## UNIVERSITY OF CENTRAL OKLAHOMA

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# Formulation, Optimization and Sensory Acceptance Study of Gluten Free Chickpea Fortified Finger Millet Tortillas

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To my father, Kul Bhakta Shakya and my mother Basundhara Shakya

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### ABSTRACT

Researches have shown potential health benefits of finger millet in many health conditions due to its nutritional content. However, absence of gluten in this flour prevents binding properties required to formulate tortillas. The objective of the study is to formulate, optimize, and perform a sensory acceptability study on chickpea fortified finger millet tortillas. We formulated a flour composition using USDA's standardized tortilla recipe consisting of finger millet and chickpea flour at the ratio of 70:30 w/w, respectively. We further optimized it with 2% sugar, 4% of glycerin and 15% of starch (rice, potato, and tapioca) to enhance functional and sensory properties. A trained sensory panel used a descriptive analysis tool to evaluate sensory acceptability, texture and flavor of the tortillas. The descriptive sensory analysis showed that the tortillas with rice and potato starch were more acceptable in comparison to tortillas with tapioca starch. From the conclusion drawn out of the descriptive analysis, tortillas with tapioca starch were eliminated from the study. The tortillas with rice and potato starch were further characterized by physicochemical and sensory acceptability study. The results showed that there was no significant difference in chemical and nutritional values but had some physical differences. The sensory acceptability study showed that overall likability was slightly higher for tortillas with potato starch in comparison to rice starch which correlates with higher scores for taste, texture and aroma of the tortillas with potato starch. Physical properties showed that the tortillas with potato starch were thicker, smaller and tougher in comparison to tortillas with rice starch which associates with low scores in appearance and tenderness in sensory acceptability study. The results indicated that incorporation of potato starch result in formulation of chickpea fortified finger millet tortillas with acceptable textural and sensory properties which would be a gluten free, nutrient dense alternative to traditional tortillas for people with celiac disease and an alternative food for diabetes patients.

Keywords: Finger millet, Celiac disease, Tortilla, Starch, Gluten free

## **Practical Application**

This research aimed to formulate a gluten free soft-shell tortilla incorporating finger millet, chickpea flour (equally nutritionally beneficial ingredients) and minimal amount of starch. After tortilla formulation was optimized, there were evaluated by adult consumers in a college setting. Findings indicate that use of potato/ rice starch can make a gluten free and nutritional chickpea fortified finger millet tortilla product that is acceptable among the sampled population.

#### **CHAPTER ONE: INTRODUCTION**

Introduction and Statement of the Problem

According to World Health Organization's (WHO) global report (2016) in 2014, more than one in three adults over the age of 18 years were overweight and more than one in ten were obese worldwide. The prevalence of obesity is high in WHO Regions of America implied by the fact that the prevalence of obesity increased with increase in country's income level (WHO, 2016). In Regions of America alone in 2014, 62.8% males and 59.8% females were overweight. Overweight and obesity has been strongly linked to diabetes and the increasing prevalence of obesity has increased incidence of diabetes as well. According to WHO global report (2016) in 2014, there were over 422 million people with diabetes worldwide in comparison to 108 million in 1980 and is expected to reach 592 million by 2035 (Guariguata et al., 2014). United States alone had more than 62 million people with diabetes (WHO, 2016), ranking it the third country with highest prevalence of diabetes globally (Guariguata et al., 2014).

Diabetes is a life threatening conditions and is of major concern, the prevalence of diabetes is so high that American Diabetes Association (ADA) has ranked it to be the seventh leading cause of death in 2010 (American Diabetes Association, 2014). Diabetes not only takes toll of health of the population but also affects the economy of the country. According to a study, the total economic costs of diagnosed diabetes in US in 2012 was \$245 billion (direct and indirect expenditure) and people with diabetes spend 2.3 times more money (in medical expenditure) than a person without diabetes (ADA, 2013). Diabetes is a multifactorial condition which can be genetic or environmental, majority of the cases of this condition are behavioral and environmental. It is caused due to rise of blood glucose levels higher than normal which is affected by the lifestyle and daily food habits of the people. The rise in blood glucose is due to a

genetic condition where there is not enough production of insulin to regulate it or due to physiological condition where people are overweight, obese and insulin produced by the body is not enough to regulate it. The only way to manage or prevent diabetes is a healthy lifestyle incorporating a nutritious balanced diet, physical activity, and maintaining a healthy weight (Center for Disease Control and Prevention, 2014).

According to Food Allergy Research & Education (FARE) 15 million Americas have food allergies and among them 5% are adults and 8% are children. Researches have shown that number of people developing food allergies is growing and increased by 50% in children from 1997to 2011 (FARE, 2016). FARE also puts eight food categories (milk, eggs, peanuts, tree nuts, soy, wheat, fish, and shellfish) that are accountable for 90% of the food allergies. Wheat is a dominant grain in the market and is in the top eight category of food allergies, thus creating a vacuum of food for people with allergies.

Wheat allergy and celiac disease are two different conditions but they have in common is wheat. Celiac disease (CD) is an autoimmune disorder that affects one in 141 Americans in which people cannot tolerate gluten because it damages the inner lining of their small intestine and prevents it from absorbing nutrients (Pietzak, 2012 & Rubio-Tapia, 2012). Wheat contains gluten and hold risk to people with celiac. Studies have also found out that 10% of the people with Type 1 diabetes (T1D) has prevalence of celiac disease in comparison to 0.5% of the general population and also have common genetic precursors (ADA, 2014; Camarca et.al, 2012). It is known that T1D affects bone metabolism and structure and new studies have shown that CD is also an underlining cause in bone impairment (Camarca et al., 2012). In short. osteopenia is a new phenomenon in people with CD, T1D or in two or more autoimmune diseases which can be managed with gluten free diet (Camarca et.al, 2012).

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Food plays a vital role in a human's health and life in general. The health condition of people depends on what they put into their body therefore, food is one of the key component in solving health problems. According to CDC (2014), the majority of the U.S. adult population consumes less than the recommended amount of whole grains and dietary fiber on a daily basis, and less than 5 percent of Americans achieve the average recommended 3-ounce amount of whole grains per day. The USDA 2010 Dietary Guidelines recommend individuals to consume at least half of their grains as whole grain varieties and to replace refined grains with whole grains (McGuire, 2011). Food had negative as well as positive effects in the health of a person; some food tends to degrade the healthy condition where as other help promote health. There are many foods being researched and many more to be explored among them millets are the ones that have been ignored for decades.

The growing trend of influence of food and culture in United States has peaked the demand of tortillas; making the tortilla industry the fastest growing sector in baking industry in U.S. Tortillas were commonly made out of wheat and corn flour; the budding tortilla industry opens up opportunities to explore various legumes and millets as alternative ingredients to provide healthier options. Millets are a good source of phytochemicals, micronutrients, and essential amino acids except lysine and threonine. There have been studies that have shown potential health benefits in many conditions such as diabetes, cardio vascular disease, aging, cancer, celiac disease, and many more (Devi, Vijayabharathi, Sathyabama, Malleshi, & Priyadarisini, 2014). However, studies for optimization of tortilla made of millet are very limited. We aim to study the formulation and optimization of tortillas made with finger millet flour and chickpea flour that could be an alternative to traditional flour and a potential functional food in managing diabetes and celiac disease.

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## **Summary of the Project Proposal**

## **Hypothesis:**

Finger millet flour and chickpea flour can be used to formulate a nutritious tortilla that is gluten free, and a functional alternative to traditional tortillas for people with diabetes and celiac disease.

## **Research Question:**

Can finger millet flour and chickpea flour tortillas be a good alternative to formulate a nutritious gluten free tortillas?

## **Objectives/ Specific Aims:**

The objective of this study is to formulate a nutritious and gluten free tortilla using finger millet flour and chickpea. The specific aims of the study include:

- 1. Formulate and optimize chickpea fortified finger millet tortilla with chickpea flour.
- 2. Evaluate the physical, chemical, nutrition analysis, and sensory analysis of the formulated tortilla.
- 3. Conduct sensory acceptance tests of the tortilla.

#### **CHAPTER TWO: REVIEW OF THE LITERATURE**

#### Diabetes

#### Introduction

WHO global report in 2014 stated that there were over 422 million people with diabetes globally in comparison to 108 million in 1980 and is expected to reach 592 million by 2035 (Guariguata et al., 2014). According to CDC (2014), the chances of developing diabetes is higher in population over the age of 65 in comparison to 20-44 year. Diabetes is a multifactorial condition leading to further complication resulting in loss of vision, cardiovascular events, end-stage renal disease, lower extremity amputations, and many more (WHO, 2016). There are two common type of diabetes Type 1 Diabetes (T1D) and Type 2 Diabetes (T2D); the distinguish between the two is not easy and often requires sophisticated laboratory tests for pancreas function (WHO, 2016). The most common kind of diabetes is T2D which is about 90% to 95% of all the diagnosed cases in comparison to 5% of T1D. Diabetes is just not detrimental to people's health but also their economy. According ADA (2013), in 2012 people with diabetes spent about 2.3 times more money in comparison to those without diabetes, totaling to \$ 245 billion in direct (medical expenditure) and indirect expenses (disability, work loss, premature death).

Type 1 Diabetes (T1D) also called insulin dependent diabetes mellitus which is developed when beta cells that produce insulin in pancreas are destroyed. Onset of T1D can be at any age however, the peak age of diagnosis is mid-teen. People with T1D have to use insulin pump or injection to survive.

Type 2 Diabetes (T2D) also called non-insulin dependent diabetes that begins with insulin resistance where cell within muscles, liver and fat tissues do not use insulin properly.

Insulin resistance is developed which creates a rise in need of insulin leading to inability of beta cells to produce insufficient quantity. Onset of T2D is later than that of T1D and occurs mostly in people with a family history, obesity, impaired glucose metabolism, physical inactivity, race/ ethnicity and old age. The change in lifestyle, genetics and environment has drastically increased incidence of Type 2 diabetes.

Gestational diabetes onset during second or third trimester of pregnancy. The increase in blood glucose during pregnancy escalates the risk of both mother and fetus. It requires treatment which would include diet, physical activity and insulin. Women with gestational diabetes have 5% to 10% chances of having type 2 diabetes later in life and the children of the women who had gestational diabetes during pregnancy have higher risk of developing diabetes or obesity.

## **Symptoms**

Unmanaged blood glucose level can create several different symptoms in many parts of a human body. The symptoms of diabetes can be severe or mild varying upon people but are very mild in cases of T2D. Some of the common symptoms of diabetes are frequent urination, feeling of thirst, feeling of hunger (even during meals), extreme fatigue, blurry vision, cuts/bruises with slow healing, weight loss (even with increase in diet in T1D), tingling, pain, or numbness in the hands/feet in people with T2D (ADA, 2015).

#### Diagnosis

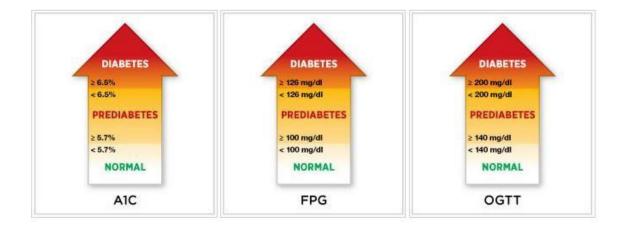
#### A1C

This test measures blood glucose for the past 2-3 months without fasting or drinking anything. Fasting Plasma Glucose (FPG)

This test checks the fasting blood glucose levels for which patients are not allowed to eat or drink at least 8 hours prior to test.

#### Oral Glucose Tolerance Test (OGTT)

This test checks blood sugar level before and after the consumption of a sweet drink which shows glucose processing in the body that aids in diagnosing diabetes.



#### Source: Diabetes.org

#### Figure 1: Chart describing diagnosis of diabetes in different test.

#### Treatment

There is no cure for diabetes; it is a multifactorial disease and self-management of diet and physical activity is the key to control and manage it. People with T1D must have insulin pump or get injections to survive, which is also true for people with severe case of T2D. People with T2D can manage their blood glucose by adapting healthier diet, regular physical activity, losing weight, medication and healthier lifestyle in general.

## **Co-existing Conditions**

Diabetes can onset numerous complications and affect several parts of a human body. Maintaining blood glucose is a major concern because low or high glucose level can result in organ failure and be as severe as death. Early diagnosis and treatment can help reduce risk of other health complications. According to CDC (2014) in 2011 about 282,000 people ended up in emergency due to hypoglycemia and 175,000 people due to hyperglycemia. People with Diabetes are also at a high risk of high blood pressure and high cholesterol along with heart disease and strokes. In 2010, the number of people hospitalized due to stroke was 1.5 times higher with people with diabetes in comparison to people without diabetes (CDC, 2014). Other complication with diabetes is related to microvascular disease concerned with kidney, eye and nerves. Uncontrolled diabetes has resulted in about 73,000 non-traumatic lower limb amputation in people with diabetes in 2010, also has been accounted for 228,924 kidney failures, and 4.2 million people over the age of 40 years have suffered retinopathy resulting in loss of vision due to diabetes. According to CDC, 2014 undiagnosed, untreated and uncontrolled diabetes have caused 234,051 deaths in 2010 alone.

#### **Food Allergies**

#### Introduction

USDA define food allergy as "a specific type of adverse food reaction involving the immune system. The body produces what is called an allergic, or immunoglobulin E (IgE), antibody to a food. Once a specific food is ingested and binds with the IgE antibody, an allergic reaction ensues." (USDA, 2011). According to American College of Allergy, Asthma & Immunology (ACAAI) 2014, there are over 50 million American that have some kind of allergies and food allergies are estimated to affect 4%- 6% of children and 4% of adults. Allergies can be caused by any food that cannot be tolerated by a person, there are more than 160 foods known to cause allergic reaction (USDA, 2011) but Food Allergen Labeling and Consumer Protection Act of 2004 has identified eight major foods that account for 90% of those reaction. Those eight foods are milk, eggs, fish, crustacean shellfish, tree nuts, peanuts, wheat and soybeans.

#### **FDA and Government Regulations**

The government of United States has passed the Food Allergen Labeling and Consumer Protection Act of 2004 (FALCPA) in August 2, 2004 to help American avoid health risks due to food allergens. FALCPA declared the major allergen in the Act of 2004 which was effective in January 1, 2006, the act requires product to contain all the ingredient list by common name in descending order of predominance (USDA, 2011), but the law is only limited to consumer packaged foods regulated by Food and Drug Administration (FDA).

Food allergies are not only caused by the presence of allergens as some ingredients but can also be caused from unintentional cross contaminations. FDA has provided guidance for the food industry to provide advisory allergen statements like "may contain [allergen]" or "produced in a facility that also uses [allergen]" to reduce consumer's health risks. However, the act passed by FALCPA does not apply for meat, poultry and egg products. Food Safety and Inspection Service (FSIS) does encourage the use of allergen statements for public health concerns and are considering amending the act (USDA, 2011).

