Whole Grain Wheat Consumption in Young Children Can Increase Through Repeated Exposure and Use of Hard White Wheat

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Authorization to Submit Thesis

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Abstract

National recommendations encourage whole grain (WG) consumption in children. The objective of this study was to assess children's preference and consumption of WG wheat (with hard white wheat and hard red wheat) and the effects of repeated exposure. Two cohorts of children ages 2-6 years of age (n=101) participated in the study. Caregivers completed a questionnaire and children participated in taste activities with WG bread and tortillas. Descriptive statistics were reported for questionnaire responses. T-test and ANOVA were used to determine intake differences and the Mann-Whitney U test was used to identify differences in children's reported liking of all samples between cohorts. Parents and children consumed WG bread (66%); however, 22% consumed wheat or white (not understanding WG). Children reported liking hard white wheat significantly more (p=.042) than hard red wheat bread, and they consumed greater amounts of hard white wheat but the difference was not significant. Repeated exposure resulted in significant differences in intake for all bread and tortillas. Results from the study demonstrated the benefit of repeated exposure and potential use of hard white wheat to increase WG consumption in young children.

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List of Abbreviations

AI Adequate Intake

DI Deionized Water

FDA Food and Drug Administration

HRB Hard Red Bread

HRDB Hard Red Dyed Bread

HRT Hard Red Tortilla

HRW Hard Red Wheat

HWB Hard White Bread

HWDB Hard White Dyed Bread

HWT Hard White Tortilla

HWW Hard White Wheat

USCB United States Census Bureau

USDA United States Department of Agriculture

Chapter One

Introduction

Many health benefits are associated with whole grain consumption, including a reduced risk of chronic diseases and some cancers (Anderson, 2002; Jacobs, Marquart, Slavin, & Kushi, 1998; Liu, Stampfer, Hu, Giovannucci, Rimm, Manson, & Willett, 1999; Thompson, 1994). Whole grains contain nutrients, specifically fiber, phytochemicals, vitamins, and minerals, (USDA, 2007; Liu, 2004; Slavin, Jacobs, Marquart, & Wiemer, 2001), which support the overall health and development of children and adolescents (Jaramillion, Yang, Hugher, Orlet-fisher, Morales, & Nicklas, 2006). Despite the benefits of whole grains, few children consume adequate amounts of whole grains (Cleveland, Moshfegh, Albertson, & Goldman, 2000; Slavin, 2004). Consumption of whole grains is influenced by many factors, including consumer confusion (Marquart, Wiemer, Jones, & Jacob, 2003; Seal, Jones, & Whitney, 2006), taste preferences (Birch, 1999; Birch, 1987; Birch & Fisher, 1998), and repeated exposure (Birch & Fisher, 1998).

Whole grains contain all three anatomical features of the kernel, including the bran, germ, and endosperm (Lang & Jebb, 2003). Each layer of the grain contains specific nutrients that are valuable to overall health and development (Lang & Jebb, 2003). Whole grains can be refined, which is the removal of the bran and germ through milling, resulting in the loss of nutrients compared to whole grains (Lang & Jebb, 2003). Refined grains do not contain fiber, phytonutrients, antioxidants, vitamins, and minerals (Kantor, Variyam, Allshouse, Putnam, & Lin, 2001). Due to the increased nutrient content of whole grains, consumption of whole grains is encouraged, as they support overall health and reduce the risk of chronic diseases (Topping, 2007).

Whole grain consumption has been recommended as a means in which to support overall health of adults and children (Slavin, 2004). Whole grain consumption is associated with significantly greater amounts of macronutrients, vitamins, and minerals, specifically carbohydrates, fiber, folate, magnesium, phosphorous, and iron (O'Neil, Nicklas, Zanovec, Cho, & Kleinman, 2011). In 2005, the Dietary Guidelines recommended Americans consume three servings of whole grains per day (USDA, 2000) and half of their grains as whole grains (Mancino, Kuchler, & Leibtag, 2008). However, children ages 2-18 do not meet the recommended intake of 3 servings of whole grains per day (Harnack, Walters, & Jacobs, 2003). As few as 9% of preschool children and 16% of adolescents are consuming 2 servings of whole grains per day (Harnack, Walters, & Jacobs, 2003).

The failure to meet whole grain consumption recommendations is due to many factors. A few of the factors include consumer confusion, individual intake preferences, and whether children have been repeatedly exposed to whole grains. Consumers indicate, despite the health benefits, that whole grain products are difficult to identify, presenting a barrier for whole grain consumption (Marquart, Wiemer, Jones, & Jacob, 2003; Seal, Jones, & Whitney, 2006). Adults and children are unaware of the quantity of whole grains they are consuming due to the difficulty identifying whole grain products (Mancino et al., 2008). The Federal Drug Administration (FDA) does not require manufactures to include the amount of whole grains in a whole grain product label (Mancino et al., 2008). Food products that contain 51% whole wheat grain and 11% fiber may make health claims about decreased risk for heart disease and cancer on the product (Mancino et al., 2008). Additionally, the term "whole wheat" is not regulated by the FDA, and does not indicate that a product is 100% whole wheat grain (Mancino et al., 2008). These barriers create consumer confusion and

affect adult selection of whole grain products, which influence children's whole grain consumption.

Another factor is taste preferences, which is the primary factor influencing children's intake of foods (Birch, 1999; Guthrie, Rapoport, & Wardle, 2000). Children's food preferences are established at an early age, and translate into adulthood (Birch, 1987; Birch & Fisher, 1998). Children inherently prefer sweet and salty compared to bitter and sour (Steiner, 1979), which influence intake and possibly whole grain consumption. Whole grains such as hard white wheat contain fewer phenolic compounds compared to hard red wheat; therefore they are lighter in color, and less bitter in taste (Ransom, Berzonsky, & Sorenson, 2006), and could create an opportunity to increase children's whole grain consumption.

Taste preferences in children are fostered by repeated exposure to foods (Birch, 1979; Birch & Fisher, 1998; Rozin, 1990; Wardel, Cooke, Gibson, Sapochnik, Sheiham, & Lawson, 2003). By repeatedly exposing children to new foods, a greater acceptance of nutrient dense foods can be developed (Anzaman, Rollins, & Birch, 2010; Rozin, 1976; Birch, 1987), impacting children's overall health (Birch & Fisher, 1998). In consideration of children's development of taste preferences, it is important to understand children's preference for whole grains and identify strategies to increase whole grain consumption.

Problem Statement

Children in the United States (US), do not meet the recommended 3 servings of whole grains per day (Harnack et al., 2003). Many factors effect children's consumption of whole grains including consumer confusion (Marquart et al., 2003; Seal et al., 2006), taste preferences (Birch, 1999; Birch, 1987; Birch & Fisher, 1998), and repeated exposure (Birch & Fisher, 1998). Whole grain wheat products, predominately those prepared with hard red

wheat flour have a greater bitter taste (Ransom et al., 2006), and due to children's aversion to bitter tastes (Steiner, 1979) their consumption may be lower. Hard white wheat has fewer phenolic compounds, and tastes less bitter than hard red wheat, and previous research indicates they are preferred by children (Keeney, Ramsay, Tsao, & Plank, 2016). However, additional research is needed to determine the effects repeated exposure on consumption of whole grain wheat bread and to confirm children's taste preferences of hard white wheat bread and tortillas.

Statement of Purpose

The primary purpose of this quantitative study was to determine whether repeated exposure of hard white wheat whole grain and hard red wheat whole grain bread and tortillas would increase children's liking and intake of whole grain wheat bread and tortillas. A secondary purpose was to determine whether children's liking and intake of bread and tortillas was greater with hard white wheat whole grain versus hard red wheat whole grain.

Research Question

Primary research question: Does repeated exposure of hard white wheat whole grain and hard red wheat whole grain bread and tortillas increase children's preference and intake of whole wheat bread and tortillas? Secondary research question: Do children report a greater preference and intake of hard white wheat whole grain versus hard red wheat whole grain bread and tortillas?

Significance of Study

There are many health benefits associated with whole grain consumption including a reduced risk of chronic diseases and certain cancers (Anderson, 2002; Jacobs et al., 1998; Liu et al., 1999). Because of these health benefits, government promotion of whole grains

has increased (Andreyeva & Luedicke, 2013). While the Dietary Guidelines recommend children consume at least 3 servings of whole grains per day and to make half of all grain consumption whole grains (USDA, 2000; Mancino et al., 2008), children are not meeting the recommended intake of whole grains (Harnack et al., 2003).

Barriers to whole grain consumption include consumer confusion (Marquart et al., 2003; Seal et al., 2006), taste preferences (Birch, 1999; Birch, 1987; Birch & Fisher, 1998), and repeated exposure (Birch & Fisher, 1998). Due to a lack of regulation in labeling and packaging of whole grain products, consumers report confusion regarding the amount of whole grains they are consuming (Harnack et al., 2003). Additionally, taste preference and repeated exposure foster consumption of foods (Birch & Fisher, 1998), but adults often fail to repeatedly offer whole grains to children after they have been rejected. Repeatedly offering foods, supports acceptance for whole grains (Anzaman, Rollins, & Birch, 2010; Rozin, 1976). Hard white wheat provides an opportunity to increase children's whole grain consumption and liking of whole grains due to the presence of fewer phenolic compounds, less bitter taste, and lighter color (Keeney, Ramsay, Tsao, & Planck, 2016). Therefore, continued research to address these issues in children's whole grain consumption is needed.

Definition of Terms

Hard Red Wheat:

> A type of whole wheat characterized by a hard endosperm and a red bran (Ransom et al., 2006).

Hard White Wheat:

> A type of whole wheat characterized by a hard endosperm and white bran (Ransom et al., 2006).

6-n- propythiouracil (PROP) test:

An edible chemical compound used to determine genetic sensitivity levels to bitter tastes (Hedge & Sharma, 2007).

Taste preference:

Predisposition for certain flavors, which influences types of foods and amounts consumed (Geller, Rovner, & Nansel, 2009).

