	3.62a	1.99a	2.12a	0a	0.57f	20.16fg
Non-stabilized	1:4	Amylase	4.91jk	2.96ab	1.59ab	1.78ab	0a	3.49cdef	34.93cdefg
Non-stabilized	1:6	Amylase	4.79jk	2.31abce	1.22abcde	1.41abcd	0a	0.94f	17.01g
Rolled	1:4	Amylase	7.11e	2.53abc	1.33abcd	1.54abc	0a	0.85f	27.64efg
Rolled	1:6	Amylase	6.64fg	1.97bce	1.13abcde	1.11bcd	0a	0.58f	16.41g

Table 11.	. Carbohydrate composition of oat beverage residues	

Mean values in a column followed by the same letter are not significantly different (p<0.05).

There was no significant difference between the mannose content among all extraction methods. All samples registered no mannose.

Wet milling produced the highest average free glucose level at 48.58%, followed by dry milling and amylase treatment, 39.32% and 24.01%, respectively (Figure 16).





There are numerous ways the residues are utilized after production. A long-lasting method has been to use the residues as animal feed. Due to the nutrient value, the feed can be nutritious and beneficial for the animals.

The residue post-production is also referred to as pulp. This pulp has been utilized in many other ways. Large companies have been incinerating the residues. This destroys the material and eliminates the cost of material transportation. Over time the practice of incineration has been frowned upon by the public, due to climate change and environmental activism. Today, companies are researching other uses for the residues. These methods include milling the pulp into a flour and reintroducing the flour back onto the process, as well as using the pulp as a nutrient additive for energy and granola bars.

#### 6. CONCLUSIONS

In conclusion, to produce a quality oat beverage, numerous aspects must be taken into consideration. When assessing the groat type, it was shown that stabilized groats would be a better option than both non-stabilized and rolled groats. This is because the heat-treated groats have more available starch for the enzymatic process, as well as inactivating natural enzymes like lipase. The water to grain ratio did not have a major effect on the samples, beside for the percent solids and yield. This was due to the simple fact that the 1:4 ratio had 50g of groat flour, while the 1:6 ratio only had 33.33g. The extraction methods explored included dry milling, wet milling, and amylase treatment. The amylase treated samples tended to be higher in free glucose and total starch, as well has producing the samples with the highest (p < 0.05) degree Brix. Another benefit of the amylase treatment occurs when the product is heated to inactivate the amylase enzyme. This heating dramatically reduces the microbial activity within the product, thus acting as two steps in one. The amylase treatment ensures that there is enough available starch for enzymatic processing, as well as having a degree Brix that closely emulates dairy milk and other benchmark plant-based beverages. To ensure a quality oat beverage, it is recommended that stabilized groats, 1:4 water to grain ratio, and amylase treatment is utilized.

#### 6.1. Future work

The process of oat beverage production has not changed very much since the 1980's. The process has been refined over the past 40 years, because of constant changes in processing and food ingredient technology. This experiment evaluated the ideal parameters to produce an oat beverage. Further research can be conducted into different enzyme formulations and manipulating the timing of said enzyme treatments. Utilizing  $\alpha$ -amylase with protease, or  $\beta$ -amylase could increase parameters like yield and solid content. Once the process of production has been

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finalized, another interesting area to investigate would be focused on food ingredient technology (FIT) and fortification. Experimenting with FIT and fortification formulations could help further the quality of the product. Experimenting to determine ideal sensory properties such as mouth feel and flavor. Also evaluating how consumption of the product effects blood sugar and insulin levels. The extraction of oat β-glucan with reimplementation into other food systems, such as granola bars and energy drinks, could be fascinating as well. Finally, more research could be conducted into packaging and shelf-life. Focusing on these variables ensure that the plant-based alternatives emulate dairy milk in regard to functionality and rheological properties.

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