#### **Symptoms**

Some of the most common food allergy symptoms are appearance of hives, flushed skin or rashes, tingling or itchy sensation in mouth, swelling of face tongue, or lips, vomiting, diarrhea, abdominal cramps, coughing or wheezing, dizziness, lightheadedness, swelling of the throat and vocal cords, difficulty in breathing, loss of consciousness and anaphylaxis (USDA, 2011)

#### Diagnosis

Most of the food related reaction occurs within two hours of consumption of food but can also can be as quickly as few minutes or within six hours (ACAAI, 2014). Diagnosis of food allergies are complicated because the test cannot conclusively determine the cause of the allergy or the allergen. When a food allergy is suspected, the person need to consult an allergist and perform analysis to confirm since allergies can be life threatening. The diagnosis process includes detail questionnaire on allergy symptoms, onset time, etc. which would direct blood test and skin prick food allergy tests, that will indicate weather food-specific IgE antibodies are present in the body. There is another way of diagnosing which is expensive, time consuming and potentially dangerous; it is the oral food challenge where patients are fed tiny amount of suspected allergen in increasing dose over the period of time under strict supervision. The results of these tests narrow down the food search causing allergies and is placed on a special diet.

#### **Management and Treatment**

The primary way of managing allergies is avoiding food that initiates it. People with diagnosed food allergies need to pay attention to ingredients, read food labels before consuming it. FALCPA mandated manufactures to list the ingredients as well as chances of cross contamination during manufacturing to avoid incidence of food allergy. Allergies are hard to classify as mild or severe, the level of allergy is determined by the environment and amount of allergen consumed. Allergic reaction can be as severe as anaphylaxis that impairs a person breathing and drops their blood pressure which can also be fatal. The first-line of treatment for it is epinephrine (adrenaline), people diagnosed with allergies should prescribe epinephrine auto-injector and carry it with them with at least two doses available in it. Epinephrine might have some adverse effect in people which needs to be addressed with their physician but even with those effects it is the best bet over anaphylaxis (ACAAI, 2014). In November 2013, President Barack Obama signed a law "School Access to Emergency Epinephrine Act" that requires

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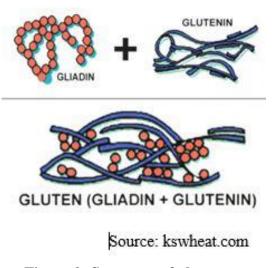
schools to have epinephrine auto-injectors (epipen) on hand, by 2014 over a dozen states adopted the law and required schools to have supplies for general use (ACAAI, 2014).

There is no proven cure to allergies but children are seen to outgrow allergies as they grow. Researches have shown that timing in introduction of certain food and eliminating solid foods with babies before 17 weeks' old have prevented onset of food allergies in children.

## Gluten

#### Introduction

Gluten is a protein fraction commonly found in wheat (giladin), rye (hordein), barley (secalin) and triticale (Welstead, 2015). It helps in maintaining the structure and holds the food together in a product, which is well defined by its name gluten meaning glue in Latin (Welstead, 2015). The very common source is wheat and its derivatives like durum, emmer, semolina, spelt, graham, kamut (Elli, 2015). Wheat gluten was first



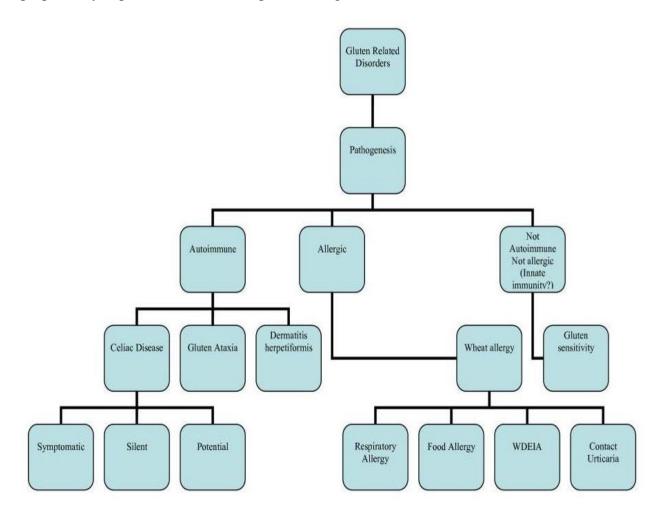
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#### Figure 2: Structure of gluten

isolated in 1745 advancing production and its utilization. According to Welsted (2015), it was in 1941 between World War I and World War II, Nutrition Society in Britain promoted increase in global production of wheat that possibly contributed to increased prevalence of diagnosis of Celiac Disease.

#### **Gluten Related Disorder**

Wheat is the most common grain grown around the world, the availability of the grain and its functional property that gluten provides had made it primary ingredient in many foods (Sapone et al, 2012). Along with wide acceptance and use of wheat, there are several health conditions that has been linked to its consumptions due to the presence of gluten. The recognized health condition mediated by gluten in wheat are wheat allergies (WA) and celiac disease (CD). There are conditions where people react to gluten neither due to allergic reaction nor involves autoimmune mechanism, but are simply intolerant to gluten also known as gluten sensitivity. The gluten induced conditions can be the result of a diverse condition ranging from immune system reaction to environmental triggers. A nomenclature and classification of gluten-related disorder proposed by Sapone et al (2012) is reported in Figure 3.



Source: Sapone et al (2012)

## Figure 3: Proposed new nomenclature and classification of gluten-related disorders.

#### Pathophysiology

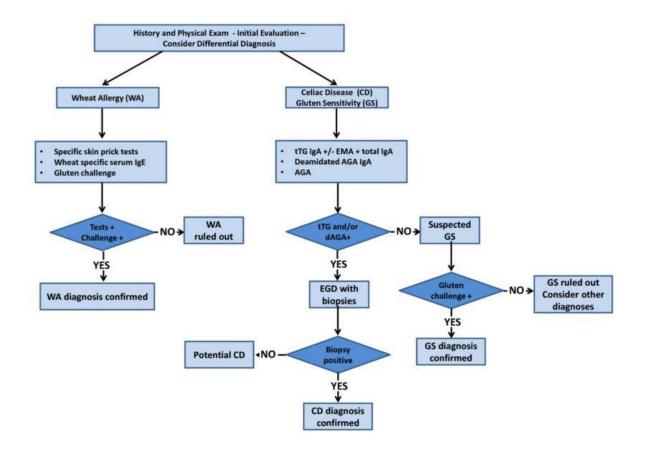
The gluten-related disorders are triggered by gluten present in products. Gluten have different physiological reaction in the body corresponding to different pathophysiology in these disorders. Wheat allergy is triggered by cross-linkage of immunoglobin (Ig)E by repeat sequence of gluten peptide that release the chemical reaction whereas CD is an autoimmune disorder that damages the small intestine (Sapone et al., 2012). Conditions like wheat allergies and gluten sensitivity develops adverse physiological reactions like discomfort and anaphylaxis whereas CD causes damage to small intestine.

## **Symptoms**

The symptoms for gluten-related disorder varies from different condition, it can be as minor as abdominal pain, headache, rashes, foggy mind, fatigue, diarrhea, depression, anemia, joint pain, numbness in hands and legs to anaphylaxis to death (Sapone et al., 2012)

#### Diagnosis

The diagnosis of gluten related disorders can be done by a combination of clinical, biological, genetic, and histological data (Sapone et al., 2012). The algorithm to differential diagnosis of gluten related disorder is shown in Figure 4.



Source: Sapone et al (2012)

# <u>Figure 4: Proposed algorithm for different diagnosis of gluten-related disorders.</u> Gluten Free Diet

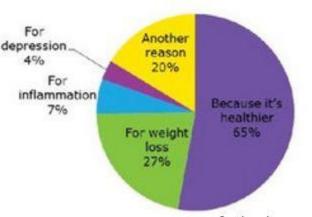
The general cause of gluten-related disorders is gluten so the treatment is gluten free diet (GFD). Excluding grain/cereals containing gluten and substituting it with other grain without gluten is the most effective way of treating gluten-related disorders. It was in 1934-1936 and 1950 Willem-Karel Dicke found out gluten was the cause for symptoms like anorexia, increased bowel movement and steotorrhea and experimented with wheat-free diet providing a framework for GFD. Medication is not normally prescribed to people following GFD except in cases of dermatitis herpetiformis to prevent rashes but adaptation of GFD increases the risk of calcium,

iron, zinc, B6, B12 and folate deficiency, so physicians test bone density frequently and prescribe gluten free dietary supplement (NIH, 2016).

#### **Market for Gluten Free Products**

According to Watson (2014), Mintel Group Ltd. reported the gluten-free market is projected to reach \$15 billion in sales this year. This market valuation takes into consideration not only products that are made specifically to be gluten-free, but those that are naturally gluten-free and are packaged and labeled as gluten-free. Alternatively, Packaged Fact, a food research

Base: 247 internet users aged 18+ who eat gluten-free foods or used to eaten gluten-free foods for reasons other than intolerance/sensitivity



Source: foodnavigator-usa.com

#### Figure 5: Consumer of gluten-free foods

company, has a similar definition of what it means for products to be gluten-free, but values the market around \$10 billion (Watson, 2014). However, the key similarity in research is market growth in the double-digits, projected at anywhere between 38-48% from 2013-2018 (Crawford, 2015). The main reason for this explosive growth in the gluten-free products market is associated with healthier options. With increased growth for healthier food options, there is growing demand for foods that replace traditional product offerings. For these reasons, gluten-free foods are no longer just considered food for people with celiac disease, but for anyone looking to have a healthier diet. With gluten-free products perceived as being healthier, there has been an increase in sales of gluten-free foods that serve as a replacement for products containing wheat. However, despite the growth in the market, there are still challenges that face the market, such as the inability for wheat flour replacements to replicate the texture of flour-based products, and the decrease in

nutrition with the addition of starches in order for the product to resemble flour-based products in texture.

#### **Finger millet**

## Introduction

Millets are small seeded species of cereal grains widely grown around the world and is the sixth most important grains (Premavalli, 2012). Millets are ancient grain that had been cultivated as early as 2700 BC in China, it has a relatively low cost, cultivation environmental and because of peoples' preference on rice and wheat over millet it is sometimes referred as "poor man's cereal" (Premavalli, 2012; Shobana et al., 2013). Millets are used as cattle feed in developed countries, and are not widely explored and are underestimated for its potential. These crops have resistance to pests and diseases, can grow under heat and drought conditions and have short growing season making these the most important crop in tropical and semiarid regions (Premavalli, 2012). The nutrition composition of the millets is comparable to other cereals, moreover they are superior source in terms of dietary fiber, minerals, B-vitamins, starch properties and physiological action (Shobana et al., 2013; Premavalli, 2012).

There are six types of most common and important millets. Amongst them finger millet has the highest amount of calcium, potassium, sodium, dietary fiber and iron (Shobana et al., 2013). Finger millet is very sparsely consumed worldwide besides in India, and eastern and central Africa where it is one of the staples. India is also the world's leading producer of finger millet. There are several in vitro and in vivo studies over health benefits of finger millet. Regular consumption of finger millet can lower the risk of cardiac problem, diabetes, gastrointestinal cancer and is a substitute cereal for patients with celiac disease due to absence of gluten (Shobana et al., 2013)

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## History

There are different theories on the origin of finger millet, some studies show that it originated in India and later migrated to Arabia and Africa about 3000 years ago and some show it originated in Africa and migrated to India. According to Fuller (2002, 2003) it originated in Africa which is supported by a linguistic evidence of the term "ragi" whose root source term is from language used in Africa. However, the earliest report on finger millet dates back to about 2300BC in Karnataka, India (Shobana et al., 2013). The scientific name of finger millet is *Eleusine coracana*, named after the Greek goddess of cereal Eleusine. Its common name finger millet was given to it from its branching of panicle that looked like fingers on the hand.

## Classification

Kingdom	Plantae
Division	Magnoliophyta
Class	Liliopsida
Order	Poales
Family	Graminae
Subfamily	Chloridoideae
Genus	Eleusine
Species	Coracana
	Source: Tamil Nadu A

Source: Tamil Nadu Agricultural University (2011)

# Table 1: Classification of finger millet

#### **Appearance and Structure**

Finger millet is a crop that falls under minor cereal grass family, it is 2 to 4 feet in height with a firm and fibrous roots system (Premavalli, 2012). The root system as well as the leaves are strongly keeled which makes it difficult to break (Van Wyk & Van Oudtshoorn, 1999). The stem of the plant is slender, erect and clumped together at the base and sometimes branches. It is green in color, hollow at internodes and solid at nodes with inflorescences at the terminal nodes (TNAU, 2011). The leaves of the plants are 220-500mm long and 6-10mm wide; are green in color, flattened, splits along the entire length and envelopes the stem with very little exposure at internodes (Van Wyk & Van Oudtshoorn, 1999). The well grown leaves tend to snap and bend down. The inflorescences at the end of the shoots branch out into few branches and consist of number of spikes ranging from 3 to 20. The spikes of the plant resemble to that of finger on a hand along with an odd one littler lower down the whorl which is also known as the thumb (TANU, 2011). The spikelet is often curved and crowded with 2 to 4 flowered enclosed by lemma and palea. The flowers are bisexual; opens one per day from bottom to top and it takes 4-8 days for flowering (Premavalli, 2012). The finger millet grain is globose, smooth and spherical that is slightly flattened at the base with a small depression called hilum. The outer layer of the grain is bot fused and can be easily removed (Wyk & Gericke, 2000); the color of the seed varies and can range from brown, reddish brown, black, orange, purple to white (TAMU, 2011).

#### **Environmental Condition and Production**

Finger millet is an annual crop that can be grown from tropical to arid climates and has a distinct ability to survive in less fertile soil (Hulse, Laing, & Pearson, 1980). According to National Research Council (1996), finger millets have been cultivated in parts of Africa where the rainfall is as low as 300 mm per annum, it is tolerant to cool climate but thrives in hot

conditions. Finger millets takes about 2.5- 6 months from germination to maturation, it has the ability to utilize rock phosphate and can grow in any soil with reasonable water holding capacity (National Research Council, 1996). According to Premavalli (2012), finger millet is a highly productive crop and yields anywhere from 1300- 5000 kilograms per hectare with a short growing season, resistant to drought conditions and resistance to pest. Finger millet is less susceptible to pest and disease and its seeds can be stored for years without any insect damage however, there is a fungal disease known as "blast" that can destroy an entire field (National Research Council, 1966).

The root system of finger millet is exceptionally strong which makes it hard to pull out from the ground by hand (Van Wyk & Van Oudtshoorn, 1999); people growing them in countries like Africa and India have to invest a lot of labor hours to this task. The labor hours are considered the most important in these rural parts of the country which is one of the reason for farmers to cultivate some other crop like maize instead of finger millet (FAO, n.d.).