Repeated exposures:

Exposure to a flavor or to a variety of flavors within a particular food group (e.g., fruits or vegetables) following 8-10 exposures of continuous food introductions (Mennella & Ventura, 2010).

Limitations

This study has several limitations. First, participating parents were primarily Caucasian, moderately high socio-economic status, with a bachelor's degree or beyond, which does not reflect the majority of the population of the United States, however the demographic is an accurate representation of the Pacific Northwest (United States Census Bureau, 2010). Results cannot be generalized to the population of the United States, but provide insight to repeated exposure and preference in children.

The majority of children who participated in the study were exposed to whole grain foods prior to their involvement in the study. Exposure to whole grain foods outside the tasting activities may have influenced children's reported preference of whole grains.

Summary

Many health benefits are associated with whole grain consumption, including a reduced risk of chronic diseases and some cancers (Anderson, 2002; Jacobs et al., 1998; Liu

et al., 1999; Thompson, 1994). Few children consume adequate amounts of whole grains (Cleveland et al., 2000; Slavin, 2004), which are needed for overall health and development of children and adolescents (Jaramillion et al., 2006). Many factors are associated with children's consumption of whole grains, including consumer confusion (Marquart et al., 2003; Seal et al., 2006), taste preferences (Birch, 1999; Birch, 1987; Birch & Fisher, 1998), and repeated exposure (Birch & Fisher, 1998).

Previous research indicates young children prefer hard white wheat versus hard red wheat (Keeny et al., 2016), however the difference in liking and intake between hard white wheat and hard red wheat bread and tortillas, as well as the effect of repeated exposure has not been studied. Further research is necessary to identify children's taste preferences of hard white versus hard red wheat whole grain bread and tortillas, whether the color of bread and tortillas affect children's liking and intake, and the affect of repeated exposure on intake and liking of whole wheat bread and tortillas.

The primary purpose of this quantitative study was to determine whether repeated exposure of hard white wheat whole grain and hard red wheat whole grain bread and tortillas would increase children's liking and intake of whole grain wheat bread and tortillas. A secondary purpose was to determine whether children's liking and intake of bread and tortillas was greater with hard white wheat whole grain versus hard red wheat whole grain. The first chapter provided an introduction, problem statement, purpose statement, research question, significance of study, definition of terms, and limitations of the study. The second chapter will review the literature on the components of whole grains, nutrients in wheat grains, whole grain consumption, and factors effecting whole grain consumption. The third

chapter is written as a manuscript and includes an introduction, the methodology, the results, the discussion, and implications from this research.

Chapter Two

Literature Review

Regular consumption of whole grains and whole grain products is associated with many health benefits, including a reduced risk for chronic diseases by promoting a healthy gastrointestinal tract, preventing cardiovascular disease, regulating blood glucose, preventing colon cancer prevention, and supporting weight management (Anderson, 2002; Jacobs, Marquart, Slavin, & Kushi, 1998; Liu et al., 1999; Morris, Marr, & Clayton, 1977; Thompson, 1994). Whole grains contain nutrients such as fiber (USDA, 2007), phytochemicals (Liu, 2004), vitamins, and minerals (Slavin, Jacobs, Marquart, & Wiemer, 2001) that support the overall health and development of children (Jaramillion, Yang, Hughes, Orlet-Fisher, Morales, & Nicklas, 2006). However, as few as 8% of adults and 3% of children consume the recommended intake of whole grains daily (Cleveland, Moshfegh, Albertson, & Goldman, 2000; Slavin, 2004).

Many factors are associated with children's consumption of whole grains, including taste preferences (Harnack, Walters, & Jacobs, 2003). Taste preferences develop during childhood and influence children's food choices into adulthood (Birch, 1987; Birch & Fisher, 1998); therefore, children's preference and consumption of whole grain varieties could influence future adult consumption of whole grains. Understanding children's preference for whole grains and identifying strategies to increase whole grain consumption could positively impact children's future health.

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secondary purpose was to determine whether children's liking and intake of bread and tortillas was greater with hard white wheat whole grain versus hard red wheat whole grain. With this information, a greater understanding of children's taste preferences of whole grains and strategies to increase whole grain consumption can be implemented to positively impact children's future health.

This literature review begins with an overview of the components of whole grains and types of wheat grain varieties. This discussion is followed by nutrients in wheat grains and their benefits to health. A detailed explanation of key nutrients in whole grains is presented to include fiber, phytochemical, vitamin, and mineral content. Last, a discussion of whole grain consumption, and factors affecting whole grain consumption, including consumer confusion, taste preferences, and repeated exposure, followed by the need for whole grain consumption in children, as well as government promotion, is reviewed.

Components of Whole Grains

A whole grain consists of three anatomical features, the bran, endosperm, and the germ, which comprise the entire grain kernel (Lang & Jebb, 2003). Each anatomical feature of a whole grain contains nutrients that contribute to the overall health benefits of whole grain consumption (Lang & Jebb, 2003). The bran is the outer layer that is rich in B vitamins and phytonutrients (Lang & Jebb, 2003). The endosperm is the middle layer that consists of 80-85% carbohydrates and 12-14% protein for hard wheats (Lang & Jebb, 2003). The germ is the inner layer that contains minerals, such as zinc and iron, and antioxidants such as vitamin E (Lang & Jebb, 2003).

Whole grains are defined by the Federal Drug Administration (FDA) as consisting of "intact, ground, cracked or flaked caropsis (fruit or kernel) of the grain whose principal

components, the starchy endosperm, germ and bran, are present in the same relative proportions as they exist in the intact grain" (AACC International, 2000, p. 191). Whole grain foods can contain the intact whole grain, or components of the whole grain can be reconstituted and recombined in the relative proportions that occur in the grain kernel (USDA, 2007). For a wheat product to be considered whole grain, it must be at least 51% whole grain flour by weight (Slavin, 2004). The most common whole wheat grain products include bread, rolls, flour, tortillas, rice, cereal, pasta, ready-to-eat breakfast cereal, and ready-to-cook breakfast cereal (Kantor, Variyam, Allshouse, Putnam, & Lin, 2001).

Whole grains can be refined, to give them a finer texture, lighter color, and extended shelf life (Lang & Jebb, 2003). Refined grains are milled grains on which the bran and germ are removed (Lang & Jebb, 2003). The removal of the bran and germ results in a grain comprised of mainly the endosperm, or carbohydrate and protein layer, thus resulting in the loss of nutrients found in the bran and germ layers, such as vitamin, mineral, phytochemical, and antioxidant (Lang & Jebb, 2003). Due to the inclusion of the bran and germ, whole grains differ from refined grains in that they have a dense texture, darker color, and provide greater amounts of vitamins, minerals, fiber, phytonutrients, and antioxidants; all of which contribute to the additional health benefits of whole grains compared to refined grains (Kantor et al., 2001). Refined grains are low in fiber, as well as phytonutrients and antioxidants that accompany fiber (Kantor et al., 2001).

Types of Wheat Grain Varieties

Many varieties of whole wheat grains exist, including red, white, hard, soft, winter, and spring types (Liu, 2007). Wheat varieties are grown for specific end products such as bread, rolls, flour, tortillas, rice, cereal, pasta, ready-to-eat breakfast cereal, and ready-to-

cook breakfast cereal (Adom, Sorrells, & Liu, 2003). The following paragraphs review the color of grains, varieties in texture such as hard and soft grains, and varieties in season including spring or winter wheat.

The color of the grain determines the wheat variety classification as red or white (Adom et al., 2003). Hard red wheat flour is characterized by high levels of gluten and is best used for bread and cakes (Marconi & Carcea, 2001). Hard white wheat is a new type of wheat in the US market (Ransom, Berzonsky, & Sorenson, 2006). Hard white wheat and hard red wheat vary by use and composition, however hard white wheat does not contain the brown color in the outer seed coat, and it is less bitter due to fewer phenolic compounds and phenolic compounds (Ransom et al., 2006). Hard white wheat is being used to increase US whole grain consumption because of the similar appearance (higher pigmentation) and taste to traditional white bread provided from refined hard red wheat (Ransom et al., 2006).

The texture of the grain determines whether wheat is classified as hard or soft wheat (Martin, Frohberg, Morris, Talbert, & Giroux, 2001). Wheat hardness is determined by a chromosomal genetic variation, of either hard or soft properties (Martin et al., 2001). The end-use of wheat (baked goods, bread, tortillas, and cereals) determines the hardness of the wheat used (Martin et al., 2001).

Wheat varieties are referred to as winter or spring. The season that the wheat is grown determines if it is a spring or winter variety (Adom et al., 2003); however, the USDA does not make a distinction between winter and spring hard white wheat varieties (Ransom et al. 2006). This results in a lack of spring, winter, red and white wheat terminology on food packages and labels in stores, however wheat is bought and sold by class.

Specifications of grains including color, texture, and season variety determine the end-use of grains as baked goods, bread, tortillas, and cereals (Martin et al., 2001). Genotypic differences in the variety of wheat grains result in varying nutritional values (Liu, 2007); therefore, understanding wheat varieties can be useful in supporting consumer overall health.

Important Nutrients in Wheat Grains That Benefit Health

Wheat grains contain nutrients vital to the health of adults and children (Topping, 2007). Among these nutrients are protein, carbohydrates, vitamins, minerals, fiber, phytonutrients, and antioxidants (Kriestensen et al., 2010; Anderson, Smith, & Gustafson, 1994; Fardet, 2010; Adom et al., 2003; Liu, 2007; Okarter & Liu, 2010). Whole wheat grains are contain as complex carbohydrates, which provide energy, aid in digestion, and control bacterial infections in the gastrointestinal tract (Topping & Clifton, 2001; Ramakrishna et al., 2000). Vitamin and mineral content in whole grains is associated with many health benefits and is protective against heart disease and some cancers (USDA, 2000). Key nutrients in whole grains and their health benefits are described below.