### **Nutritional Content**

Studies were conducted on 76 varieties of finger millet from all over the world for years and the nutritional compositions of finger millet were determined; it consists of 73 to 82% of carbohydrate, 4 to 8% of protein, 1 to 4.5% of lipid, 200 to 450mg calcium, 5 to 15mg iron, 0.4 to 4mg B-vitamins, 3 to 12% crude fiber and seven essential amino acids (Premavalli, 2012). The carbohydrate composition of finger millet consists of 15 to 20% of dietary fiber and 2 to 4.5% free sugar and mostly starch consisting of amylose, amylopectin, and other starch fractions (Shobana et al., 2013)

Finger millet is an incomplete protein because of deficit of essential amino acids (lysine and threonine). But it is rich in glutamic acid which is 16 to 21% of the total amino acid content

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in finger millet and it fulfill 50% of WHO recommendation. The common amino acids found in finger millet are isoleucine, leucine, methionine, phenylalanine, valine, tryptophan, and histidine (Premavalli, 2012).

Lipid content in finger millet ranges from 1 to 4.5%, lower than other millet and wheat grains which not only influences nutritional value but also favors the stability of the grain. According to Mahadevappa and Raina (1978) finger millet has 70 to 71% of neutral lipids, 10-12% of glycolipids and 5-6% of phospholipids.

Finger millet is also a very good source of micronutrients especially calcium and iron. According to Gopalan et al., (2009), it has 344mg% of calcium, 3.9mg% of iron and 283 mg% of phosphorus in comparison to other cereal grains and millet. Finger millet is high in phosphorus which results in significant amount of phytic acid which is a main storage form of phosphorus. Phytic acid is an anti-nutrient since it impairs the absorption of zinc and calcium which will lead to mineral deficiency in a long run. The phytic acid content of finger millet is lower in comparison to many other cereal grain and is not of major concern because according to Sripriya et al (1997) phytic acid can be decreased by 60% and the bioavailability can be improved by germination and fermentation of the grain.

Along with micro and macro nutrients finger millet also contains non-nutrients that has health benefits even if they were once considered anti-nutrients. Non-nutrients are compounds that do not contribute to nutritional strength but evert positive and non-positive action resulting health benefits or inhibitory action. One of the major non-nutrients found in finger millet are polyphenols. Phenols are large and diverse compounds that do not play any role in nutrition but have antioxidant, anti-mutagenic, anti-oestrogenic, anti-carcinogenic, and anti-inflammatory properties that are beneficial in preventing and minimizing incidence of diseases.

	Whole wheat	Finger millet	Dias A.	Whole grain
	flour	flour	Rice, flour flour	sorghum flour
Carbohydrate(g)	71.9	72	80.13	76.64
Fat (g)	2.5	1.3	1.42	3.34
Protein (g)	13.21	7.3	5.95	8.43
Fiber (g)	10.7	11.5	2.4	6.60
Kcal	340	328	366	359
Phenols (mg)	20.5	102	2.51	43.1
Starch (%)	64	59.0	77.2	73.8
Calcium (mg)	34	344	10	12
Phosphorous (mg)	357	283	98	278
Iron (mg)	3.6	3.9	0.35	3.14
Magnesium (mg)	137	137	35	123
Zinc (mg)	2.6	2.3	0.80	1.63
Potassium (mg)	363	408	76	324
Potassium (mg)	363	408	76	

Source: USDA, Shobana (2013), Devi (2011).

# Table 2: Comparison of finger millet flour with whole wheat, rice and whole grain sorghum

flour. Nutrients and minerals in 100 g of edible portion.

#### **Health Benefits**

The rise in obesity and related health conditions has increased the demand of foods containing high dietary fiber and phytochemicals such as phenols (Devi, et al., 2011). Whole grain cereals such as finger millet are a good source of phytochemical and dietary fiber. The polyphenols and dietary fiber content in the finger millet attributes to several health benefits due to its antioxidative, antimutagenic, and antiglycemic properties. Researches has shown that regular consumption of finger millet aids in controlling postprandial blood glucose surge which is attributed to its phenolic content. Phenols in finger millet partially inhibits amylase and glucosidase during enzymatic hydrolysis of complex carbohydrate delaying absorption of glucose, controlling postprandial blood glucose levels (Shobana et al. 2013). The high dietary fiber content in the finger millet slows gastric emptying and forms un-absorbable complex with carbohydrate in the gut delaying absorption of carbohydrate which aids in controlling blood glucose as well as reducing risk of gastrointestinal disease (Kawai et al. 1987). In a study on diabetic rats it was found out that feeding them finger millet controlled their blood glucose level and improved antioxidant status hastening their wound healing process (Rajasekaran, Nithya, Rose, Chandra, 2004). The increased levels of reactive oxygen species (ROS) in damaged cells leads to necrosis and converts superficial wounds into deeper wounds (King, 2001), which is prevented by antioxidants from finger millets and stimulate wound healing.

#### **Current Products**

Finger millet is widely consumed and staples in southern Africa and India. It is a very versatile grain that can be used in many different types of foods. It can be fermented, germinated, puffed, milled and baked or cooked into food products. Finger millet is an ancient grain and most of the products were traditional foods which have diverse into commercial products.

Traditionally finger millet was considered poor man's food and used to make staples like unleavened bread, porridge, finger millet balls, and some non-alcoholic as well as alcoholic drinks. In the last two decades the properties of finger millet have been in the lime light which has contributed to new products being manufactured. There are nearly 40 processed foods that have been documented most of which are in India. (Premavalli, 2013)

## **Principles of Flour Tortilla Production**

#### Market

The tortilla industry is the fastest growing sector in the baking business with recorded sales of \$12 billion in 2015 overtaking white sandwich bread sales (Kabbani, 2016). The versatility of the tortilla has surged it into an alternative of bread. Tortillas are also popular among health-conscious consumers due to its short ingredient list. (Schafer, 2015) According to Schafer (2015), the shift of food trends to multigrain and whole grain has increased demand and consumption of wheat tortillas significantly. Wheat tortillas are the most popular followed by corn tortillas mainly due to preference of Hispanic population (Schafer, 2015). Moreover, we cannot neglect the love of Americans for Mexican food which has amplified the sale of tortillas in fast food restaurants. The future of tortillas is growing attributed to its versatility opening new markets in Europe, Asia and parts of Africa, along with its adoption into Special Nutrition Program for Women, Infants, and Children (WIC) program (Schafer, 2015).

#### Process

There are three different methods for flour tortilla production: hot press, die cut and hand stretch (Qarooni, Posner, Ponte, 1993). The process of making tortillas varies with the ingredients used as well as the purpose of the production. According to Bello, Serna Saldivar, Waniska, Rooney, (1991) hot press is the most common method in commercial production. Dough mixing is the first and the most important step in tortilla production. Tortilla dough can be mixed in two different ways; first method mixes all the ingredients together. Second method mixes ingredients in three different steps; dry ingredients are first mixed in then shortening are added in and mixed and finally water is added until dough is developed (Bello et al., 1991). The amount of water and mixing time varies with ingredient and affects the properties of dough. According to Bello et al. (1991), the dry ingredients are mixed for 2 minutes in slow speed and then shortening is added in and mixed in for another 8 minutes. Finally, water is added and mixed for 1 minutes at low speed and slowly increased at medium until dough is developed. Warm water (38 to 52°C) is added to the mixture to produce a dough temperature of 30 to 36°C (Bello et al., 1991). The dough is then rested for 2 minutes, then divided into 50g dough balls and rested for 5 minutes. Pressing and resting of the dough is different between different methods, according to Bello et al. (1991) 750-1450 psi pressure for 1.4 sec is applied to flattened the dough ball.

The temperature and pressure of the press ranges from 177 to 237° C and 278 to 758 N/cm<sup>2</sup> respectively (Janson 1990), which creates a skin on the tortilla restricting the amount of steam and carbon dioxide released during baking making it puffy. Tortillas are baked in a conveyor belt in between 260°C for 17 to 40 seconds, according to Bello et al. (1991), hot press tortillas are baked in at 232°C the middle tier in 40 seconds. Cooked tortillas are then cooled in similar conveyer belt; the cooled tortillas are then packaged.

#### Ingredients

#### Flour

The properties of the flour affect the quality of the tortilla in many ways. According to Waniska et al. (2004) flour properties should contain protein content (10-12%) of intermediate

quality and low levels of starch damage to produce good quality tortilla (as cited in Barros et al., 2010). The flour requirement for tortillas also depends on its cooking procedure. Tortillas manufactured by hand stretch and hot-press requires lower protein content (9.5-11.5%) where ones manufactured by die-cut requires high protein content (11.5- 14%).

#### Water

Water is a crucial ingredient in any baked product, it effects the properties of dough, aids in gluten development and activates leavening agents (Serna-Saldivar, Rooney, Waniska, 1988). Water aids in dissolving and rehydration ingredients which help in physical and chemical changes like starch gelatinization (Minarro, Albanell, Aguilar, Guamis, Capellas, 2012). The variation of water in a formulation is dependent on the ingredients being used in the formulation. Warm water is generally prefered to create an optimum dough temperature.

#### Fat

Shortenings reduce dough stickiness, improves shelf-life, adds flavor and palatability to a product (Serna-Saldivar, Rooney & Waniska, 1988; Minarro et al., 2012. Shortening is the most commonly used fat in tortillas but according to Bejosano, Novie& Waniska (2006) good quality tortillas can also be made without the use of hydrogenated shortening. Unmodified oils that are liquid at room temperatures can be used to make good quality tortillas considering some changes in processing conditions as a result of softer dough with use of liquid fat (Bejosano et al., 2006). In gluten-free breads, the fat-liquid provides and extra interfacial source that allows bubble formation and expansion by stabilizing gas cells (Minarro et al., 2012).

#### Salt

Lipolytic activity in flour reduces baking quality during storage of a product which can be reduced by addition of salt. Salt reduces hydrolytic and oxidative rancidity giving better storage results (Doblado-Maldonado, Arndt & Rose, 2013). Salt also acts as a preservative limiting bacterial growth and enhancing flavor of the tortillas (Quilez et. al, 2006).

#### Sugar

Addition of sugar in the formulation not only aids in enhancing the flavor of the product it also assists in incorporating air cell resulting in better texture. It also retains moisture prolonging freshness (Minarro et al., 2012).

#### Leavening Agent

Sodium bicarbonate, potassium bicarbonate and ammonium bicarbonate are some of the most commonly used leavening agents in baked cereal products (Bejosano & Waniska, 2004). The leavening agents produce carbon dioxide into the dough creating bubbles resulting in expansion of the dough. The carbon dioxide gas also influences the flexibility, opacity, even flavor and thickness of the tortilla (Cepeda, Waniska, Rooney & Benjosano, 2000). Leavening agent when used in gluten–free dough increase volume and aid in softer texture during baking (Minarro et al., 2012).

#### Starch

Starch is the main component of bread and plays a vital role in structure and mechanical properties (Ziobro, Korus, Witczak, & Juszczak, 2012). According to Miyazaki, Hung, Maeda, & Morita (2006), 75- 85% of the flour is starch which dilutes gluten to a suitable level and absorbs water from gluten by gelatinization giving a structure for gas formation and retention so that the bread does not collapse while cooling. The role of starch is even more essential in case of gluten-free baking. Starches act when the temperature increases by competing with other components available in the system forming a gel. Starches can be extracted from various sources including cereals, roots and tubers. The main sources of starch are corn, rice, wheat and

potato which have significantly different in structures, compositions, physio-chemical, and functional properties (Singh, Singh, Kaur, Sodhi, & Gill, 2003).

Starch is a major polysaccharide in plants that form a semi crystalline and amorphous region in an alternative fashion varying in levels of crystallization (Jerkins & Donald, 1994). Starch consists of copolymers of linear chain amylose, branched amylopectin biopolymers represented in the form of amorphous region and crystallinity respectively along with lipids, proteins, potassium, phosphorus, phospholipids, and many more compounds (Vandeputte & Delcour, 2004; Singh et al., 2003). The presence of these compounds significantly affects the functional properties of the starches. The higher phosphate monoester content in potato starch results in pasting with higher light transmittance (higher paste clarity) whereas higher phospholipids in cereal starch (rice) results in pasting with lower transmittance (Singh et al., 2003).

The presence of phospholipids not only affect the light transmittance it also effects the swelling power and solubility of the starches. Phospholipids have a tendency to form a complex with amylose and amylopectin which limits swelling as well as solubility of the starch. The higher content of phospholipid complex in cereal starches (rice, wheat, tapioca) reduces swelling power, solubility and transmittance. In contrast higher swelling power, solubility and transmittance in potato starches relates to high phosphate group on amylopectin which repulses phosphate on adjacent chains increasing hydration and weakening bonding within crystalline domain (Galliard, 1987; Singh et al., 2003).

#### **Preservatives and Additives**

Studies have shown that gluten contributes significantly to shelf stability and aids in resisting breaking during consumption in tortillas (Wang & Flores 1999 and Pascut & Waniska

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2004). Tortillas require longer shelf-life than bread because they are not consumed on the day they are baked (Mondal et al., 2009). However, the higher retention of protein functionality and decreased starch dispersion and firming gives tortillas longer shelf-life in comparison to bread (Bejosano & Waniska, 2004). According to Friend, Serna-Saldivar, Waniska & Rooney (1992) propionate, sorbate and sufficient acidulate (fumaric, citric, phosphoric, etc) can be used to achieve longer shelf-life by reducing the pH of tortillas.

#### **Structural Development and Quality Assessment**

Gluten is the protein fraction found in wheat, barley, rye, oats and their crossbred varieties that plays a vital role in overall structure and quality of baked cereal products (Badiu, Aprodu, & Banu, 2014). Dry gluten consists of 75-86% protein and the remaining are carbohydrate and lipid (Bloksma, 1971). The gluten protein forms a continuous network which is responsible for elasticity and extensibility and retains carbon dioxide to provide structure to the baked products (Badiu et al., 2014). Gluten is composed of high molecular weight and low molecular weight glutenin and gliadins and their allelic variants (Mondal et al., 2009). Glutenins and gliadins have different functional properties which interact to develop network that provides quality and strength to the dough (Mondal et al., 2009). Gliadins are soluble in aqueous alcohols and form a viscous fluid mass when it isolated and hydrated (Plyer, 1988) whereas isolated glutenins form a tough, rubbery mass when hydrated. The rubbery mass produced during hydration of glutenins is responsible for elasticity of the dough. Along with the structure of baked products, gluten also play a vital role in texture, with 16% water gluten has a glass transition temperature at room temperature which aids in producing an elastic dough while mixing (Hoseney, 1994).