The fiber content of whole grains for health. The fiber in whole grains can contribute to an individual's consumption of dietary fiber, which provides numerous health benefits (USDA, 2007). Dietary fiber is defined as the components of plant cells that are not broken down by human digestive enzymes (Burkitt & Trowell, 1986). Fiber is classified as soluble and insoluble fiber (Gropper & Smith, 2013). Wheat grains contain soluble fiber in the endosperm, and insoluble fiber in the bran, both of which benefit health (Topping, 2007). Insoluble fiber, such as wheat bran, aids digestion (Topping, 2007).

It is recommended that individuals should consume fiber daily, which is indicated as Adequate Intakes (AI), and determined based on the amount of fiber healthy people consumed in reference to their age and gender (USDA, 2007). The AI for women ages 19 to 50 years is 26 grams, and 38 grams per day for men, (USDA, 2007). Children ages 3 to 5 years should consume half of the recommended intake of fiber, which is 11 grams per every 1000 calories (USDA, 2007). The most common sources of fiber are fruits, vegetables, and grains (USDA, 2007), such as whole wheat.

Consumption of fiber from whole grains is the leading source of dietary fiber in the prevention and treatment of chronic diseases (Anderson et al., 1994). High fiber intake is associated with a decreased risk of coronary heart disease, reduced blood pressure, weight management, glycemic control, gastrointestinal function, and some cancers (Anderson et al., 1994). Since whole grains contain the bran layer, fiber intake increases as whole grain intake increases (O'Neil, Nicklas, Zanovec, Cho, & Kleinman, 2011). Hard white wheat offers an opportunity to increase children's consumption of whole grains and fiber due to its similar taste and appearance as traditional white bread (Ransom et al., 2006). With the numerous health benefits of fiber, ensuring young children consume sufficient amount is paramount.

The phytochemical content of whole grains for health. Phytochemicals are biologically active components found in plants, such as whole grains (Liu, 2004). Some phytochemicals are responsible for the deep colors of fruits and vegetables as well as the darker brown in whole grains; they may be responsible for the added benefits of whole grains beyond basic nutrition (Liu, 2004). Health benefits of whole grains also are attributed to antioxidant components as well as phytochemicals, which have been shown to reduce oxidative damage in the body (Adom & Liu, 2002).

Whole grains contain significant amounts of phytochemicals (Liu, 2004; Slavin, 2001). Different types and varieties of whole grains influence the concentration of phytochemicals (Adom et al., 2003). The most important groups of phytochemicals found in whole grains consist of phenolics, carotenoids, lignans, β-glucans, and inulin (Liu, 2007). Carotenoid pigmentation is responsible for the color of whole grains and contain antioxidant properties, thus contributing to their role in decreasing risk of chronic diseases and cancer (Liu, 2007). β-glucans aid in maintaining healthy blood cholesterol blood sugar levels as well as support immunity (Liu, 2007). Inulin acts like a prebiotic to stimulate growth of healthy bacteria in the intestines to support colon health (Liu, 2007). As a result, consumption of whole grains creates the opportunity to ingest a diverse variety of phytochemicals beneficial to health.

The vitamin and mineral content in whole grains for health. The vitamin and mineral content in whole grains is higher than that of refined grains (O'Neil et al., 2011). Whole grains contain vitamins and minerals found in the intact bran of the grain kernel (O'Neil et al., 2011). Specifically, vitamins such as B vitamins, vitamin E, and minerals such as selenium, zinc, copper, calcium, potassium, and magnesium, which are lacking in the typical American diet, are presented as whole grains (Slavin et al., 2001). B vitamins in whole grains include thiamin, niacin, riboflavin, and pantothenic acid (O'Neil et al., 2011). Vitamin E compounds play a role in immunity, DNA repair, and other metabolic processes (Traber, 1999). Calcium and zinc are of specific concern due to their aide in physical growth and development of children (Mahan & Escott-Stump, 2008). Zinc is necessary for proper growth of children as well as appetite, and taste acuity (Mahan & Escott-Stump, 2008). Some children do not meet the recommendation for zinc intake (Mahan & Escott-Stump, 2008),

creating an opportunity for whole grains to increase children's zinc intake. Calcium intake in childhood is pertinent for mineralization and maintenance of bone growth (Mahan & Escott-Stump, 2008). It is recommended that children ages 3-8 years consume 700-1000 mg of calcium per day to achieve optimal bone growth (Gropper & Smith, 2013). Adequate intake of vitamins and minerals are necessary for growth and development of children by supporting overall health and reducing the risk of chronic diseases (Mahan & Escott-Stump, 2008), and whole grains provide more vitamins and minerals.

Benefits of Whole Grains to Overall Health

Consumption of whole grains is associated with a reduced risk of chronic diseases such as cardiovascular disease, diabetes, certain cancers, and excess weight gain occur less frequently in individuals who consume whole grains (Topping, 2007). Whole grain intake is important for children's improved diet quality, growth, development of a healthy gastrointestinal tract, learning, play, and disease prevention (Slavin et al., 2001). Whole grain intake is associated with improved diet quality and nutrient intake in children and adolescents (O'Neil et al., 2011). Children's long-term health status is affected by their current preferences and food intake (Jaramillion et al., 2006). Therefore, identifying children's preference for whole grains has implications for children's current and future health. The specific health benefits such as a healthy gastrointestinal tract, prevention of cardiovascular disease, diabetes, cancer, and weight management are described.

A healthy gastrointestinal tract from whole grain consumption. A healthy gastrointestinal tract is characterized by the absence of disease, immune responses, minimal inflammation, and efficient digestion (Jonnalagadda el al., 2011). Fiber is characterized as soluble and insoluble fiber (Gropper & Smith, 2013). Soluble fiber retains water and

becomes a gel like substance that slows down digestion, where as in-soluble fiber does not retain water and aids in the formation of fecal bulk, causing it to pass more quickly through the gastrointestinal tract (Gropper & Smith, 2013). Consumption of fiber from whole grains, which provides phytonutrients, vitamins, and minerals, supports a healthy gastrointestinal tract (Jonnalagadda el al., 2011). A healthy gastrointestinal tract reduces the risk of diverticulitis and certain cancers (Jonnalagadda et al., 2011). The fiber content of whole grains aids in digestion, promotes fecal bulk, decreased transit time, and increases the removal of carcinogenic compounds (Sengupta, Tjandra, & Gibson, 2001). Whole grains also have been suggested to have probiotic effects, increasing beneficial bacteria in the gastrointestinal tract and, influencing immune response (Jonnalagadda el al., 2011). The amount of microflora in the gut is associated with short chain fatty acids in whole grain foods and are beneficial to health (Topping, 2007). Whole grains contribute to the maintenance of gastrointestinal function and protect against diseases (Slavin, 2013).

Cardiovascular disease prevention from whole grain consumption. Whole grain consumption reduces the risk for coronary heart disease because elevated cholesterol, specifically low-density lipoprotein, is a risk factor for coronary heart disease (Topping, 2007). Cholesterol that is bound to fiber inhibits the formation of micelles, therefore cannot be absorbed in the small intestine and is excreted in the feces as well as bile recirculation and subsequent removal of cholesterol from the blood for formation of new bile (Gropper & Smith, 2013). Thus, whole grain intake is associated with decreased serum cholesterol levels, a risk factor for cardiovascular disease (Harnack et al., 2003).

Blood glucose regulation from whole grain consumption. Whole grains contain properties which aide in blood glucose homeostasis (Slavin, 2004). High fiber foods, such as

whole grains can support blood glucose regulation. For example, high fiber foods take longer to eat, increase satiety, slow gastric emptying, increase feelings of fullness, decrease serum insulin, decrease food intake due to decrease insulin stimulating appetite, decrease energy availability, and increase fermentation of foods (Anderson et al., 1994).Whole grains decrease the rate of glucose entry (from digested carbohydrates) into the blood as well as the total amount of glucose released, therefore an increase in fiber intake from whole grains leads to improved glucose regulation in the blood, resulting in decreased prevalence of diabetes (Slavin, 2004).

Cancer prevention from whole grain consumption. Epidemiologic evidence indicates whole grain intake has been associated with a decreased risk of colon cancer (Slavin, 2004; Harnack et al., 2003). Whole grain consumption increases fecal bulk, leading to waste removal of carcinogenic toxins in the colon, decreasing an individual's risk for colon cancer ; as a result, whole grain consumption is associated with an inverse relationship to certain cancers, including cancers of the gastrointestinal tract (Jonnalagadda el al., 2011).

Weight management from whole grain consumption. Increased fiber intake is associated with whole grain consumption and has been associated with decreased weight gain in adults (Slavin, 2004). Whole grains enhance satiety, causing individuals to feel fuller longer, therefore, individuals consume less food and it is easier to maintain a healthy weight (Slavin, 2004). Body weight and diet are also correlated with health status, reducing the risk of chronic diseases including obesity (Healthy People, 2020).

Obesity rates in children ages 2-19 years are 18.2% for males and 16% for females, and there have been no statistically significant changes in obesity rates since 2012 (Healthy People, 2020). High fiber foods, such as whole grains, reduce risk for obesity and assist in

weight loss and weight maintenance (Andersonet al., 1994). Therefore, increased consumption of whole grains may assist in the reduction of obesity.

Whole Grain Consumption

Due to the health benefits of whole grains, consumption has been recommended to support adult and child health in the US (Slavin, 2004). The 2005 Dietary Guidelines, were the first to suggest Americans consume half of their grains as whole grains (Mancino, Kuchler, Leibtag, & 2008), consisting of three servings of whole grains per day (USDA, 2000). Children are consuming the recommended amount of grains, however daily consumption of whole grains is lacking (Harnack et al., 2003). Few children ages are meeting these recommendations, with only 9 percent of children ages consuming 3 or more servings of whole grains per day (Harnack et al., 2003). However, mean whole grain intake increases steadily from young children ages 2-5 years to adolescents ages 6-18 years, but as few as 9.3% of preschool children and 16.4% of adolescents are consuming 2 or more servings of whole grains per day (Harnack et al., 2003). Consumption patterns of whole grains is shifting (Kantor et al., 2001). Intake of grain foods has increased since record lows in the 1970's, however whole grain consumption remains low (Kantor et al., 2001).