According to Waniska (1999), the ideal tortilla should be evenly opaque, have about 2 mm thickness with ample diameter and have at least 3-weeks shelf life. The air bubbles produced from gluten network when hydrated affects the opacity of the tortillas. Larger the number of smaller air bubbles the lighter diffraction giving tortillas its opacity (Serna-Saldivar, Rooney & Waniska, 1988). The diameter is another characteristic of tortillas that requires extensible dough that could resist shrinkage during processing (Bejosano & Waniska, 2001). Studies have suggested that gliadin is associated with extensibility and loss of strength in the dough which is compensated by glutenin that increases dough strength (Edward, Dexter & Scanlon, 2001 and MacRitchie, 1985). According to Mondal et al. (2009), functionality of gliadin may contribute to a good quality tortilla that require extensibility which can be achieved with high or low gliadin content depending on the allele.

The diameter of tortillas also requires extensible dough that resists shrink-back during processing. The dough extensibility, in-turn, depends again on the gluten proteins. Thus, dough extensibility during hot pressing and retention of tortilla flexibility after baking requires a gluten functionality that is unique to the strong viscoelastic gluten functionality needed for bread.

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#### **CHAPTER THREE: METHODOLOGY**

#### **Material and Methods**

#### **Preliminary Research**

According to Deora, Deswal & Mishra (2014), most of the commercial gluten-free mixes usually contain carbohydrate, the substitute gluten-free flour, as well as addition of different starch to replace absence of gluten which increases the amount of carbohydrate and restrict other nutrients. The lack of nutrients in gluten-free diet has led to some cases of malnutrition in people with CD (Marco & Rosell, 2008). The objective of this research is to formulate a tortilla using two nutritionally superior grains: finger millet and chickpea to produce a nutrient dense product that accompanies several essential health benefits.

Chickpea is a legume (*Leguminosease* family) that originated in Asia, it contains high amount of protein (23-27%) and lipids (5.8-6.2%) compared to other legumes (Dodok, Abid, Hozova, Halasova & Polacek et al., 1993). Chickpea is high in lysine which makes it an excellent protein to complement finger millet which is deficient in lysine. The combination on finger millet flour and chickpea would be a good match to fulfill the requirement of all the basic macro and micronutrients. The struggle in formulation of the tortilla is the absence of gluten in these flour. However, according to Boye, Zare & Pletch, (2010), the specific amino acids content of chickpea has characteristic of high foam expansion and stability in comparison to other legumes which is beneficial in gluten-free product development. The aim in formulation of the tortillas was to use minimum amount of starch to substitute absence of gluten and produce a tortilla that would be acceptable to consumers. Chickpea was the most appropriate alternative for enhancing the nutritive value along with improvement in physical and overall quality of the gluten-free tortillas.

	Chickpea
Carbohydrate(g)	12.2
Fat (g)	6.04
Protein (g)	20.47
Fiber (g)	12.2
Kcal	378
Calcium (mg)	57
Phosphorous	252
( <b>mg</b> )	
Iron (mg)	4.31
Magnesium	79
( <b>mg</b> )	
Zinc (mg)	2.76
Potassium (mg)	718

Source: USDA

#### Table 3: Nutrients and minerals content of Chickpea in 100 g of edible portion.

Finger millet is high in many micronutrients and dietary fiber but is low in protein in comparison to wheat (7.3 g in comparison to 11.6 g of protein). In order to reach the targeted 12.1 g of protein content per 100 g, we added chickpea flour at the ratio of 70:30 w/w, finger millet/chickpea. Once the base of the flour was finalized we took USDA's standardized tortilla recipe and substituted wheat flour with the flour mixture of finger millet and chickpea. The standardized recipe calls for salt, baking powder, shortening, and water.

Ingredients 3.5	Amount in us metrics
Flour	2 cups
Baking powder	1 tsp.
Salt	<sup>1</sup> ⁄ <sub>2</sub> tsp.
Oil	2 tbsp.
Warm Water	<sup>3</sup> ⁄4 cup
	Source: whatscool

#### Source: whatscooking.fns.usda.gov

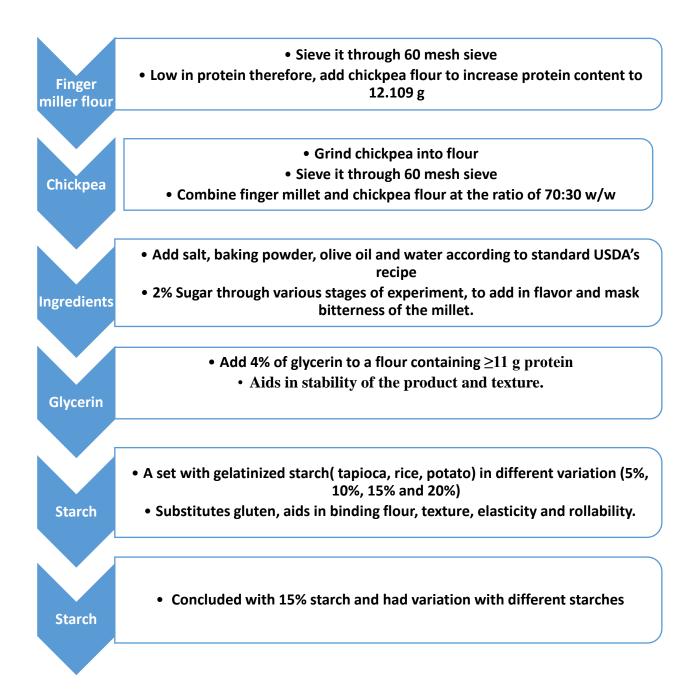
#### Table 4: USDA's standardized tortilla recipe

Finger millet has a distinct characteristic earthy taste which might not be acceptable to the general population. To boost the flavor and mask the bitterness of the millet we added sugar to the blend. According to Fernholz (2008), in a similar study of tortillas with sorghum flour he added 5% of sugar to mask the bitterness and enhance flavor. We experimented with 5% of sugar (based on 100 g flour) and found the tortillas to be very sweet, so we experimented with 1%, 2% 3% and 4 % of sugar and found 2% worked the best. Sugar at 2% enhanced the flavor and masked the bitterness of the tortilla without making it too sweet.

According to a study by Pourfarzad, et.al. (2011), propylene glycol has a great effect on quality and shelf life of bread. Addition of propylene glycol reduces the quantity of water required to get a suitable dough; significantly decreasing the moisture content making products stable. It also delays the staling in storage time longer than 2 days and soften the product by stabilizing water-starch system incorporating in the structure of water surrounding the starch chain (Pourfarzad, et.al 2011). It has been reported that addition of 4% of polyols to a flour containing  $\geq 11.0\%$  protein produced acceptable tortillas, stable during storage and rollable

(Suhendro, Waniska, Rooney & Gomez, 1995). Based on Suhendro et al., 1995 study for this research we added 4% propylene glycol to lower moisture content, make tortillas more stable by extending their shelf life.

Finger millet is a cereal grain that does not contain gluten which is beneficial as an alternative to patients with celiac disease but absence of gluten inhibits physiological property of dough like binding of the dough, rollability, and elasticity. To overcome this problem, we experimented with different types of starch (rice, tapioca, potato) and xanthan gum in different percentages to get an optimum result. We experimented with 5%, 10% 15% and 20% of starch to determine optimum quantity of starch that would bind the tortilla keeping in mind of the use least possible amount of starch. There were physical differences in tortillas with 5%, 10% and 15% starches. The description of the formulation and optimization of the tortillas is shown in the Figure 6.



#### Figure 6. Process of formulation and optimization of the tortillas.



5% Starch



10% starch



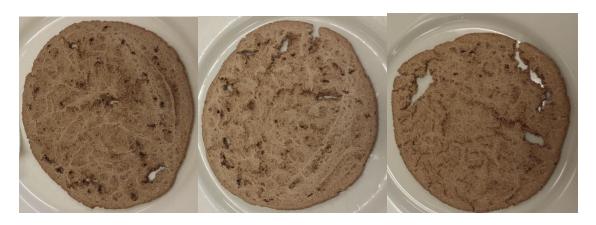
15% starch

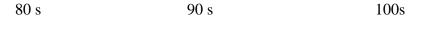


#### Figure 7: Comparision of tortillas with different % of starches.

The objective observation showed that the tortillas with 5% and 10% starch were not rollable, they cracked whenever they were rolled whereas the tortillas with 15% and 20% starch did not crack when rolled. There was no significant difference in objective observation of tortillas with 15% and 20% starch, therefore we selected 15% of starches to keep the quantity of starch low and to help in binding of the dough.

In the formulation we used extra virgin olive oil instead of regular oil to add in more flavor. The tortilla preparation process was based from a research based on sorghum flour tortilla. There was no research that backed up on cooking time of the chickpea fortified finger millet tortillas so, we experimented with cooking time and conducted an objective observation to determine cooking time. We pressed and cooked tortillas on a Cuisinart CPP-200 International chef tortilla maker at 204° C for 80, 90 and 100 s simultaneously.





#### Figure 8: Comparision of tortillas cooked at different times.

The tortillas cooked for 80 s were soft, doughy and did not look cooked where as the tortillas cooked for 100 s were hard and brittle. The tortillas cooked for 90 s were still doughy but when cooked longer were getting hard so we took 90 s as cooking time for the tortillas.

#### **Characteristics of Finger Millet Flour and Chickpea Flour**

#### Moisture

The moisture contents of the flours were determined using the AACC Approved Method 44-15-02 where 5 grams of flour were placed in an oven maintaining a temperature of 103°C for 60 min.

#### Ash

The ash content was analyzed using the AACC Approved Method 08- 01; the samples from moisture analysis were taken forward for ash by further heated at 590°C for 21 hours and is cooled for 1 hour in desiccator and weighed.

#### Protein

The crude protein of the flours was determined at Oklahoma State University, Robert M Kerr Food and Agricultural Products Center, Stillwater using AOCS approved M4 – Determination of Crude protein by Leco combustion Method, Revision 8. Protein of rest of the ingredient was obtained from the manufacturing sources.

#### **Tortilla Formulation**

The formulation of the tortilla is shown in the Table 5. The ingredients used were as follows: finger millet flour (Swad, Skokie, IL) and Chickpea (Swad, Skokie, IL) was bought from an ethnic Indian store in Portland, OK. Baking powder (Clabber Girl, Terre Haute, IN), iodized salt (Morton, Chicago, IL), sugar (Great value, Walmart store Inc.), olive oil (Great value, Walmart store, Inc.), glycerin (Plant Guru, Plainfield, NJ), starches (Ingredion,

Ingradianta 2.5	Amount in us	Amount in	Ontimization	Dariyad from
Ingredients 3.5	metrics	chickpeas (g)	Optimization	Derived from
			70:30 ratios	
Flour	2 cups 206.0	206.0	finger millet/	Standardized
Flour		206.0	chickpea flour,	recipe
			w/w	
Baking	1 top	3.5	1.69%	>>
powder	1 tsp.	5.5	1.09%	
Salt	<sup>1</sup> ∕₂ tsp.	2.8	1.37%	>>
Oil	2 tbsp.	16.2	7.84%	>>
Sugar		4.1	2% of the flour	Experiment
Glycerin		8.2	4% of the flour	Experiment
Storah		20.0	15% of the	Even a view avet
Starch	Starch 30.9	flour	Experiment	
		170 7	62.83% of the	Standardized
Water		170.7	entire mixture	recipe

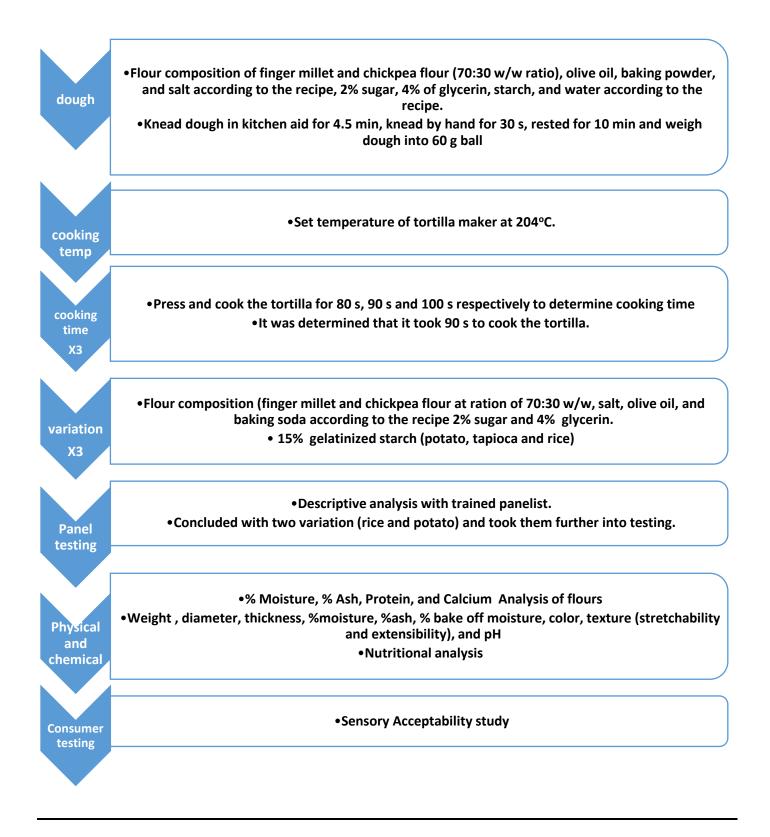
Englewood, CO), Xanthan gum (Bod's red mill, Milwaukie, OR), and water.

#### Table 5: Formulation of chickpea fortified finger millet tortillas

#### **Tortilla preparation**

The dry ingredients (finger millet flour, chickpea flour, starch, baking powder, sugar, and salt) were mixed for 1 minute and 30 s on speed 1 in a KitchenAid mixer. Olive oil and glycerin were added and mixed for 45 s at speed 1. The sides were scraped down with a spatula and the ingredients were mixed further for another 45 s at speed 2 until no clumps are visible. Warm water (38°C) was slowly added while mixing at speed 1 and increasing to speed 3 for a total mixing time of 1 minute and 30 s. The dough was kneaded for 30 s with a hook in the mixer, rested for 10 min, and then placed in a plastic container with a lid to retain moisture. The dough was carefully weighed into 60 g balls and stored in a plastic container with a lid during preparation.

Cuisinart CPP-200 International chef tortilla maker was used to press and bake tortillas at 204° C. The dough balls were placed in between the plates and pressed for 6 s and cooked in for 1 min and 30 s. The tortilla was then cooled on a cooling rack for 2 minutes and stored in a resealable plastic bag. The tortillas were rested for 2 hours before analyzed. The process of preparation of the samples and the methods is described in Figure 9 below.



#### Figure 9: Flow chart of processing and analysis

#### **Physical and Chemical Measurements**

#### Weight

The tortillas were weighed on analytical balance Tree HRB203 one at a time. The values were recorded to the nearest 100<sup>th</sup> decimal.

#### Diameter

The diameter of the tortillas was measured using a 6-inch LCD digital caliper. Two values were taken from each tortilla; the first value was recorded and the caliper was turned at 90 degrees to get second value.