Assessment of children's whole grain intake, and its effect on overall health status is important (Fox, Condon, Briefel, Reidy, & Deming, 2010). Young children ages 2-5 years who consume greater amounts of whole grains also consume significantly greater amounts of vitamins and minerals, carbohydrates, fiber, folate, magnesium, phosphorus, and iron as well (O'Neil et al., 2011). Furthermore, those children also consume significantly lower amounts of added sugars, total fat, saturated fatty acids, mono unsaturated fatty acids, and cholesterol

(O'Neil et al., 2011). Thus, whole grain consumption is accompanied by improvements in nutrient intake.

Wheat is the most commonly consumed grain in the United States, constituting 66-75% of total grain consumption and is consumed in the form of leavened bread (Slavin, 2004). Bread is indeed the most common food source of whole grains consumed by children, accounting for 21.7% of children's total whole grain intake and the other forms such as ready to eat cereals, corn and chips (Harnack et al., 2003). Whole grains are typically consumed inside the home (Bellisel, Hebel, Colin, Reye, & Hopkins, 2014). Parents who live with children in the home consume fewer whole grains than those who do not live with children in the home, indicating the possibility of parents influence on children's consumption of whole grains (O'Neil et al., 2011). Thus, parental involvement with children during meals, influences whole grain consumption.

Factors That Affect Whole Grain Consumption

A number of factors affect adult and child consumption of whole grains. Low consumption of whole grains has been associated with lacking consumer awareness of whole grain related health benefits, consumer confusion, taste preferences, and repeated exposure (Harnack et al., 2003). Despite the health benefits, consumers report that whole grains are difficult to identify (Marquart, Wiemer, Jones, & Jacob, 2003; Seal, Jones, & Whitney, 2006). Because of difficulty identifying whole grains, adults and children are unaware of their true whole grain consumption (Mancino et al., 2008); furthermore, the primary factor that affects children's whole grain consumption is taste (Birch, 1999; Guthrie, Rapoport, & Wardle, 2000). Other factors include consumer confusion, taste preferences, and repeated exposure are reviewed in the following paragraphs as addressed in the following:

Consumer confusion affect on whole grain consumption. Whole wheat consumption is correlated with the amount of whole grain products available on the market (Okarter & Liu, 2010). Whole grain breads are difficult to identify, which present a barrier for consumers to increase whole grain consumption (Marquart et al., 2003; Seal et al., 2006). In efforts to increase whole grain consumption, one of the aims of the Federal Drug Administration (FDA) was to revise food labeling requirements (Andreyeva & Luedicke, 2013). Unfortunately, the FDA does not require food manufactures to label their whole grain products with the amount of whole grain in the food item, causing consumer confusion of how much whole grains (in grams) they are consuming (Mancino et al., 2008). This revision instilled action by food manufactures to market their food products containing whole grains through advertising of their healthful attributes (Mancino et al., 2008).

Currently, an item may be labeled as "100% whole grain" or "10g of whole grains" as long as it is not misleading (Mancino et al., 2008). If the food product contains at least 51% whole grains and 11% fiber, food manufactures can make a whole grain health claim and market their product as a food that may decrease risk of heart disease or cancer (Mancino et al., 2008). Terms like "whole wheat" are often used by food manufactures, but they are not actually 100% whole grain products (Mancino et al., 2008). In order for a grain product to receive the Basic Stamp from the Whole Grains Council, it must contain at least 8g of whole grain (Whole Grains Council, N.D.).

Taste preference affect on whole grain consumption. Many factors effect children's taste preferences such as texture, color, and exposure to foods (Birch & Fisher, 1998). Taste is the primary factor influencing children's intake and consumption (Birch, 1999; Guthrie et al., 2000). Taste preferences developed in childhood could translate into

adulthood (Brannen & Fletcher, 1999, Ramsay, Rudley, Tonnemaker, & Price, 2015), and are influenced by environmental factors as well as genetic factors (Birch, 1999).

Children and adults have distinct taste preferences (Birch, 1987; Birch & Fisher, 1998), and taste preferences influence food intake (Birch, 1979; Birch & Fisher, 1998; Guthrie et al., 2000), as well as overall health status (Jaramillo et al., 2006). Children have inherent taste preferences that develop in utero and throughout early childhood, translating into adulthood (Birch & Dietz, 2008; Cooke & Fildes, 2011). Some factors that influence children's food preferences include home availability and accessibility, parenting style and parenting practices, and role modeling (O'Neil et al., 2011).

Children inherently prefer sweet and salty tastes compared to bitter and sour (Steiner, 1979). Whole grains, such as hard red wheat contain additional phenolic compounds, therefore their bitter taste is more apparent than in hard white wheat (Ransom et al., 2006). Thus, hard white wheat could provide an opportunity to increase whole grain consumption in children.

Repeated exposure affect on whole grain consumption. Research has found that repeated exposure fosters children's taste preferences (Birch, 1979; Birch & Fisher, 1998; Rozin, 1990; Wardle, Cooke, Gibson, Sapochnik, Sheiham, & Lawson, 2003). The development of food preferences also depend on the level of familiarity with foods; therefore, repeatedly exposing children to new foods, tastes, and flavors become an effective technique for children to accept a wide variety of nutrient dense foods (Anzaman, Rollins & Birch, 2010; Rozin, 1976; Birch, 1987; Birch, 1999). By repeatedly offering nutrient dense foods to children, children are to accept a wide variety of nutrient dense foods and establishing a healthy diet (Skinner et al., 2002)

Children do not innately like all foods, requiring an increased amount of exposures to increases their familiarity and liking of that food (Birch & Marlin, 1982; Birch, 1979). Repeated exposure may result in previously disliked foods becoming liked foods, and may prevent food neophobia (Cooke, 2007).

Government Promotion of Whole Grains

In consideration of the health benefits of whole grains, the US government promotes their consumption (Mancino et al., 2008). One promotion effort is aimed at increasing children's consumption of a variety of nutrient dense foods such as whole grains, fruits, vegetables, milk products, lean meats and protein sources (Healthy People, 2020). To increase children and adolescents consumption of whole grains per day, the National School Lunch Program and the School Breakfast Program have updated their guidelines, consistent with the dietary guidelines, for children regarding whole grains (Sox & Greenfield, 2009). Furthermore, Healthy People 2020 states several objectives to improve children's consumption of whole grains, specifically to increase the proportion of individuals ages 2 and older who consume the recommended 6 servings of grain products, 3 of those servings should constitute whole grains per day (Harnack et al., 2003).

Whole grain sales increased with the release of new whole grain products beginning in 2003 (Mancino et al., 2008). Tortillas are one of the fastest growing whole grain products in the consumer market (Kantor et al., 2001). Improving the nutritional content of tortillas is important as consumers are becoming more health conscious (Kantor et al., 2001). With the health benefits of whole grain consumption, government promotion of whole grains targets children and adolescents (Harnack et al., 2003). There is a demand, specifically for hard

white wheat, due to its more favorable appearance compared to hard red wheat, in the domestic and export markets (Ransom et al., 2006).

Conclusion

Many factors contribute to children's consumption of whole grains including overall health benefits, consumer confusion and taste (Harnack et al., 2008). Consumption of whole grains is associated with health benefits such as increased consumption of fiber, vitamins, minerals, phytonutrients, and antioxidants, various chronic diseases including cardiovascular disease, diabetes, colon cancer, and excess weight gain occur less frequently in those who consume whole grains (Topping, 2007).

Currently, children do not consume the recommended amount of whole grains (Harnack et al., 2008). Barriers to consumption of whole grains include consumer confusion and taste preferences (Marquart et al., 2003; Seal et al., 2006). Whole grains are difficult to identify, therefore consumers are unaware of whole grains and the amount of whole grain in a given product (Marquart et al., 2003). Taste, texture, appearance, and smell are also reported as barriers to whole grain consumption, with taste as the dominant barrier to consumption (McMackin, Dean, Woodside, & McKinley, 2013).

Children have distinct taste preferences develop in utero throughout early childhood and translate to adulthood (Birch, 1987). Inherently, children prefer sweet tastes compared to bitter (Steiner, 1979). Whole grains, specifically hard white wheat, contain fewer phenolic compounds than hard red wheat, therefore are less bitter (Ransom et al., 2006). Since food preferences depend on familiarity with foods, repeatedly exposing children to foods (such as whole grains) leads to acceptance of a variety of foods (Anzaman et al., 2010; Rozin, 1976; Birch, 1987; Birch, 1999). Expanding children's taste preferences to whole grain foods

provides an opportunity to increase children's acceptance and liking of whole grains, influencing their overall health status.

Whole grains are typically consumed as leavened bread by children (Slavin, 2004). Hard white wheat has positive attributes that children are familiar with and is a food that can increase children's intake of whole grains. Therefore, the primary purpose of this quantitative study was to determine whether repeated exposure of hard white wheat whole grain and hard red wheat whole grain bread and tortillas would increase children's liking and intake of whole grain wheat bread and tortillas. A secondary purpose was to determine whether children's liking and intake of bread and tortillas was greater with hard white wheat whole grain versus hard red wheat whole grain. With this information, a greater understanding of children's taste preferences of whole grains and strategies to increase whole grain consumption can positively impact children's future health.

Chapter Three

Children's Whole Grain Consumption and Liking of Hard White Versus Hard Red Wheat

Introduction

Regular consumption of whole grains by adults and children is associated with many health benefits, including the promotion of a healthy gastrointestinal tract, cardiovascular disease prevention, blood glucose regulation, colon cancer prevention, and promotion of weight management (Anderson 2002; Jacobs, Marquart, Slavin, & Kushi, 1998; Liu, et al., 1999; Morris, Marr, & Clayton, 1977; Thompson, 1994). Whole grains contain nutrients such as fiber (USDA, 2007), phytochemicals (Liu, 2004), vitamins, and minerals (Slavin, Jacobs, Marquart, & Wiemer, 2001) that are particularly important for the overall health and development of children (Jaramillion et al., 2006). However, as few as 3% of children consume the recommended intake of whole grains daily (Cleveland, Moshfegh, Albertson, & Goldman, 2000; Slavin, 2004).

Whole grains consist of all three grain anatomical features, the endosperm, germ, and bran (Lang & Jebb, 2003). Each layer of the grain contains specific nutrients that benefit overall health (Lang & Jebb, 2003). Whole grains can be refined, resulting in the loss of nutrient content found in the bran and germ (Lang & Jebb, 2003). Refined grains are lighter in color, softer in texture, and have a longer shelf life compared to whole wheat grains (Lang & Jebb, 2003). Therefore, whole grains contain a greater amount of nutrients compared to refined grains, and are darker in color, and a dense texture (Kantor, Variyam, Allshouse, Putnam, & Lin, 2001), resulting in a unique taste.

There are variety of whole wheat grain types (Liu, 2007). Wheat grain varieties are determined by the color, texture, and season wheat is grown (Liu, 2007). Wheat grains are classified as red or white depending on the color of the bran (Adom, Sorrells, & Liu, 2003). The texture of the grain determines if the wheat is considered hard or soft (Martin, Frohberg, Morris, Talbers, & Giroux, 2001). Hard white wheat is lighter in color and tastes less bitter compared to hard red wheat due to fewer phenolic compounds (Ransom, Berzonsky, & Sorenson, 2006).

The type of whole grain may influence consumption as many factors are associated with children's consumption of whole grains, including taste preferences (Harnack, Walters, & Jacobs, 2003). Taste preferences and exposure to foods develop during childhood and influence children's food choices into adulthood (Birch, 1987; Birch & Fisher, 1998); indicating, children's preference and consumption of whole grain varieties could influence future adult consumption of whole grains. Understanding children's preference for whole grains and identifying strategies to increase whole grain consumption also could positively impact children's future health.

Children's taste preferences are fostered by exposure to foods (Birch, 1979; Birch & Fisher, 1998; Rozin, 1976; Wardel et al., 2003). An acceptance to a wide variety of nutrient dense foods is dictated by food preference, familiarity with foods, and repeatedly exposing children to new foods, tastes, and flavors (Anzaman, Rollins, & Birch, 2010; Rozin, 1976; Birch, 1987). Repeatedly offering nutrient dense foods, such as hard white wheat, expands children's food preferences and fosters a healthful diet for children's growth and development (Skinner, Carruth, Bounds, & Ziegler, 2002).

Therefore, the primary purpose of this quantitative study was to determine whether repeated exposure of hard white wheat whole grain and hard red wheat whole grain bread and tortillas would increase children's liking and intake of whole grain wheat bread and tortillas. A secondary purpose was to determine whether children's liking and intake of bread and tortillas was greater with hard white wheat whole grain versus hard red wheat whole grain.

Methods

Children from child care centers in the Pacific Northwest were invited to participate in the study. Parents completed a demographic questionnaire. Trained researchers conducted hedonic tasting activities with samples of bread and tortillas prepared with hard white wheat whole grain flour and hard red wheat whole grain flour. The University of Idaho Institutional Review Board approved this study.

Cohorts. Children were recruited into two cohorts (n=101). Children in cohort 1 (n=48) participated in a baseline activity, 7 repeated exposure activities, a post activity, and a large portion size tasting activity. Children in cohort 2 (n=53) participated in a baseline activity and a large portion size tasting activity.

Center and participant recruitment. Purposive sampling was used to recruit participants (children 3-5 years of age) from Child Care Centers (CCC) in the Pacific Northwest for cohort 1 and 2. The Idaho State Training and Registry System (STARS), a public record of child care providers in Idaho, was used to identify Idaho CCCs. The Idaho STARS Region 2 Quality Child Care Consultant was contacted to facilitate communication with CCC directors. In the first cohort, a sample of children attending CCCs, in Moscow, ID and Lewiston, ID were identified to access participants. A second cohort of children attending

a CCC in Pullman, WA and the Child Development Center and the University of Idaho Children's Center in Moscow, ID were identified to access participants.

Recruitment began fall 2015, in which CCC directors were contacted to assist in the recruitment of parents and children. Multiple CCCs were contacted for participation in the study. A letter was sent to CCC directors explaining the details of the study and an invitation to participate (See appendix A). The initial contact was used to determine whether a CCC was willing to participate and whether their center met the inclusion criteria for participation. Inclusion criteria consisted of serving children between 3-5 years of age and located in the Pacific Northwest. Of the 10 centers contacted, 7 agreed to participate in the study. Child care centers serving children from a lower income status and limited consumption of 100% whole grain bread and/or tortilla products were sought out for participation in this study. Parental consent was obtained for each child participating in the study.

The primary investigator and research assistants met face-to-face with each CCC director, who responded to the letter, to further explain the study and establish rapport. Child care center directors (n=7) were given information about the tasting activity and a description of their involvement in the study as well as the expectation of the parent and child.

Using rolling enrollment, families in the initial cohort were asked to complete consent forms (see appendix B) and a demographic questionnaire (see appendix C). Children from Tendercare Children's Center (n=8) in Lewiston ID, from Wonder World Learning Center LLC (n=15) in Lewiston ID, Emmanuel Lutheran (n=27) in Moscow, ID and Moscow Day School (n=10) in Moscow ID participated in the first cohort. In the second cohort children were recruited from the University of Idaho Child Development Lab (n=17), The University

of Idaho Children's Center (n=18) in Moscow, ID, and The Learning Center (n=6) in Pullman, WA. Parents of children who completed the study received compensation of $20.

Demographic questionnaire. A questionnaire was developed and administered to parents to capture bread and tortilla purchasing and consumption habits by the adults and children in the family. The 22-item questionnaire (see appendix C) contained questions pertaining to adult consumption of bread and tortillas per day, frequency of whole grain bread and tortillas and non-whole grain bread and tortillas purchased, and the type of whole grain and non-whole grain bread and tortillas typically consumed by the parent. Parents were asked to provide demographic information such as the child's birth date, gender, whether the child had food allergies, their relationship to the child, the parent pregnancy status, the parent employment status, level of education, household income, ethnicity, marital status, and parent height and weight. Also, the frequency and amount of child consumption of bread and tortillas per day, and the type of whole grain (whole or non-whole grain) bread and tortillas typically consumed by the child were captured.

Bread preparation. All wheat products were prepared in the Food and Research Center Laboratory by a trained researcher. The wheat flour was donated by Ruben McLean, Manager Quality/Audit Compliance Grain Craft, and the yeast was donated by Katherine O'Brien from the University of Idaho Wheat Quality Lab. A bread recipe and protocol was developed by trained research assistants to create standardized bread to 100g bread pan loafs. A moisture test and mixing test was conducted, using the AACC method, at the Washington State University Wheat Quality Laboratory to determine the moisture content of the hard white wheat and hard red wheat breads for water standardization between type of wheat variety. The bread recipe was tested repeatedly to ensure evenness in baking, cell size, and

bread texture. Ingredients for bread were weighed to the nearest 100th of a gram to standardize bread loaves. The addition of food grade commercially available McCormic dye was used to match the color of hard white whole wheat to hard red whole wheat. A CR 400 Chroma Meter (Konica Minolta Sensing Americas, Inc. New Jersey, USA) was used to assess L*, a*, and b* color parameters, and matched (Smith, Deobald, Worden, Keeney, & Ramsay, 2016). Dye added to the dough was measured to the nearest 0.1 uL using a variable volume classic pipette. Four versions of bread recipes were developed: hard red wheat, hard white wheat, hard red wheat dyed, and hard white wheat dyed.

Tortilla preparation. Tortillas were prepared in the Food and Research Center Laboratory by a trained researcher. A tortilla recipe and protocol was developed by trained research assistants to create standardized tortillas. Tortillas were tested repeatedly to ensure evenness in thickness and diameter, as well as texture. Two versions of tortillas were developed: hard white wheat, and hard red wheat. Tortillas were baked on a skillet and pressed using a tortilla press. For complete tortilla recipe see appendix D.

Preparation of samples for tasting. Following baking, bread was transported to the Margret Richie School of Family and Consumer Sciences Carmelita Spencer Foods Laboratory for tasting activity portion sizing. Each loaf of bread was sliced to remove the crust. The bread was cut into ¼ inch slices and further cut into ¼ inch cubes. Three cubes were placed in a clear 1 oz cup with a lid. Each tortilla was prepared using a cookie cutter to remove the center of the tortilla, and a pizza cutter was used to cut the outside ring into ¼ inch triangles. Three triangles were placed in a clear 1 ounce cup with a lid. Circular stickers were numbered, color coded, and placed on the bottom of each cup and lid to indicate type of bread and tortilla. One cup of each type of bread and tortilla was stacked, and placed in a

clear bread bag for transport to tasting activities. All cups were covered and kept at room temperature. Bread and tortilla portion size protocols are provided in appendix E.

Taste activity setting. The taste activity setting was similar for cohort 1 and 2. Over the course of five months, trained graduate and undergraduate research assistants visited each CCC one to three times a week to conduct taste activities with each child. Children in cohort 1 participated in repeated exposure of bread and tortillas, while cohort 2 did not. The taste activities followed the validated protocol of capturing children's liking and preferences using a three-point hedonic face scale (faces developed by Dr. Samantha Ramsay, adapted from Birch, 1979; for full script, see appendix F). The taste activity followed the procedure described below.

Set up and preparation for tasting activity. Trained researchers visited each CCC with a mobile tasting activity kit including hunger dolls, 3-point hedonic face scale, pointer finger for the child to use, water pitcher, water cup, and pre-prepared bread and tortilla samples. Children were greeted by a research assistant and asked if they would like to participate in a tasting activity. Children were brought to a taste activity location and offered to pour themselves water and to taste one of each sample: hard white wheat tortilla, hard red wheat tortilla, hard white wheat bread dyed, hard red wheat bread, hard red wheat bread dyed, and hard white wheat bread.