#### Thickness

A 6-inch LCD digital caliper was used to measure the thickness of the tortilla. The tortillas were measured one at a time and the values was recorded in nearest millimeters to 100<sup>th</sup> decimal.

#### **Moisture Content**

The moisture content of each tortilla sample was obtained using the AACC method 44-

15.02. The weight of the samples was changed to 5g instead of 2g to increase accuracy.

#### **Bake off Moisture Percent**

The moisture loss during baking was calculated using method described in (Hathorn, Biswas, Gichuhi, & Bovell-Benjamin, 2008). The following formula was used to calculate bake off moisture %.

% weight loss =  $\frac{\text{Weight of dough} - \text{Weight of baked bread}}{\text{Weight of dough}} \times 100.$ 

#### Color

A Hunter Lab MiniScan XE Plus (Hunter Associates Laboratory Inc., Reston, VA) was used to measure the color of the tortilla samples. L, a and b values were given as an output. "L" was the measurement for lightness (0 = black and 100 = white); "a" value that indicated red and green colors (+a = red and –a = green), and "b" value indicated yellow (+b) and blue (-b) colors. The device was calibrated at the beginning of the test using a white and black tiles included in the package. The measurements were conducted at FAPC, OSU, Stillwater **Figure** 



### Stretchability

A TA-XT2i Texture Analyzer (Texture Technologies Corp., Scarsdale, NY) was used to analyze stretchability and flexibility. The 60mmTA-108 Tortilla Fixture (Texture Technologies Corp., Scarsdale, NY) and a 20mm TA-108 acrylic rounded edge probe (Texture Technologies Corp., Scarsdale, NY) was used. The probe was set with 20.0 g force and moved at a test speed of 1.70mm/s; the probe was programmed to travel 30.0mm total before returning to its original position. The tortilla was inserted through the four screws, placed in the

Figure 10: Hunter Lab MiniScan XE Plus



Figure 11: TA-XT2i Texture Analyzer

fixture, and screwed in using the bolts. As the test ran, the probe pushed through the center of the tortilla; maximum peak force values and distance values were recorded.

#### Extensibility

Extensibility of each tortilla was tested using a TA.XT2i Texture Analyzer and the TA-96 miniature tensile grips (Texture Technologies Corp., Scarsdale, NY) were used with 5.0g force, moved at a test speed of 1.00mm/s and traveled distance of 25.00m. For each sample, stainless steel dog bone template (Figure 12) was used to cut tortilla pieces with an average tortilla height of 3.44mm. Each of those pieces were placed in the tensile grips, tightened by hand and the tests were run. The tortilla pieces were pulled up vertically and the maximum peak force values and distance values were recorded. **Figur** 

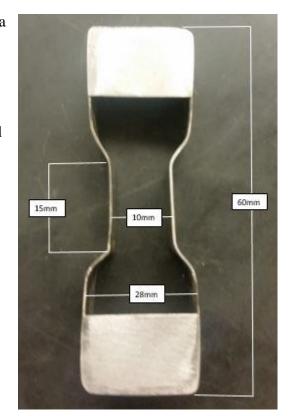


Figure 12: Stainless Steel Dog Bone Template

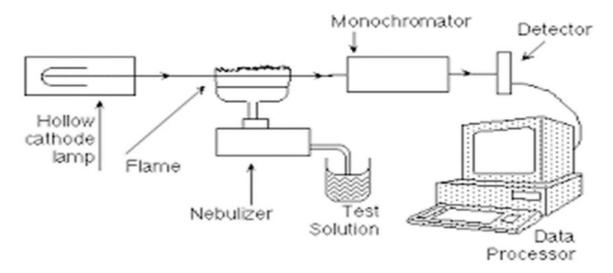
#### pН

pH was determined by homogenizing 10 g of samples in 100 ml of distilled water. The pH value was determined using a pHmeter pH-009 (I) pen type that had been calibrated against standard buffers 7 and 4.

#### **Calcium Analysis**

Calcium analysis was done at University of Central Oklahoma according to method used in (Bazzi, Kreuz, & Fischer, 2004), where Flame Atomic Absorption Spectrometry (FAAS) method measured concentration by an absorption or emission characteristics. Some of the advantages of using Atomic Absorption Spectrometry are it has greater sensitivity, and detection limits in comparison to other methods, it conducts direct analysis of some liquid samples, it has low spectral interference, and it require very small sample size (García & Báez, 2012)

According to Garcia & Baez (2012), in FAAS method sample are placed near a machine and a tube connected to the machine is inserted into the sample. When the test is initiated the tube suctions the sample up towards a nebulizer, which breaks the liquid into a fine mist. The glass bead in the nebulizer further breaks the mist into very fine liquid particles called aerosol. This aerosol is then passed through the flame to a monochromator, to a detector followed by an amplifier. The hollow cathode lamp placed before the flame acts a specific analyst that provides energy to an atom to leave the ground state.



Source: https://web.nmsu.edu/~kburke/Instrumentation/AAS1.html

#### Figure 13: Flame Atomic Absorption Spectrometry Diagram

Calcium content was analyzed with slight modification to FAAS method (Bazzi et al., 2004), approximately 5 grams (triplicate) of finger millet and chickpea flour were weighed, placed in a crucible and then in a muffle furnace at 575°C for 24 hours. The crucibles were then allowed to cool for an hour in a desiccator and once cooled they were weighed. After the

crucibles were weighed the sample residues were transferred into a beaker and 5-ml of 12 M HCL was used to dissolve it. This solution was then transferred into a 100-ml volumetric flask and diluted with deionized water to the mark and mixed thoroughly. The solution was then filtered using a silica funnel to remove remaining matrix. Each sample, a 5-mL aliquot was pipetted into 50-mL volumetric flask, along with 5-mL of La Matrix Modifier solution, diluted with deionized water to the mark and mixed thoroughly. A Standard Addition solutions was made with a 1-mL aliquot pipetted into a 50-mL volumetric flask, along with 5-mL of La Matrix Modifier solution and 1-5 milliliters of standard CaCl<sub>2</sub> standard solution, which was diluted with deionized water to the mark and mixed thoroughly. Once all the Standard Solutions were made, the absorbance was then measured using the Flame Atomic Absorption Spectrometry Machine. The absorbance's were recorded, and entered into an Excel sheet, where all calculations and analyses were done.

#### **Nutritional Analysis**

Nutritional analysis of the tortillas was performed using Genesis R&D Software (ESHA Research, Salem, OR; Version 9.12.1.0) at the University of Central Oklahoma.

#### **Sensory Analysis**

Sensory evaluation of the tortillas was conducted at University of Central Oklahoma in two different evaluations. A descriptive analysis of 3 sample tortillas was done by trained panel (dietetic interns and graduate students), the result of the analysis lead to elimination of one sample. The remaining two samples were taken further into additional testing and sensory acceptance study. Sensory acceptance study was conducted by students and staff. Institutional Review Board's (IRB) approval was granted for all stages of this study through University of Central Oklahoma.

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#### **Descriptive Analysis**

Descriptive analysis was conducted at University of Central Oklahoma (UCO). To familiarize the panelist with the sensory analysis a modified Spectrum<sup>TM</sup> method (Meilgaard, Carr, & Civille, 2006) was used and 15 point scale was used to score the taste testing. **Fi** 



Figure 14: Setup for descriptive sensory analysis

The panelist in the descriptive analysis were asked to fill in a brief questionnaire on demographics, education and consumption of tortillas. The study was conducted with 8 trained panelists (7 females ,1 male) age ranging from 18-40 years (62.5% at the age group of 18-25years), 62.5% of the panelist had completed Bachelors and rest of them had completed Masters degrees. As per consumption of tortillas, 37.5% of the panelist claimed to eat tortillas at least once a week., 37.5% once every two weeks and 25% once a month. The panelists gave informed consent to participate in the sensory testing and did not have any history of food allergy.

The panelists were trained in two sessions; in the first sessions sensory panelist were trained to define the attributes and references on taste, texture, odor, and appearance. A list of attributes was referred from a similar study on sorghum flour which accounted for all the characteristics of the finger millet flour and a 15-point numbered absolute scale was used to score perceived intensity. In the second session the attributes and references were analyzed and refined with standard compounds. The final session, sampling the tortillas were conducted the next day to eliminate panelist fatigue and were provided with three samples of tortillas on a white paper plate with random three- digit numeric codes assigned on each sample along with

scoring ballots. In the session the tortilla samples were evaluated following standard taste compounds: 02.%, 0.4% and 0.8% of NaCl solution with scores of 4, 7 and 15 respectively for saltiness; 1%, 2%, 4% and 8% sucrose solution with scores of 3, 6, 11 and 15 respectively for sweetness; 0.02%, 0.04% and 0.06% of caffeine with the score of 2, 7 and 14 respectively for bitterness; butter roll and canned biscuit dough with scores of 5 and 15 respectively for doughiness and toasted wheat germs with a score of 10 for nuttiness. For evaluation of texture by mouth following standard texture compounds were used: cream cheese, sharp cheddar cheese and peanuts with scores of 1, 4 and 13 respectively for hardness; corn muffin and graham crackers with scores of 2 and 7 respectively for fracturability; post grape nuts with a score of 14 for grittiness. To evaluate texture by hand following standard texture compounds were used: Kool-aid gels, orange peel, potato chips and crunchy granola bars with scores of 1,6,10 and 14 respectively for roughness; flour tortilla and pita pocket bread with scores of 8 and 14 respectively for tearability. For odor granulated sugar, honey maid graham crackers and clover honey with scores of 2, 8 and 14 respectively for sweet and a bag of millet flour with a score of 11 for musty odor were used as standard odor compounds. The evaluation of the appearance was done following standard compounds: Flour tortilla and whole wheat flour tortilla with a score of 2 and 13 respectively for evenness of the color; wheat flour tortilla and regular flour tortilla with score of 5 and 11 respectively for variation in fluctuation on surface of the tortilla. The sensory sessions were conducted at 22-24°C, the panelists were provided with unsalted crackers and distilled water to cleanse their palate after each sample in separated booths for all the sessions.

#### Sensory acceptability study

The sensory acceptability study was held at University of Central Oklahoma Fifty (50) untrained volunteering panelists were asked to answer some survey questions on demographics,

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education and consumption of tortillas. They were also asked to complete a consent and allergy release form before the taste testing. If the participants had allergies they were not be allowed to participate in the taste testing. Tapioca starch tortillas were eliminated from this study due to the consensus result of descriptive panel as the most unacceptable tortilla samples. Only rice starch and potato starch tortillas were used in this study.

The volunteer panelist consisted of 50 untrained accessors consisting in 35 females and 15 males, age ranging from 18-80 years (76% of the panelist being in the 18-25 age group, 14% in 26-30 age group, 2% in 31-35 age group, 4% in 36-40 age group, 2% in 51-55 age group and 2% in 71-80 age group). Among 50 accessors, 12% had complete high school, 76% had completed some college courses, 6% had completed Bachelors, 4% had Masters and 2% had complete PhD. As per consumption of tortilla, 2% of the panelist claimed to eat tortillas every day, 36% ate at least once a week, 32% ate once in 2 weeks, 26% ate once a month, 2% once a year and 2% never ate.

The sample tortillas were placed on a white paper with three- digit numeric code assigned to each. Samples were given to the panelist one at a time to eliminate bias; unsalted cracker and distilled water was provided to cleanse their palate between tastings. A 9-point hedonic scale ballot was provided to score each sample. The 9-point hedonic scale displayed degrees of like and dislike (1, extremely dislike; 9 extremely like) and was given section under every attribute for additional comments. The attributes tested were for appearance, aroma, texture, tenderness, taste, and overall likability. The study was conducted at 22-24°C temperature room in separated booths.

#### **Statistical Analysis**

All the analyses were conducted in triplicates. Means and standard deviations of all samples were reported for color, moisture, nutrition, and sensory evaluations. One-way ANOVA was performed using General Linear Model Procedure to identify significant difference (p < 0.05) among the samples followed by Tukey's test. All Statistical analyses were carried out using SPSS (SPSS 20.0, IBM Crop, Armonk, NY) and Microsoft Excel 2016 MSO, Version 16.0.6001.1078.

#### **CHAPTER FOUR: RESULTS AND DISCUSSIONS**

#### **Physical and Chemical Measurements**

#### Moisture, Ash and Protein Content of Finger Millet Flour and Chickpea Flour

Table 6 shows the average moisture, ash and protein content of finger millet flour, chickpea flour, potato starch, and rice starch. The results agree with those of Sertac et al, (2010) who reported chickpea containing 17.55-23.32% of protein, 2.54-3.41% of ash and 6.39-10.57% of moisture. Also results also agree with previous reports of finger millet showing approximately 7% protein, 1.7-4.13% ash and 13.2% of moisture (Singh & Raghuvanshi, 2012; Shobana et al., 2013). The slight variation comparing to literature reports could be explained in part by different varieties of the grains used. There was no significant difference in moisture content of the flours but there were significant differences in ash and protein content. The ash of chickpea flour was higher that finger millet flour which shows there is higher quantity of minerals in chickpea flour in comparison to finger millet flour. The data for potato starch and rice starch was obtained from their manufacturer.

Table 6: Comparison of moisture, ash and protein content results of finger millet flour and
chickpea flour *

Sample	Moisture (%) Ash (%)		Protein (%)
Finger millet flour	$10.7 \pm 0.09^{a}$	$1.83\pm0.13^{a}$	5.2
Chickpea flour	$10.3\pm0.11^{\rm a}$	$2.41\pm0.31^{b}$	19.7
Potato Starch	4%	0.21%	< 0.1%
<b>Rice Starch</b>	12%	0.24%	0.43%

\*Means  $\pm$  standard deviation with different superscripts within columns indicate significant differences among treatments (p<0.05).

#### Weight, Diameter, Thickness and Bake Off Moisture Percentage

The Table 7 shows the averages for weight, diameter, and thickness of the two finger millet flour samples. There was significant difference in weight and diameter but no significant differences in thickness of the tortillas. The weight of the tortillas is indirectly proportional to the bake off moisture % of the tortillas. The bake off moisture % of T-RICE tortilla made with rice is higher than T-POTATO tortilla made with potato which shows that more moisture (liquid) is baked off in the process of making the tortilla in comparison to T-POTATO. The moisture bake off from the tortilla is also a characteristic of starch, it shows that potato starch absorbs and holds more water than rice starch. The potato starch has larger, irregular granules and higher content of phosphate group in comparison to rice starch which aids in higher swelling power without disintegration (Kaur, Singh & Sodhi, 2002; Galliard, 1987).

The diameter and the thickness of the tortillas are indirectly proportional; higher the diameter co-relates to lower thickness due to the spreadability of the tortillas. T-RICE made with rice had higher spreabability with higher diameter and lower thickness which is supported by the study that states rice starch has unique spreadable characteristics which is valuable in food as well as pharmaceutical application (Wani et al., 2012).

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 Table7: Comparison of weight, thickness and diameter results of chickpea fortified finger

 millet tortillas with different starches\*.

	Weight (g)	Thickness (mm)	Diameter (mm)	Bake off moisture (%)
T-RICE <sup>1</sup>	$46.50\pm0.39^a$	$3.28\pm0.36^{\text{a}}$	$144.79\pm2.19^a$	33.5
T-POTATO <sup>2</sup>	$49.25\pm0.94^{b}$	$3.61\pm0.09^{a}$	$133.51\pm3.12^{b}$	28.0

\*Means  $\pm$  standard deviation with different superscripts within columns indicate significant differences among treatments (p<0.05).

1 Chickpea fortified finger millet tortilla with rice starch.

2 Chickpea fortified finger millet tortilla with potato starch.

#### Moisture Content, Ash and pH

Table 8 shows the average moisture content, ash and pH of two chickpea fortified finger millet tortilla sample. There was no significant difference in moisture content, ash or pH of the sample tortilla. These results were expected as two samples differ only with respect to starch. The composition of starches is very similar consisting of polymers and minor compound however the physio-chemical properties and functional characteristics were prepared in an aqueous system and annealing could occur during heating (Singh et al., 2003; Wani et al., 2012).

#### Table 8: Comparison of moisture and ash results of chickpea fortified finger millet tortillas

with different starches\*

Sample	Moisture %	Ash %	рН
T-RICE <sup>1</sup>	$26.4\pm0.09^{a}$	$2.86\pm0.07^a$	$6.48\pm0.08^{a}$
T-POTATO <sup>2</sup>	$26.6\pm0.04^{\rm a}$	$2.86\pm0.03^{a}$	$6.48\pm0.02^{\rm a}$

\*Means  $\pm$  standard deviation with different superscripts within columns indicate significant differences among treatments (p<0.05).

1 Chickpea fortified finger millet tortilla with rice starch.

2 Chickpea fortified finger millet tortilla with potato starch.

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#### Color

Table 9 shows the average 'L', 'a', and 'b' values which were significantly different in both the samples. The tortillas T-POTATO made with potato starch were lighter in color in comparison to T-RICE made with rice starch with a 'L' value of 57.1. The values of 'a' were higher in T-RICE which indicates that it has more redness and as for values of 'b' T-POTATO is higher which shows it has more yellow color. According to Singh et al. (2003), the higher phosphate monoester content in potato starch results in pastes with higher light transmittance whereas higher phospholipids in cereal starch (rice) results in pastes with lower transmittance. The transmittance properties of the starches explain the lighter color of tortillas with potato starch in comparison to tortillas with rice starch. According to Yang, Hattori, Kawaguchi & Takahashi, (1998) maillard reaction occurred between potato starch and lysine resulting in higher yellowness, which explains the yellower color of tortillas with potato starch in comparison to tortillas with rice starch.

# Table 9: Comparison of color results of chickpea fortified finger millet tortillas with different starches\*

Sample	L	a	b
T-RICE <sup>1</sup>	$52.56\pm0.99^{a}$	$8.32 \pm 0.15^{a}$	$16.64\pm0.33^a$
T-POTATO <sup>2</sup>	$57.09 \pm 1.41^{\text{b}}$	$7.82\pm0.39^{b}$	$17.62\pm0.83^{\text{b}}$

\*Means  $\pm$  standard deviation with different superscripts within columns indicate significant differences among treatments (p<0.05).

1 Chickpea fortified finger millet tortilla with rice starch.

2 Chickpea fortified finger millet tortilla with potato starch.

#### **Texture (Stretchability and Extensibility)**

T-RICE tortillas made with rice starch had significant higher force meaning they were

firmer but had insignificant difference in distance with only a slightly higher distance indicating

only slight extensible. According to Frenholz (2008), a higher force indicates greater stretchability, the higher force on T-RICE suggests that it has the higher stretchability in comparison to T-POTATO tortillas with potato starch. However, Frenholz (2008) also states that gluten-network in wheat tortillas creates flexibility so, stretchability test may not be a good indicator for a gluten-free tortilla due to the absence of gluten-network.

The table 10 shows the average force and distance of the tortillas testing its extensibility. There was significant different for force and distance with the lowest value of 1184.93g. According to Suhendro et al., 1999, a low force value and longer distance of extension indicates soft and extensible tortillas whereas higher force value and shorter rupture distance indicates hard and brittle tortillas. T-RICE made with rice has low force and longer distance whereas T-POTATO made with potato has higher force and shorter distance making indicating T-RICE being softer than T-POTATO.

 Table 10: Comparison of extensibility and strechability texture results of chickpea fortified

 finger millet tortillas with different starches\*

	Extensibil	Extensibility		ability
Sample	Distance Force (g) (mm)		Force (g)	Distance (mm)
T-RICE <sup>1</sup>	$1184.93 \pm 125.288^{a}$	$17.35 \pm 1.81^{a}$	$424.6\pm38.31^{\text{a}}$	$3.92\pm0.75^{\text{a}}$
T-POTATO <sup>2</sup>	$1427.77 \pm 245.65^{b}$	$14.63\pm0.59^{b}$	$372.14 \pm 53.77^{b}$	$3.82 \pm 1.09^{a}$

\*Means  $\pm$  standard deviation with different superscripts within columns indicate significant differences among treatments (p<0.05).

1 Chickpea fortified finger millet tortilla with rice starch

2 Chickpea fortified finger millet tortilla with potato starch

#### **Calcium Analysis**

The calcium analysis using Flame Atomic Absorption (FAA) showed that finger millet has 43.553mg of calcium per 100g and chickpea has 14.167mg per 100g. The plots for calcium absorbance in finger millet and chickpea is shown below and the calculation are in Appendix 1.

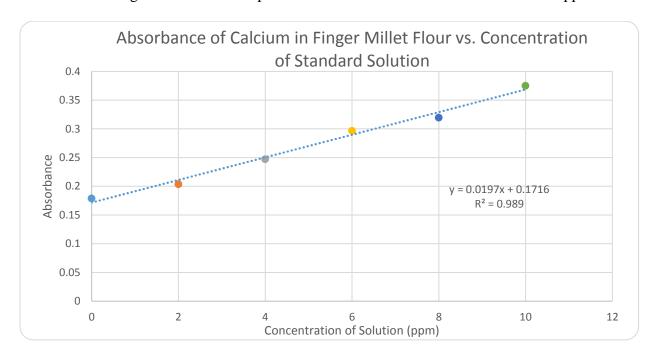
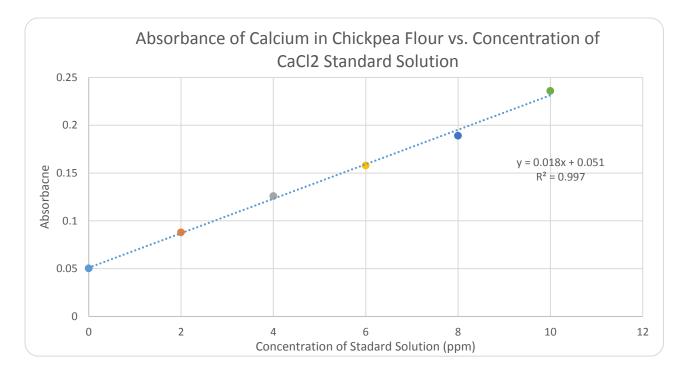


Figure 15: Calcium absorbance of the finger millet flour vs the concentration of the

calcium standard solution (ppm)



## Figure 16: Calcium Absorbance of Chickpea Flour vs. the Concentration of the Calcium Standard Solution (ppm)

#### Nutrition Label

There was no significant difference in nutritional facts of the tortillas since the only difference in the formulation was the use of different starch which had similar properties. The composition of starches is very similar consisting of polymers and minor compound however the physio-chemical properties and functional characteristics is subjected to aqueous system, biological origin and annealing (Singh et al., 2003; Wani et al., 2012).

Nutrition Facts Serving Size (28g) Servings Per Container	Nutrition Facts Serving Size (28g) Servings Per Container
Amount Per Serving	Amount Per Serving
Calories 70 Calories from Fat 15	Calories 70 Calories from Fat 15
% Daily Value*	% Daily Value
Total Fat 1.5g 2%	Total Fat 1.5g 2%
Saturated Fat 0g 0%	Saturated Fat 0g 0%
Trans Fat 0g	Trans Fat 0g
Cholesterol 0mg 0%	Cholesterol 0mg 0%
Sodium 100mg 4%	Sodium 100mg 4%
Total Carbohydrate 12g 4%	Total Carbohydrate 12g 4%
Dietary Fiber 1g 4%	Dietary Fiber 1g 4%
Sugars 1g	Sugars 1g
Protein 2g	Protein 2g
Vitamin A 0% • Vitamin C 0%	Vitamin A 0% • Vitamin C 0%
Calcium 4%  • Iron 4%	Calcium 4%  • Iron 4%
*Percent Daily Values are based on a 2,000 calorie diet. Your daily values may be higher or lower depending on your calorie needs: Calories: 2,000 2,500	*Percent Daily Values are based on a 2,000 calorie diet. Your daily values may be higher or lower depending on your calorie needs: Calories: 2,000 2,500
Total Fat         Less than         65g         80g           Saturated Fat         Less than         20g         25g           Cholesterol         Less than         300mg         300mg           Sodium         Less than         2,400mg         2,400mg           Total Carbohydrate         300g         375g           Dietary Fiber         25g         30g	Total Fat         Less than         65g         80g           Saturated Fat         Less than         20g         25g           Cholesterol         Less than         300mg         300mg           Sodium         Less than         2,400mg         2,400mg           Total Carbohydrate         300g         375g           Dietary Fiber         25g         30g
Calories per gram: Fat 9 • Carbohydrate 4 • Protein 4	Calories per gram: Fat 9 • Carbohydrate 4 • Protein 4

**Tortilla with Rice Starch** 

**Tortilla with Potato Starch** 

#### Figure 17: Nutritional facts comparision of tortillas made rice and potato starch

#### **Sensory Analysis**

#### **Descriptive Analysis**

For flavor, the only significant difference was in sweetness and doughy after taste of the tortillas. The tortillas with potato starch was the sweetest compared to the tortilla with tapioca starch which has an average score of 1.4 (least sweet comparable to 0.465 or sucrose solution). The doughy profile was high (5.5 score comparable to butter roll) for tortillas with potato starch while the scores were similar for tortillas with rice or tapioca starch. Overall the highest acceptance scores were observed on tortillas with potato starch compared to those with rice or tapioca starch.

In attributes of texture, there was no significant difference across the parameters both in hand and mouth feel texture. But the scores for roughness and tearbility was slightly higher for

tortilla with potato starch which correlates with the physicochemical texture data that indicated it is harder in comparison to tortilla with rice starch.

Shape is the only attribute that has significant difference in context of appearance of the tortillas. The tortillas with rice starch were rounder than other tortillas with a high score of 14.1. The data indicates that tortillas with rice starch and tortillas with tapioca starch were rounder and smoother than tortilla with potato starch.

In aspect of odor and overall likability there was no significant difference in the tortillas, but the scores indicated that tortilla with tapioca starch, tortilla with rice starch and tortilla with potato starch had least sweet and musty odor respectively. The panelist prefered tortilla with potato starch with an overall likability scores of 11.1 and disliked tortilla with tapioca starch with the least score of 6.9. Due to low score tortilla with tapioca was eliminated from the experiment and tortilla with rice and potato starch were taken further for testing and sensory acceptability study.

## Table 11. Comparison of flavor attributes in description analysis of chickpea fortified

#### finger millet tortillas with different starches\*

Sample	Sweet <sup>1</sup>	Salty <sup>II</sup>	Nutty <sup>III</sup>	Bitter <sup>IV</sup>	Doughy V
T-RICE <sup>1</sup>	$1.6\pm0.74^{a}$	$2.3\pm0.71^a$	$6.5\pm2.62^{a}$	$1.3\pm0.71^{a}$	$2.9\pm.099^{a}$
T-POTATO <sup>2</sup>	$2.6\pm0.92^{b}$	$2.6\pm1.51^{a}$	$6.1\pm3.18^{a}$	1.0 ±0.00 <sup>a</sup>	$5.5 \pm 2.56^{b}$
T-TAPIOCA <sup>3</sup>	$1.4\pm0.52^{\rm a}$	$1.8\pm0.89^{a}$	$5.6\pm2.20^{\mathrm{a}}$	$2.0 \pm 1.77^{a}$	$3.0 \pm 1.69^{a}$

Flavor

\*Means  $\pm$  standard deviation with different superscripts within columns indicate significant differences among treatments (p<0.05).

1. Chickpea fortified finger millet tortilla with rice starch.

2. Chickpea fortified finger millet tortilla with potato starch.

3. Chickpea fortified finger millet tortilla with tapioca starch.

I. Sweet intensity was evaluated on a scale from 1 (not detectable) to 15 (extremely sweet)

II. Salty intensity was evaluated on a scale from 1 (not detectable) to 15 (extremely salty)

III. Nutty intensity was evaluated on a scale from 1 (not detectable) to 15 (extremely nutty)

IV. Bitter intensity was evaluated on a scale from 1 (not detectable) to 15 (extremely bitter)

V. Doughy intensity was evaluated on a scale from 1 (not detectable) to 15 (extremely doughy)

## Table 12. Comparison of texture attributes in description analysis of chickpea fortified

#### finger millet tortillas with different starches\*

#### Texture

Sample	Texture (in hand)		Texture (by mouth)		
Sample	Roughness <sup>I</sup>	Tearability <sup>II</sup>	Hardness III	Fracturability <sup>IV</sup>	Grittiness V
T-RICE <sup>1</sup>	$5.5\pm1.07^{\rm a}$	$12.3\pm2.38^{a}$	$8.6\pm3.96^{a}$	$7.3\pm3.20^{a}$	$5.6\pm3.99^{a}$
T-POTATO <sup>2</sup>	$6.1 \pm 2.90^{a}$	$12.5\pm2.14^{\rm a}$	$5.8 \pm 1.66^{\rm a}$	$5.9\pm2.85^{a}$	$3.1 \pm 1.45^{a}$
T-TAPIOCA <sup>3</sup>	$5.4 \pm 1.19^{\rm a}$	$12.4\pm1.77^{a}$	$7.9\pm3.39^{\rm a}$	$6.4 \pm 2.19^{a}$	$5.0\pm3.07^{\rm a}$

\*Means  $\pm$  standard deviation with different superscripts within columns indicate significant differences among treatments (p<0.05).