The arrangement and place settings were consistent for each taste activity for all participants. The taste activity included one child sized table, and two chairs located in discrete locations. Tables and chairs were developmentally appropriate for child height and size. The tables were set up with a water pitcher, disposable drinking cups, and a disposable napkin. Lighting was similar in all child care centers. The three-point hedonic face scale was

arranged to face the child, and within reach of the child using the pointer finger. Tasting activities were performed in an area where distractions to the child being tested were minimal.

The tasting activity procedure was consistent for cohort 1 and 2. At the initial tasting activity, both cohorts of children were offered 3 ¼ inch cube/triangle samples of bread/tortillas per cup. The first cohort of children were presented with a tray with six disposable cups, each containing 3 ¼ inch cube/triangle samples of bread/tortillas per cup for repeated exposure. At the final tasting activity, both cohorts of children were offered larger samples: one inch slices of bread, equivalent to three slices of bread and 3/8 tortilla per serving. For all tasting activities, bread and tortilla samples were presented at room temperature.

Assent was obtained from each child prior to conducting each tasting activity by asking the child "would you like to help me with a project today?" Next, children were asked "Do you have any allergies?" To assess children's level of hunger and satiety at the time of each tasting activity, hunger dolls were used (See figure 3.1 and appendix F).

Figure 3.1 Hunger Dolls Used in Taste Preference Activities

FULL – Say to the child "you can point to this doll if your stomach is feeling full." 	**NEUTRAL** – Say to the child, "you can point to this doll if you are not really hungry, but you're not really full either." 	**HUNGRY** – Say to the child, "you can point to this doll if your stomach is feeling hungry." 

* Hunger dolls developed by Dr. Samantha Ramsay and Patti Perry

To assess hunger, each doll was presented to the child one at a time with an explanation "This is my friend, and she is really hungry. See how her tummy is all the way

empty? That means that she is very hungry. You can point to this doll if you are feeling very hungry," "This is my friend, and she is really full. See how her tummy is filled up all the way? That means that she is really full. You can point to this doll if you are feeling really full," "This is my friend and she is not really hungry, not really full. Can you see how her tummy is only filled halfway up? That means that she is not really hungry, not really full. You can point to this doll if you are feeling not really hungry, not really full". After all three hunger dolls were explained, the child was given a pointer finger, the hunger dolls were mixed up, and the child was asked to choose the hunger doll that matched the description: "Can you point to my friend that is really hungry?" "Can you point to my friend that is really full?" "Can you point to my friend that is not really hungry, not really full?" Children were then asked to point to the hunger doll that reflected their current level of hunger: "Can you point to the doll that tells me how your tummy is feeling right now?" After identifying their hunger, children returned the pointer finger to the research assistant and continued to the next portion of the taste preference activity.

At the beginning of the taste activity, children's level of understanding of the three-point hedonic face scale (see figure 3.2) was determined using a protocol developed by Leanne Birch (1979), modified by Dr. Susan Johnson, and modified for the current study by Dr. Samantha Ramsay (see appendix F). A trained research assistant closely followed a script, but did not read verbatim (for full script see appendix F).

Figure 3.2 Hedonic Faces Used in Taste Preference Activities

Yummy Just OK Yucky

*Faces developed by Dr. Samantha Ramsay, adapted from Birch, 1979.

Each face was presented to the child one at a time with a brief explanation: “This is the yummy face. See how she is smiling? That means she likes it and she thinks it’s yummy,” “This is the yucky face. See how she is frowning? That means she doesn’t like it and she thinks it’s yucky,” “This is the just ok face. See how she isn’t smiling or frowning? That means she thinks it’s just ok”. After all three faces were explained, they were placed on the table one at a time, with a second explanation: “This is the yummy face. See how she is smiling? You can point to this face if you try a food and you like it,” “This is the yucky face. See how she is frowning? You can point to this face if you try a food and you don’t like it,” “This is the just ok face. See how she isn’t smiling or frowning? You can point to this face if you try a food and you think it is just ok”. After each face was explained, the child was asked to choose the face that matched each description: “If you tried a food and you liked it, which face would you point to?” “If you tried a food and you didn’t like it, which face would you point to?” “If you tried a food and you thought it was just ok, which face would you point to?”. Children who demonstrated their understanding of the hedonic scale continued with the taste preference activity.

Conducting the tasting activity. Following the child's demonstrated understanding of the hunger dolls and hedonic faces, the tasting activity was conducted. The child was presented with a tray containing six samples of bread and tortillas and asked to take the first cup they would like to try. "Please pick up the cup, taste it, and place the cup in front of the face that tells me how it tastes to you." Once the child placed the cup in front of the face, they were asked "what face is that". The researcher recorded the rating, and order for that sample and repeated until each sample was rated. Once all samples were rated, any duplicates in a preference were ranked according to the child's report of which sample is the yummiest or yuckiest. All responses were recorded on an activity sheet (see appendix G), as well as the child's behavior: like, lick, spit, smell, swallow, and refuse.

Anthropometrics. Following baseline and final tasting activities, children's height and weight were obtained (See appendix G). Children were asked to remove their shoes, stand in front of a tape measure with their heels pressed against the wall and look straight ahead (CDC, 2014). The child was weighed using a portable scale to the nearest hundredth of a pound.

Plate waste as a proxy for intake. Plate waste in each sample cup was documented from baseline, at repeated exposure, and the large portion sample following completion of all tasting activities. Each sample was weighed to the nearest tenth of a gram prior to the activity and after the activity to estimate the difference in gram weight as a proxy for children's intake. To see the plate waste record sheet see appendix H.

Activity cleanup. Taste activity stations were cleaned between each activity. Tables and chairs were cleaned and sanitized with a water and bleach solution. Spilled bread and tortillas were removed from the area after each tasting activity. All cups, lids, water bottles,

and spoons were disposed of. Trays and faces were sanitized with a water and bleach solution towelette after each tasting activity. Following the completion of the tasting activity for that day, supplies were placed in the mobile kits for transport to the Margaret Richie School of Family and Consumer Sciences Eating Laboratory for storage.

PROP test. A PROP test was conducted to identify children who were bitter tasters. The PROP test solution was prepared in the Carmelita Spence Foods Lab at the University of Idaho, using distilled water (DI water) and a powder form of PROP to create a 1 liter batch with a concentration of a 0.56mmol/L. Each batch was separated into 10mL individual samples for the PROP test activity. Each child was presented with a 60mL plastic cup containing 10mL of the PROP solution and a cup of apple juice to drink after trying the solution. Children were asked to sip and spit out the PROP solution, then they were asked if they could taste anything. If the child said no they were classified as a non-taster. If the child said yes, or had a noticeable facial response they were either classified as semi-tasters, or bitter-tasters depending on the severity of their reaction. Children who were classified as bitter tasters, consumed apple juice after tasting the PROP solution to cleanse their pallet. Facial expressions were observed and documented to support verbal responses and to identify ambiguous responses, or conflicting responses. PROP test results did not influence children's eligibility to participate in the study. For full PROP protocol see appendix I.

Data analysis. Descriptive statistics were used to analyze questionnaire demographic data, including parents' bread and tortilla purchasing and consumption habits, and parents' daily consumption of bread and tortillas. The type and frequency of whole grain bread and tortillas and non-whole grain bread and tortillas purchased was collected. Descriptive statistics were used to analyze parents' self-reported height, weight, age, and Body Mass

Index (BMI). Means, median, range, and standard deviation were identified. Body mass index was calculated as kilograms of body weight divided by height in squared meters for each primary parent using self-reported height and weight (CDC, 2014). Descriptive statistics, counts, and frequencies were used to identify children's preference and intake of bread and tortillas at baseline and final activity.

Children's intake was reported to the nearest one-hundredth of a gram. Independent sample t-tests were used to identify difference in children's intake between cohort 1 and cohort 2 at baseline and final activities. The independent-samples Mann-Whitney U test was used to identify a difference in children's reported liking of all bread and tortilla samples between cohort 1 and cohort 2. Statistical significance was set at $p<0.05$ for all tests and SPSS Statistics (IBM Corp. Released 2011. IBM SPSS Statistics for Windows. Version 22. Armonk, NY: IBM Corp) was used to complete the analysis. Independent sample t-tests were used to determine an intake difference in hard white wheat versus hard red wheat between cohort 1 and cohort 2.

Results

One-hundred and one children participated in the study. The sample included primarily Caucasian children (n=91), and they were typically from a married household (n=91). The education level of the primary caregiver ranged from a high school degree/some college (n=18), a two year degree (n=4), a four year degree (n=49), and a graduate degree (n=30). Family income ranged from less than $35,000 (n=13), $35,000-$41,999 (n=3), $42,000-$51,999 (n=6), $52,000-$58,999 (n=8), $59,000-$73,999 (n=12), and $74,000 and above (n=57). The average Body Mass Index (BMI) was in the healthy range for adults (23.98±5.8). Full demographics are reported in table 3.1.

Table 3.1 Parent and Child Demographic Information

	N (%)
Relationship to Child	
Mother	94 (93)
Father	6 (6)
Other	1 (1)
Ethnicity	
Caucasian	91 (91)
Hispanic or Latino	3 (3)
Black or African American	1 (1)
American Indian or Alaskan Native	1 (1)
Asian or Asian American	3 (3)
Other	1 (1)
Marital Status	
Single	7 (7)
Divorced	1 (1)
Separated	1 (1)
Married or in Committed Relationship	91 (91)
Employment Status	
Chose not to work	18 (18)
Part-time	22 (22)
Full-time	53 (53)
Other	8 (8)
Highest Level of Education	
High School degree/Some College	18 (18)
Two year degree	4 (4)
Four year degree	49 (49)
Graduate degree	30 (30)
Household Income	
Less than $35,000	13 (13)
$35,000-$41,999	3 (3)
$42,000-$51,999	6 (6)
$52,000-$58,999	8 (8)
$59,000-$73,999	12 (12)
Greater than $74,000	57 (57)
Child Gender	
Male	53 (52)
Female	48 (48)
Child Age	
2 years	1 (1)
3 years	22 (22)
4 years	35 (34.5)
5 years	39 (38.5)
6 years	4 (4)

Children (n=58) participated in the PROP test to determine sensitivity to bitter tastes (on a three-point scale: bitter-taster, semi-bitter-taster, and non-bitter-taster). The majority of children, 43% (n=25), were bitter tasters, while 24% (n=14) were semi-tasters, and 32% (n=19) were non-tasters.