1. Chickpea fortified finger millet tortilla with rice starch.

2. Chickpea fortified finger millet tortilla with potato starch.

3. Chickpea fortified finger millet tortilla with tapioca starch.

I. Roughness intensity was evaluated on a scale from 1 (not detectable) to 15 (extremely rough)

II. Terability intensity was evaluated on a scale from 1 (easily pulled apart) to 15 (extremely hard to pull apart)

III. Hardness intensity was evaluated on a scale from 1 (extremely easy to bite down) to 15 (extremely hard to bite down)

IV. Fracturability intensity was evaluated on a scale from 1 (extremely easy break) to 15 (extremely hard to break)

V. Grittiness intensity was evaluated on a scale from 1 (absence of gritty particles) to 15 (extremely presence of gritty particles)

# Table 13. Comparison of appearance attributes in description analysis of chickpea fortified finger millet tortillas with different starches\*

Sample	Evenness of the color <sup>I</sup>	Shape <sup>II</sup>	Surface <sup>III</sup>
T-RICE <sup>1</sup>	$9.8\pm3.73^a$	$14.1\pm0.35^a$	$6.0\pm2.97^{\rm a}$
T-POTATO <sup>2</sup>	$8.3\pm3.77^{\rm a}$	$8.6\pm3.02~^{b}$	$6.5\pm3.33^{a}$
T-TAPIOCA <sup>3</sup>	$11.3 \pm 2.12^{\mathrm{a}}$	$12.5 \pm 2.5$ <sup>a</sup>	$5.6\pm2.06^{\rm a}$

### Appearance

\*Means  $\pm$  standard deviation with different superscripts within columns indicate significant differences among treatments (p<0.05).

1. Chickpea fortified finger millet tortilla with rice starch.

2. Chickpea fortified finger millet tortilla with potato starch.

3. Chickpea fortified finger millet tortilla with tapioca starch.

I. Evenness of the color intensity was evaluated on a scale from 1 (very even) to 15 (extremely uneven)

II. Shape intensity was evaluated on a scale from 1 (not round) to 15 (perfectly round)

III. Surface intensity was evaluated on a scale from 1 (presence of blistering) to 15 (absence of blistering)

# Table 14. Comparison of odor and overall likability attributes in description analysis of

### chickpea fortified finger millet tortillas with different starches\*

Sample	Sweet <sup>I</sup>	Musty <sup>II</sup>	Overall likability <sup>III</sup>
T-RICE <sup>1</sup>	$2.0\pm1.07^{a}$	$6.8\pm2.36^{a}$	$9.3\pm2.76^a$
T-POTATO <sup>2</sup>	$2.1\pm1.46^{a}$	$5.8\pm3.69^{a}$	$11.1\pm3.72^{\rm a}$
T-TAPIOCA <sup>3</sup>	$1.8 \pm 1.49^{\rm a}$	$7.3\pm2.76^a$	$6.9\pm4.32^{\rm a}$

### **Odor and Overall likability**

\*Means  $\pm$  standard deviation with different superscripts within columns indicate significant differences among treatments (p<0.05).

1. Chickpea fortified finger millet tortilla with rice starch.

2. Chickpea fortified finger millet tortilla with potato starch.

3. Chickpea fortified finger millet tortilla with tapioca starch.

I. Sweet intensity was evaluated on a scale from 1 (not detectable) to 15 (extremely sweet)

II. Musty intensity was evaluated on a scale from 1 (not detectable) to 15 (extremely musty)

III. Overall likability intensity was evaluated on a scale from 1 (extremely dislike) to 15 (extremely like)

### Sensory acceptance study

Table 15 shows the average scores from the consumer acceptability test. Appearance is the only attribute that has significant difference with a score of 6.3 for tortilla with rice starch and 5.6 for tortilla with potato starch. The overall likability score for tortilla with potato starch were slightly higher in comparison to tortilla with rice starch which correlates with higher score in taste, aroma and texture. In contrast, the appearance and the tenderness score were low for tortilla with potato starch which correlates with it being smaller, thicker and tougher tortillas from physicochemical testing. According to Wani et al., (2012), rice starch has bland taste, smooth, creamy and spreadable characteristics which corresponds with lower scores in taste but higher scores in appearance and tenderness of the tortillas with rice starch.

## Table 15. Comparison of scores from consumer acceptance study of chickpea fortified

finger millet tortillas with different starches\*

Sample	Overall likeability	Appearance	Texture	Tenderness	Aroma	Taste
T-RICE <sup>1</sup>	$6.0 \pm 1.77^{a}$	$6.3\pm1.51^{a}$	$5.8 \pm 1.68^{a}$	$6.0\pm1.76^{a}$	$5.9\pm1.39^{a}$	$5.9 \pm 1.82^{a}$
T-POTATO <sup>2</sup>	$6.3 \pm 1.7^{a}$	$5.6 \pm 1.70^{b}$	$6.0\pm1.75^{a}$	$5.9 \pm 1.93^{\rm a}$	$6.1 \pm 1.51^{a}$	$6.0 \pm 1.87^{\mathrm{a}}$

\*Means  $\pm$  standard deviation with different superscripts within columns indicate significant differences among treatments (p<0.05).

1 Chickpea fortified finger millet tortilla with rice starch

2 Chickpea fortified finger millet tortilla with potato starch

### **CHAPTER FIVE: CONCLUSION AND FUTURE DIRECTIONS**

The prevalence of obesity and the health conditions that are strongly linked to it (diabetes, cardiovascular disease) and the growing incidences of food allergies are of major concern globally. These health conditions are not only talking toll in the health but also the economy of the people. Global organization like WHO, CDC and many more are taking measure to control it by educating people and spreading awareness. The change in lifestyle and diet are one of the few measure to reduce risk of these conditions. The availability of healthier food choices and the awareness of functional ingredients are of utmost importance. Finger millet is an ancient millet grain that has superior nutritional values and has shown to aid in many health condition. Formulation of food incorporating finger millet could provide alternative and boost heathier diet leading to better health.

This research was successful in formulating, optimizing and conducting sensory acceptance study on a nutrient dense gluten-free chickpea fortified finger millet tortilla incorporating minimal amount of starch. Physical, chemical, textural and sensory testing showed differences between tortillas with different starches. The results indicated that incorporation of potato/rice starches may result in formulation of chickpea fortified finger millet tortillas with acceptable textural and sensory properties which would be a gluten free, nutrient dense alternative to traditional tortillas for people with celiac disease and a potential medicinal food for people with diabetes. The overall acceptability was higher for tortillas with potato starch due to its flavor neglecting the textural characteristics. The textures of the tortillas with potato starch was not ideal and comparable to commercial tortillas. Further research should include hydrocolloids and emulsifiers (sodium stearoyl lactylate, DATEM, and others) to improve overall quality of the tortillas. The shelf- life of the tortillas has not been studies in this research,

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so further research is required in the field of shelf- life along with research on effect of high protein flour composition as an alternative of gluten.

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### **APPENDICES**

### Appendix 1: Percentage of calcium ion in finger millet and chickpea grain

1. Obtained mass of CaCl<sub>2</sub> to make 1000ppm Calcium stock solution

 $\blacktriangleright \frac{1000\mu gCa2+}{mL} \times \frac{1000mL}{\frac{1}{L}} \times \frac{1gCa2+}{10^{6}\mu g} \times \frac{molCa2+}{40.078gCa2+} \times \frac{1 molCaCl2}{1 molCa2+} \times \frac{110.984gCaCl2}{1 molCaCl2} = 2.7692gCaCl2 \text{ per Liter}$ 

- Obtained 1.386g of CaCl<sub>2</sub>, and diluted into 500-mL Volumetric Flask to make 1000ppm primary standard
- 2. 250-mL of 100ppm Working Standard Solution using 25mL of 1000ppm Primary Standard Solution:  $M_1V_1=M_2V_2$

• M2=
$$\frac{(25mL)\times(1000ppm)}{250mL}$$

M2 = [S]i = 100ppm

- 3. 5% (m/v) of La matrix
  - ▶ Obtained mass of La(NO<sub>3</sub>)<sub>3</sub>: 7.325g
  - Volume of 12.1M HCl used to dilute: 40mL
  - Diluted with deionized water: 250mL
- 4. For each sample, an increment of 0, 1, 2, 3, 4 and 5 mL of 100ppm standard solution (initial volume, Vo) was added, together with 1mL of aliquot, 5mL of LA matrix solution and diluted to 50mL (Final volume, V<sub>f</sub>). Therefore, the concentration of standard solution.
  - ►  $[S]_f = [S]_i \times (\frac{Vo}{Vf})$
  - Example: By added 1mL of standard solution into the sample solution and diluted to 50mL. The final concentration of standard solution was:

$$[S]_{f} = (100 \, ppm) \times (\frac{1mL}{50 \, mL})$$

 $[S]_f = 2ppm$ 

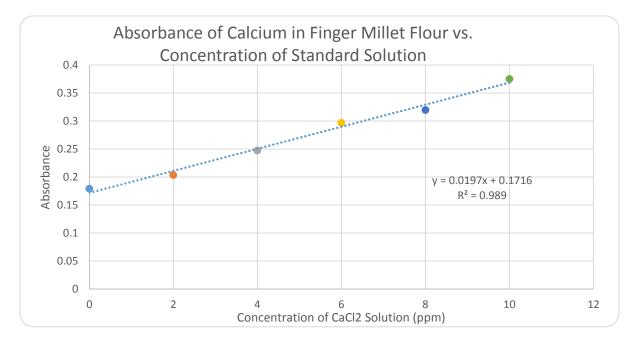
### 5. <u>% Calcium in Finger Millet Grain:</u>

Table 1: Calcium Absorbance and concentration of the Calcium standard solution

Standard	[S]f Final concentration of	Trial 1	Trial 2	Trial 3	Average
Solution (mL)	the Standard Solution (ppm)				Absrobance
0	0	0.0509	0.0494	0.0507	0.050333
1	2	0.0851	0.0877	0.0912	0.088
2	4	0.1229	0.1258	0.1293	0.126
3	6	0.1562	0.1574	0.1599	0.157833
4	8	0.1971	0.1912	0.179	0.1891
5	10	0.2458	0.2372	0.2245	0.235833

Figure 1: Calcium Absorbance of the Finger Millet Grain and the Concentration of the Calcium

### Standard Solution (ppm)



A. Final Concentration of the Finger Millet Solution

$$\blacktriangleright$$
 y= 0.0197x + 0.1716

 $\blacktriangleright \quad y=I_{s+x}*(V/V_o)$ 

► Is+x: is a measurement of analytical signal

•  $x = [S]_i^* (Vo/V_f)$ 

From equation of the line, setting y=0, the x-intercept is:

$$x = (0 - 0.1716)/0.0197$$
$$x = -8.7107$$

The magnitude of the intercept on the x-axis is the final concentration of FM solution, [X]<sub>f</sub>=

8.7107ppm, after dilution to the final sample volume.

- B. Initial Ca2+ Concentration of in 50-mL sample solution
- ▶ 1mL of FM solution (Vo) was drawn to prepare a 50mL aliquot solution (V) for FAA.

$$\blacktriangleright [X]_{f} \times V = [X]_{i} \times V_{o}$$

•  $[X]_{i=8.7107} \text{ ppm} \times (\frac{50mL}{1 mL})$ 

[X]<sub>i</sub>= 435.5330 ppm or mg/L

- C. Mass of Ca<sup>2+</sup> in 100mL FM Solution
- Mass of  $Ca^{2+} = [X]i (mg/L) \times Volume of Solution (L)$ 
  - Mass of  $Ca^{2+} = 435.5330 \frac{mg}{L} \times \frac{1L}{1000mL} \times 100mL$

Mass of  $Ca^{2+} = 43.5533 \text{ mg} = 0.04355g$ 

D. %  $Ca^{2+}$  in 0.274g of Finger Millet Grain after the furnace

 $\frac{Mass of Ca^{2+}in the solution}{Mass of FM in the solution} x100$ 

 $\% Ca^{2+} = \frac{0.04355g \ Ca^{2+}}{0.274g \ FM} x100 = 15.8954\% \sim 16\%$ 

- 6. Uncertainty in the x-intercept
- Standard deviation of x-intercept:

$$S_{\chi} = \frac{S_{\gamma}}{|m|} \sqrt{\frac{1}{n} + \frac{(y - \overline{y})^2}{m^2 \sum (x_i - \overline{x})^2}}$$

- ►  $s_y = 0.008684$
- $\blacktriangleright m = 0.1293 \qquad m^2 = 0.01672$
- ▶ n=6
- $(y \overline{y}) = 0.2701$

$$\Sigma((x_i - \overline{x})^2 = 70)$$

- ►  $s_x = 1.1739$
- The final concentration of Ca2+ in the sample with uncertainty: 8.7107 ppm  $\pm 1.1739$

# %Ca<sup>2+</sup> in Chickpea Grain:

Table 2: Average Absorbance and the	Concentration	of the Standard	Solution	(ppm)

Standard Solution	[S]f Final concentration	Trial 1	Trial 2	Trial 3	Average
(mL)	of the Standard Solution				Absorbance
	(ppm)				
0	0	0.0509	0.0494	0.0507	0.050333
1	2	0.0851	0.0877	0.0912	0.088
2	4	0.1229	0.1258	0.1293	0.126
3	6	0.1562	0.1574	0.1599	0.157833
4	8	0.1971	0.1912	0.179	0.1891
5	10	0.2458	0.2372	0.2245	0.235833

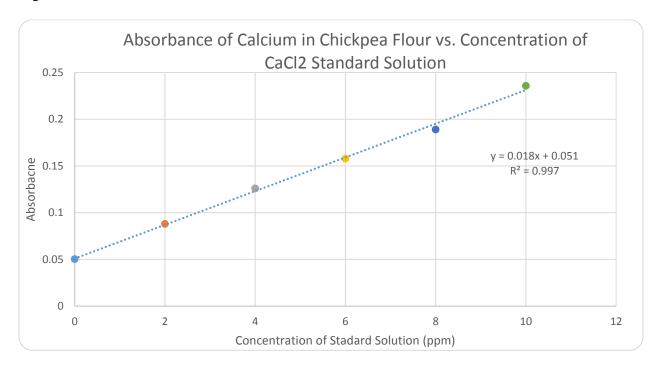
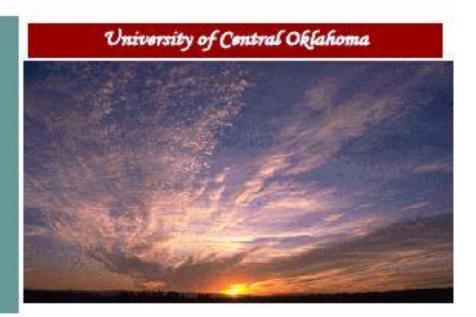


Figure 2: Absorbance of Calcium vs. the Concentration of the Standard Solution

- Mass of Calcium ion in the solution: 14.1667 mg = 0.01417 g
- Mass of the Chickpea Grain after the furnace: 0.338g
- ▶ % Ca2+ in the solution: 4.1913% ~ 4.2%

### **Appendix 2: Recruitment flyer**



# You are invited to Participate

# In a sensory evaluation of Finger millet and Legume tortilla.

Your participation in this project will involve completing a brief survey followed by a food sensory evaluation, together which will last between 20 to 30 minutes. Ouring the session, you will be asked a few demographic questions for the sole purpose of keeping a record of demographics involved in the study. There is no direct benefit for participating. Individuals with known food allergies are excluded from participating. The food sensory evaluation sessions will be held on [add dates and times here] in the Central Station at HES building. Please contact

Kritika Shakya if you plan to attend

For information, contact

This project has been approved by the UCO IRB # 15200."