Bread and tortilla consumption and purchasing habits. As indicated from the questionnaire that captured bread consumption patterns, almost all parents (n=75) and children (n=81) consumed one or more servings of bread products daily with few reported allergies or intolerances (n=15). Bread was reported as typically consumed at breakfast (n=59), lunch (n=75), and dinner (n=67) by both parents and children; however the primary parent reported the lunch meals as having the highest consumption of bread by children (n= 81). Tortillas were reported as less frequently consumed, with both parent and child consumption at breakfast (n=2) and lunch (n=11) relatively low, but the reported most frequent meal that tortillas were consumed was the dinner meal (n=34). Sixty-six percent (n=66) of parents and children consumed whole grain breads, however 22% (n=22) reported consuming wheat or white (not knowing the difference between wheat and whole wheat). Both parents and the children consumed far less whole wheat tortillas; the majority (82%, n=82 and 86%, n=86) of parents and children respectively, consumed tortillas made from non-whole grain flour.

Children's reported hunger during tasting activities. Prior to conducting each tasting activity, children's level of hunger was assessed. The hunger scale ranged from full, neutral, and hungry. Hunger was included as a cofactor in an ANOVA to identify whether a difference in intake was present among bread and tortilla samples at baseline and final activities. A Tukey Post Hoc test indicated no statistical difference ($p>.05$) in children's

intake of bread and tortillas compared to hunger level for cohort 1 and cohort 2 at baseline and final activities. For additional information see table 3.2.

Table 3.2 Children's Reported Hunger of Full, Neutral, and Hungry During Taste Preference Activities

	Baseline N (%)			Final N (%)		
	Full	**Neutral**	**Hungry**	**Full**	**Neutral**	**Hungry**
Cohort 1	20 (43)	10 (21)	17 (36)	19 (41)	13 (28)	14 (30)
Cohort 2	17 (34)	13 (26)	20 (40)	18 (34)	19 (36)	16 (30)

* Tukey Post Hoc test indicated no difference $p \leq .05$

Preference for hard white wheat versus hard red wheat bread and tortillas.

Children's preference (liking) of bread and tortillas was determined. During the baseline tasting activity children rated the HWT (n=54) as "yummy" most frequently, followed by HWB (n=53), HRB (n=52), HWBD (n=51), HRBD (n=49), and HRT (n=39). The wheat sample most frequently reported as "yucky" was HRT (n=22). Of the two dyed bread samples, HWBD (n=51) was rated "yummy" more frequently than HRBD (n=49), and reported as "yucky" less frequently (n=4 and n=11, respectively). During the final tasting activity children rated the HWT (n=62) as "yummy" most frequently, followed by HWBD (n=53), HRT (n=51), HWB (n=50), HRB (n=48), and HRBD (n=42). HRT (n=17) was reported as "yucky" most frequently, followed by HRBD (n=15). See table 3.3 for additional information.

Table 3.3 Children's Reported Liking of Wheat Samples

	Baseline		Final	
	Yummy	**Yucky**	**Yummy**	**Yucky**
	N (%)		N (%)	
HWT	54 (18)	11 (15.5)	62(20)	8 (11)
HRT	39 (13)	22 (31)	51 (17)	17 (23)
HWDB	51 (17)	4 (0.5)	53 (17)	8 (11.5)
HRB	52 (17.5)	8 (11)	48 (16)	11 (15)
HRDB	49 (16.5)	11 (15.5)	42 (14)	15 (20.5)
HWB	53 (18)	15 (21)	50 (16)	14 (19)

*HWT= Hard white tortilla, HRT= hard red tortilla, HWDB= hard white dyed bread, HRB= hard red bread, HRDB= hard red dyed bread, HWB= hard white bread

Repeated exposure resulted in a significantly higher rating of HWB (p=0.042). Sixty-three percent (n=30) of children who had repeated exposure to all samples rated HWB as "yummy" compared to forty-two percent (n=20) of children who did not have repeated exposure. The most frequent reported rating of HWB for cohort 1 was "yummy" (n=30), while cohort 2 rated HWB as "yummy" (n=20) and "just ok" (n=19). For additional information see table 3.4.

Table 3.4 Children's Reported Liking of Wheat Samples Using Hedonic Scale at Baseline and Final by Cohort

		Cohort 1 N (%)			Cohort 2 N (%)		
		Yummy	**Just Ok**	**Yucky**	**Yummy**	**Just Ok**	**Yucky**
Baseline							
	HWT	25 (64)	10 (26)	4 (10)	29 (57)	15 (29)	7 (14)
	HRT	15 (38)	12 (30)	13 (32)	27 (54)	11 (22)	12 (24)
	HWDB	26 (62)	16 (38)	0 (0)	25 (51)	20 (42)	4 (8)
	HRB	23 (59)	12 (31)	4 (10)	29 (60.5)	15 (31)	4 (8.5)
	HRDB	22 (58)	11 (29)	5 (13)	27 (55)	16 (33)	6 (12)
	HWB	27 (69)	7 (18)	5 (13)	26 (53)	13 (25.5)	10 (20.5)
Final							
	HWT	33 (69)	12 (25)	3 (6)	29 (60.5)	14 (29)	5 (10.5)
	HRT	26 (54)	13 (27)	9 (19)	25 (51)	16 (33)	8 (16)
	HWDB	26 (54)	19 (40)	3 (6)	27 (59)	14 (30)	5 (11)
	HRB	26 (54)	18 (38)	4 (8)	22 (46)	19 (40)	7 (14)
	HRDB	20 (42)	19 (40)	9 (19)	22 (47)	19 (40)	6 (13)
	HWB**	30 (63)	13 (27)	5 (10)	20 (42)	19 (39)	9 (19)

*HWT= Hard white tortilla, HRT= hard red tortilla, HWDB= hard white dyed bread, HRB= hard red bread, HRDB= hard red dyed bread, HWB= hard white bread
**Significant difference p≤.05 using Independent-samples Mann-Whitney U test

Intake for hard white wheat versus hard red wheat bread and tortillas.

Children's intake of bread and tortillas was collected. During the baseline tasting activity, the average consumption for cohort 1 was greatest for HWBD (μ=0.77 g) and least for HRT (μ=0.38 g). The average consumption for cohort 2 was greatest for HRBD (p=0.157, μ=0.73

g) and least for HRT (p=0.55, μ=0.34 g). During the final tasting activity, cohort 1 consumed the greatest amount of HWB (μ=8.8 g) and the least amount of HRB (μ=6.9 g). The average consumption for cohort 2 at the final tasting activity was greatest for HRBD (μ=5.1 g) and least for HWT (μ=3.7 g). There was no significant difference in intake between hard white wheat and hard red wheat bread and tortillas.

Independent sample t-tests were used to identify a difference in children's intake between cohort 1 and cohort 2 at final large portion activity. Children who were repeatedly exposed to wheat samples consumed significantly greater ($p \leq .05$) gram amount of bread and tortillas for all samples than children who did not receive repeated exposure (see table 3.5).

Table 3.5 Children's Baseline Intake of Bread and Tortillas at Baseline and Final by Cohort

		Mean (SD)				
		Cohort 1 Repeated Exposure	Cohort 2			
Baseline				**t**	**df**	**p-value**
	HWT	0.38 (0.39)	0.39 (0.35)	-0.21	99	0.833
	HRT	0.39 (0.41)	0.34 (0.27)	0.59	99	0.555
	HWBD	0.77 (0.78)	0.70 (0.54)	0.50	99	0.613
	HRB	0.60 (0.65)	0.72 (0.55)	-0.95	99	0.342
	HRBD	0.57 (0.60)	0.73 (0.53)	-1.4	99	0.157
	HWB	0.64 (0.67)	0.70 (0.49)	-0.52	99	0.601
Final						
	HWT	8.1 (6.4)	3.7 (4.6)	4.03	99	0.000*
	HRT	7.7 (5.6)	4.5 (5.1)	3.00	99	0.003*
	HWBD	8.4 (5.7)	4.7 (5.0)	3.47	99	0.001*
	HRB	6.9 (5.5)	4. (4.6)	2.08	99	0.040*
	HRBD	7.9 (5.8)	5.1 (5.2)	2.58	99	0.011*
	HWB	8.8 (5.2)	4.6 (4.9)	4.07	99	0.000*

*HWT= Hard white tortilla, HRT= hard red tortilla, HWDB= hard white dyed bread, HRB= hard red bread, HRDB= hard red dyed bread, HWB= hard white bread
* Significant difference $p \leq .05$

Discussion

Results from this study demonstrated the effect of repeated exposure to significantly increase children's consumption of all types of whole grain bread and tortillas regardless of color and taste differences. Children who had repeated exposure to bread and tortillas, consumed greater amounts of hard white wheat compared to hard red wheat, but the difference was not statistically significant. Children reported a significantly more frequent preference for hard white wheat bread compared to hard red wheat bread, as indicated by a higher rating of "yummy". Additionally, those children who were bitter tasters, as indicated by the PROP test, consumed significantly greater amounts of hard white wheat tortillas. Reported hunger did not impact intake of hard white wheat and hard red wheat bread and tortillas.

The results from this study support previous research on the benefit of repeated exposure to support intake of foods, (particularly whole grain wheat) in young children. Children who participated in repeated exposure of hard white wheat and hard red wheat consumed significantly greater amounts of all samples of whole wheat bread and tortillas. As repeated exposure fosters children's taste preferences (Birch, 1979; Birch & Fisher, 1998; Rozin, 1990, Wardel et al, 2003), the implications of repeated exposure to support whole grain consumption is of value in consideration of the national Dietary Guideline recommendations to consume half of grains as whole grains (Birch, 1979; Birch & Fisher, 1998; Rozin, 1990, Wardel et al, 2003).