Nritika Shakya 405-837-1951 kshakya 16 uco edu



**Appendix 3: Informed consent form** 

# University Of Central Oklahoma

# Informed Consent Form

### **Research Project Title:**

Researcher: Dr. Kanika Bhargava and Kritika Shakya

- A. Purpose of this research: To compare the sensory characteristics of millet tortilla with commercial wheat tortilla. The goal of the research is to formulate and optimize a tortilla made out of millet and chickpea flour that is nutrient dense and is beneficial for people with diabetes.
- B. Procedures/treatments involved: Sensory evaluation done by the participants would help develop an optimized recipe. Participant would be asked to taste some samples and rate each on a questionnaire provided to them. Participants will be asked some basic demographic information such as age, gender, race/ethnicity, and education level.
- **C. Excepted length of the participation:** This study will take approximately 20-30 minutes to complete in 1 session.
- **D.** Potential benefits: There are no direct benefits to participation in this study. The benefit of this study will be determine if it is possible to develop a tortilla with similar sensory characteristics to commercial tortilla as a nutrient dense, gluten free product beneficial for people with diabetes.
- **E.** Potential risks or discomforts: The possible risks involved involves an allergic reaction to the ingredients in the tortilla.

In order for you to participate in this study, you must understand the risks involved regarding allergics to the ingredients. If you answer "yes" to any of the following questions and proceed to participate in the study, you should accept the responsibility to any reaction caused by ingredients in taste tests.

- Do you have any known allergies to food or food intolerance? Yes \_\_\_\_\_ No\_\_\_\_\_
- Are you allergic to millet or grains? Yes \_\_\_\_\_ No
- F. Medical/mental health contact information (if required): Students counseling services may be reached at (405)974-2215 for medical concerns contact Mercy Hospital at (405)755-1515, for medical emergencies call 911.

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- G. Contact information for researchers: If you have any questions, please feel free to contact Dr. Kanika Bhargava at 405-974-5556. You may ask questions now, or if you have any additional questions later, we will be happy to answer them.
- H. Contact information for UCO IRB: email at IRB@uco.edu or by phone at (405)974-5497
- I. Explanation of confidentiality and privacy: Any information obtained in connection with this research study that can be identified with you will be disclosed only with your permission; your results will be kept confidential. In any written reports or publications, no one will be identified or identifiable and only group data will be presented. The research results will be secured in a locked drawer in Dr. Bhargava's office and only the student researcher and faculty involved in the research will have access to the records. The data will be destroyed by shredding at the end of 3 years period after conclusion of the study.
- J. Assurance of voluntary participation: Participation in this research study is voluntary. You may also refuse to answer any questions you do not want to answer. If you volunteer to be in this study, you may withdraw from it at any time without consequences of any kind. There is no compensation given for signing up or participating in this study.

#### Affirmation by Research Subject:

I hereby voluntarily agree to participate in the above listed research project and further understand the above listed explanations and descriptions of the research project. I also understand that there is no penalty for refusal to participate, and that I am free to withdraw my consent and participation in this project at any time without penalty. I acknowledge that I am at least 18 years old. I have read and fully understand this Informed Consent Form. I sign it freely and voluntarily. I acknowledge that a copy of this Informed Consent Form has been given to me to keep.

Research Subject's Name:

Signature:	Date:
APPROVED	APPROVAL
DEC = 7 2015	DEC - 8 2016
UCO IRB	EXPIRES

### Appendix 4: Allergy and medical release form

Informed consent with allergy and medical release The following survey is a precursor to a sensory evaluation put on by researchers at University of Central Oklahoma. The goal of the sensory evaluation is to evaluate a potential new food product and its acceptance by the general public. The sensory evaluation and survey should take no longer than 20-30 minutes to complete. If you have any questions regarding this sensory evaluation you can contact Kritika Shakya at 405-837-1951 or by email at kshakya1@uco.edu, you can also contact Dr. Bhargava by email at kbhargava@uco.edu. If you would like to contact the Institutional Review Board at UCO they can be reached by phone at 405-974-5497 and by email at irb@uco.edu. Your privacy is very important your information will remain locked in Dr. Bhargava's office and will be shredded at the end of the mandatory period of 3 years. In order to participate in the sensory evaluation you need to answer the following questions. Do you have any known allergies or food intolerances? If yes please state specifically what you are allergic to and what effect it has on you. Are you allergic or intolerant to millet or grains? If yes please state specifically what you are allergic to and what effect it has on you. APPROVAL APPROVED DEC - 7 2015 UCO IRB EXPIRES

## **Appendix 5: Demographic survey**

Please com	Please complete the information below:				
Age:					
□ 18-25	□ 26-30	□ 31-35	□ 36-40	□ 41-45	□ 46-50
□ 51-55	□ 56-60	□ 61-70	□ 71-80	□ 81-90	□ Over 90
Gender:					
□ Male	□ Female				
Education Completed:					

$\Box$ High School	□ Some College	$\Box$ B.S.	$\Box$ M.S.
□ Ph.D. MD	□ Other		

# About how often do you eat flour tortillas? (Soft tacos, burritos, wraps, etc.)

□ Every Day	$\Box$ At least once a Week	$\Box$ Once every Two Weeks
□ Once a Month	□ Once a Year	□ Never

# Thank you for your willingness to help.

# Appendix 6: Definitions and references sheet for descriptive analysis

# **Appearance**

Evenness of color:	Degree to which the color is free from variations of fluctuations.
	Tortilla placed on a white paper
	References: Mission flour tortilla (fajita size) = 2
	Mission 96% fat free whole wheat flour tortilla = $13$
Shape (round):	Being such that every part of the surface of the circumference is
	equidistant from the center.
Surface:	Degree to which the outer face presents variations of fluctuations by
	means of blistering and puffing.
	References: Mission 96% fat free whole wheat flour tortilla = $5$
	Mission flour tortilla (fajita size) = 11
<u>Texture (in hand)</u>	
Roughness:	The property of having a surface marked by irregularities, protuberances,
	or ridges
	References: Kool-aid gels (Soarin' Strawberry) = 1
	Orange peel $= 6$
	Lay's Classic potato chips = 10
	Nature Valley crunchy granola bar (Vanilla nut) = 14
Tearability:	Amount of force required to pull the tortilla apart.
	With a strip of tortilla, hold the top with one hand and pull down on the
	bottom of the strip with the other hand.
	References: Mission flour tortilla (fajita size) = 8

Kangaroo Quality pita pocket bread = 14

<u>Odor</u>

Sweet:	Aromatic associated with sugar, such as sucrose or honey.
	Reference: Great value extra firm granulated sugar = 2
	Nabisco Honey Maid graham cracker = 8
	Great value clover honey $= 14$
Musty:	Aromatic associated with a dust or earthy from grain.
	Reference: Bag of millet flour = 11
<u>Flavor</u>	
Salty:	A fundamental taste factor of which sodium chloride solution is typical.
	Reference: 0.2% NaCl solution = 4
	0.4% NaCl solution = 7
	0.8% NaCl solution = 15
Sweet:	A fundamental taste factor of which sucrose solution is typical.
	Reference: $1.0\%$ sucrose solution = $3$
	2.0% sucrose solution = $6$
	4.0% sucrose solution = 11
	8.0% sucrose solution = $15$
Bitter:	A fundamental taste factor which caffeine solution is typical.
	Reference: $0.02\%$ caffeine solution = 2
	0.04% caffeine solution = 7
	0.06% caffeine solution = 14

Doughy:	A flavor associated with wet flour or dough
	Reference: King's Hawaiian savory butter roll = 5
	Pillsbury grands homestyle canned biscuit dough = 15
Nutty:	A sweet, light brown, slightly musty and/0r earthy flavor associated with
	nuts, grains. And seeds.
	References: Kretschmer Original toasted wheat $germ = 10$
<b>Texture (by mouth</b>	<u>)</u>
Hardness:	The relative resistance to deformation.
	Bite down evenly using front teeth.
	References: Kraft Philadelphia original cream cheese = 1
	Great value sharp cheddar cheese $= 4$
	Great value party peanuts = 13
Fracturability:	Force with which sample breaks.
	Cite down evenly using front teeth until sample breaks.
	References: "Jiffy" prepared corn muffin = 2
	Graham crackers = 7
Grittiness:	Amount of gritty particles perceived in the sample during mastication.
	Measure after 5-7 chews with molar.
	References: Post Grape nuts = 14

### **Appendix 7: Descriptive sensory evaluation instrument**

Date: \_\_\_\_\_

Panelist #: \_\_\_\_\_

Sample #:

**Instruction:** 

You have been presented with sample of tortillas. Inspect each sample in the order

presented and indicate its characteristics. Score with a check on numeric intensity scale of

0-15 where 0 is least intense and 15 is extremely intense.

### Please rate the flavor of the sample

1	2	3	4	5	6	7	8	9	10	11	12	13
14	15											
Salty	y											
1	2	3	4	5	6	7	8	9	10	11	12	13
14	15											
Nutt	y											

1	2	3	4	5	6	7	8	9	10	11	12	13
14	15											
Bitte	er											
1	2	3	4	5	6	7	8	9	10	11	12	13
14	15											
Dou	ghy											
1	2	3	4	5	6	7	8	9	10	11	12	13
14	15											
<u>Plea</u>	se rate t	he odor	<u>of the</u>	<u>sample</u>								
Swee	et											
1	2	3	4	5	6	7	8	9	10	11	12	13
14	15											
Mus	ty											

1	2	3	4	5	6	7	8	9	10	11	12	13		
14	15													
<u>Pleas</u>	<u>Please rate the texture (in the hand)</u>													
Roug	ghness													
1	2	3	4	5	6	7	8	9	10	11	12	13		
14	15													
Tear	ability													
1	2	3	4	5	6	7	8	9	10	11	12	13		
14	15													
<u>Pleas</u>	se rate tl	he textu	re (by th	he mouth	<u>h)</u>									
Harc	lness													
1	2	3	4	5	6	7	8	9	10	11	12	13		
14	15													
Frac	turabili	ty												

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1	2	3	4	5	6	7	8	9	10	11	12	13		
14	15													
Gritt	iness													
1	2	3	4	5	6	7	8	9	10	11	12	13		
14	15													
<u>Pleas</u>	se rate th	ie apped	arance o	of the sa	<u>mple</u>									
Even	<u>Please rate the appearance of the sample</u> Evenness of the color													
1	2	3	4	5	6	7	8	9	10	11	12	13		
14	15													
Shap	e (roun	d)												
1	2	3	4	5	6	7	8	9	10	11	12	13		
14	15													
Surfa	ace													
1	2	3	4	5	6	7	8	9	10	11	12	13		
14	15													

Muic		<u>1 uii iin</u>	uviii y (	j mis si			CAH CHI	ciy uisii				
<u>disli</u>	ke and I	<u>15 is ex</u>	tremely	like.								
1	2	3	4	5	6	7	8	9	10	11	12	13
14	15											

# Rate the overall likability of this sample where 1 is extremely dislike 8 is neither like nor

## **Appendix 8: Sensory acceptability evaluation instrument**

	y Evaluatio	511					Dat	ticipant # e
Please rate sensory	abaractoristics	of the comple by	abaaking the	appropriate how	y using the fo	llowing coole	San	nple #
	characteristics	of the sample by	checking the	appropriate bos	x using the to	mowing scale		
EXAMPLE:								
Dislike Extremely	□ Dislike Very Much	□ Dislike Moderately	Dislike Slightly	⊠ Neither Like nor Dislike	□ Like Slightly	□ Like Moderately	□ Like Very Much	□ Like Extremely
First, record your pa Next, take one plate Before you begin to Fake a normal-sized preference. Addition and spit out) the san	with a labeled taste the samp l bite, chew it hal bites can be	l sample and reco ble, assess its app 8 times, and then	ord its code in earance and a rate it for tex	the upper right roma. ture, tenderness	-hand corner	of this form. verall likeabil		
Feel free to ask que:	stions regardin	g the tasting inst	ructions durin	g any point of t	he sensory ev	valuation.		
Appearance:								
Dislike Extremely	□ Dislike Very Much	□ Dislike Moderately	Dislike Slightly	□ Neither Like nor Dislike	□ Like Slightly	□ Like Moderately	□ Like Very Much	□ Like Extremely
		like about the sar						
Aroma: D Dislike Extremely	Dislike Very Much	□ Dislike Moderately	Dislike	□ Neither Like nor Dislike	□ Like Slightly	□ Like Moderate	Like Ly Very Much	□ Like Extremely
Dislike Extremely	Dislike Very Much	□ Dislike Moderately	Dislike Slightly	□ Neither Like nor Dislike	Like	Like	Like ly Very	Like
□ Dislike	Dislike Very Much	□ Dislike Moderately	Dislike Slightly	□ Neither Like nor Dislike	Like	Like	Like ly Very	Like
Dislike Extremely Specifically, what	Dislike Very Much	□ Dislike Moderately	Dislike Slightly ample's arom	□ Neither Like nor Dislike	Like	Like Moderate □ Like	Like ly Very Much	Like
Dislike Extremely Specifically, what Texture: Dislike	Dislike Very Much did you like/d Dislike Very Much	□ Dislike Moderately islike about the s Dislike Moderately	Dislike Slightly ample's arom Dislike Slightly	□ Neither Like nor Dislike na? □ Neither Like nor Dislike	Like Slightly Like	Like Moderate □ Like	Like ly Very Much	Like Extremely Like
Dislike Extremely Specifically, what Texture: Dislike Extremely	Dislike Very Much did you like/d Dislike Very Much	□ Dislike Moderately islike about the s Dislike Moderately	Dislike Slightly ample's arom Dislike Slightly	□ Neither Like nor Dislike na? □ Neither Like nor Dislike	Like Slightly Like	Like Moderate □ Like	Like ly Very Much	Like Extremely Like

Specifically, what did you like/dislike about the sample's tenderness?

Dislike	Dislike	Dislike	Dislike	Neither	Like	Like	Like	Like
Extremely	Very Much	Moderately	Slightly	Like nor Dislike	Slightly	Moderately	Very Much	Extremely
ïcally, what d	id you like/di	slike about the sa	mple's taste?					

Dislike	Dislike	Dislike	Dislike	Neither	Like	Like	Like	Like
Extremely	Very	Moderately	Slightly	Like nor	Slightly	Moderately	Very	Extremely
	Much			Dislike			Much	

Specifically, what did you like/dislike about the sample overall?