Previous research identified children had a significant difference in consumption of hard white wheat compared to hard red wheat, but these preferences of hard white and hard red wheat bread and tortillas has been less frequently studied (Keeney, Ramsay, Tsao, &

Planck, 2016). Thus the current study aimed to determine whether a difference in preference existed since whole grains, such as hard red wheat, contain a greater amount of phenolic compounds, which creates a more apparent bitter taste than hard white wheat (Ransom et al., 2006). Results from the present study indicated children's gram consumption of hard white wheat was greater than their consumption of hard red wheat in both bread and tortillas, however this difference was not statically significant. Children inherently prefer sweet and salty tastes compared to bitter and sour (Steiner, 1979), which would explain the greater intake of HWW and reported liking.

In this study children were tested for bitter sensitivity. Children who were bitter tasters consumed significantly greater amounts of hard white tortillas. Children generally have more sensitive taste responses compared to others (Birch, 1987; Birch & Fisher, 1998) and children who have even greater bitter sensitivity may have more aversive responses to certain foods (Birch & Dietz, 2008), such as whole wheat bread as indicated in the present study. Hard white wheat may create an opportunity to increase whole grain consumption and liking in children who have more taste sensitivity (such as bitter tasters) due to the less stringent taste and lighter color that is more palatable for children (Steiner, 1979).

Children have a variability in food intake, thus hunger was assessed prior to collecting intake at baseline and final tasting activities. Results from this study did not indicate hunger as an influence on children's consumption of bread and tortillas. Children tend to consume more palatable foods regardless of hunger (Birch, 1987). For example, children consume bread more frequently versus foods with less palatability such as vegetables, wherein hunger could have a greater impact (Birch & Fisher, 1998). Research using less palatable foods may benefit by assessing hunger.

Implications for Future Studies & Practice

Children generally liked and consumed all bread samples, but confirming previous research, generally children's intake for HWW bread and tortillas was higher than HRW bread and tortillas. From the questionnaire, caregivers reported offering bread as predominantly whole grain (from hard red wheat), which may have influenced children's frequent reported liking of HRB and the dyed HWW and HRW breads due to their familiarity of whole grain bread color. Regardless of parent reported offering of whole grain bread and tortillas, repeated exposure increased children's intake of all bread and tortilla whole grain samples.

Children do not consume the recommended amount of whole grains, and their consumption of whole grains impacts nutrient intake; therefore strategies to increase whole grain intake are needed. Results from this study reinforce the use of repeated exposure as a way to increase whole grain consumption in children from both HWW and HRW bread and tortillas. In addition, children who are bitter sensitive could benefit from products made with HWW compared to HRW as a means to increase their whole grain consumption. The effects of repeated exposure increasing all wheat sample intake, as well as the potential use of hard white wheat to increase intake is valuable information for parents, child care settings, and school lunch programs who feed children. To support Dietary Guideline efforts, marketing and dissemination of these results is needed to include: 1) the benefit of whole wheat grains, 2) awareness of the hard white wheat whole grain 3) the benefit to offering hard white wheat whole grain to increase children consumption of whole grains, and 4) the effect of repeated exposure to increase children's intake of whole grain wheat products.

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Appendix A

Dear Director:
Thank you for your willingness and effort to support our bread study. We can be reached by email idahowheatstudy@outlook.com and phone (520) 468-9054 at any time when a parent might have a question that we could answer for.

You will find the following materials in this package:

- The parent consent form with an introduction letter.
- The demographic info package for parents who have submitted their consent form.

We would like you to distribute the package to all parents of children ranged 3 to 5 years old.

We will ask parents to sign and fill the demographic info and return the package back to you and/or your teachers.

We will be back to pick up the signed form and the demographic info by October 23rd.

Thank you for your assistance.
Sincerely,

Samantha Ramsay, PhD, RDN, LD
Assistant Professor
University of Idaho

Samantha Worden, BS, MS Candidate
Research Assistant
University of Idaho

Ling-Ling Tsao, PhD
Assistant Professor
Family and Consumer Sciences
University of Idaho

Lauren Keeney, BS, MS Candidate
Research Assistant
University of Idaho

Appendix B

University of Idaho

CONSENT FORM

"Use and Preference of Bread Products"

Researcher: Samantha Ramsay, Assistant Professor, School of Family & Consumer Sciences, University of Idaho- Phone: 208-885-6026

PURPOSE AND BENEFITS

You and your child have been selected to participate in a study to understand your use of bread products. This study has been approved by the University of Idaho Institutional Review Board. You will be given a copy of this form for your records. The information we gain from this study will help us further our understanding of child and parent use and preference for bread products.

PROCEDURES

The study will consist of two main activities.

- Completion of a questionnaire at the beginning of the study as well as a follow up questionnaire at the end of the study.
- Your child's participation in taste preference activity of bread products no more than 8 times during the study. The child interview and taste preference activities will be audio and video recorded.

RISKS, STRESS, OR DISCOMFORT

The identities of the participants are confidential and pseudo names for you and your child will be used on all data, aside from the questionnaires. If at any time you are uncomfortable with a question, activity, or the snack you may discontinue the activity. All audio recording will be transcribed without the names of participants. A total of 6 samples of wheat products will be offered. Two of the bread samples will contain a commercial food dye to match color.

In compensation for your and your child's participation in the full research study, you will receive a **$20 gift card** and your child will continue to be provided with a free bread snack at each observation. The taste preference activities and observations will occur mid-October through the spring semester.

If you have any questions about this study now or later, please call Dr. Samantha Ramsay. She can be reached at 208-885-6026. You can also email her at idahowheatstudy@outlook.com

Participant's Statement

This study has been explained to me. I volunteer and agree to allow my child to take part in this research. I have had a chance to ask questions. If I have general questions about the research, I can ask the researcher listed above. If I have questions regarding my rights as a participant, I can call the University of Idaho Institutional Review Board at (208)885-6162. This project has been reviewed and approved for human participation by the Human Assurances Committee at the University of Idaho IRB. I will receive a copy of this consent form.

I _____, agree to participate in the study, and I agree for my child,
Please Print

_____, to participate in the study.
Please Print

Your Signature **Date**

Samantha Ramsay, PhD, RDN, LD
University of Idaho, Family and Consumer Sciences
PO Box 443183,
Moscow, ID 83844
Ph. 208-885-6026

Samantha A. Ramsay Samantha Ramsay October 9, 2015

Printed name of researcher **Signature of researcher** **Date**

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Appendix C

CONSUMPTION OF GRAIN PRODUCTS

Thank you for participating in our study! We would like to learn about your consumption of bread. Bread is defined as a baked food made from a mixture of flour and water; purchased or homemade.

We define bread products as: bread slices, rolls, bagels, English muffins, hamburger or hotdog buns, etc. We define tortillas as a round unleavened bread that is flat, thin and made of corn or flour.

The USDA's Choose My Plate defines a serving size of bread as one ounce (i.e. one slice of bread, one small roll, ¼ of a large bagel or one 'mini' bagel, half an English muffin, half a hamburger or hotdog bun). The USDA's Choose My Plate defines a serving of tortillas as one eight inch tortilla.

Please answer the following questions by placing the number of servings of bread products (described above) you or your child consume for each meal or snack. If you or your child do not consume that meal or snack, write "0". If you have more than one child who is between the ages of 3-5 years, please select the child who will be participating in the study.

1. **In a typical <u>DAY</u>, how many servings of bread products do you consume for:**

   Breakfast _____servings
   Morning Snack _____servings
   Lunch _____servings
   Afternoon Snack _____servings
   Dinner _____servings
   Evening Snack _____servings

2. **In a typical <u>DAY</u>, how many servings of bread products does your <u>*child*</u> consume for:**

   Breakfast _____servings
   Morning Snack _____servings
   Lunch _____servings
   Afternoon Snack _____servings
   Dinner _____servings
   Evening Snack _____servings

3. **What type of grain do you <u>typically</u> select in the bread products <u>you</u> eat? (Mark one)**

   O White
   O Wheat
   O Whole wheat
   O I don't know
   O Do not eat bread products

4. **What type of grain do you <u>typically</u> select in the bread products your ***<u>child</u>*** eats? (Mark one)**

   O White
   O Wheat
   O Whole wheat
   O I don't know
   O Do not eat bread products

5. **For the following questions, please check the box that applies: (Mark one)**

	Never	1/Month	1/Week	Multiple times/week
How often do you purchase whole grain bread?	O	O	O	O
How often do you purchase non-whole grain bread?	O	O	O	O

Please answer the following questions by placing the number of servings of tortilla products (described above) you or your child consume for each meal or snack. If you or your child do not consume that meal or snack, write "0". If you have more than one child who is between the ages of 3-5 years, please select the child who will be participating in the study.

6. **In a typical <u>DAY</u>, how many servings of tortilla products do you consume for:**

   Breakfast _____servings
   Morning Snack _____servings
   Lunch _____servings
   Afternoon Snack _____servings
   Dinner _____servings
   Evening Snack _____servings

7. In a typical <u>DAY</u>, how many servings of tortilla products does your *<u>child</u>* consume for:

Breakfast _____servings

Morning Snack _____servings

Lunch _____servings

Afternoon Snack _____servings

Dinner _____servings

Evening Snack _____servings

8. What type of grain do you <u>typically</u> select in the tortilla products <u>you</u> eat? (Mark one)

O White
O Wheat
O Whole wheat
O I don't know
O Do not eat tortilla products

9. What type of grain do you <u>typically</u> select in the tortilla products your *<u>child</u>* eats? (Mark one)

O White
O Wheat
O Whole wheat
O I don't know
O Do not eat tortilla products

10. For the following questions, please check the box that applies: (Mark one)

	Never	**1/Month**	**1/Week**	**Multiple times/week**
How often do you purchase whole grain tortillas?	O	O	O	O
How often do you purchase non-whole grain tortillas?	O	O	O	O

Child's Information

11. What is your child's birthday (MM/DD/YYYY)

_____

12. What is your child's gender? (Mark one)

O Male
O Female

13. Does your child have food allergies or food intolerances? (Mark one)

O No
O Yes

If yes please specify in the box below.

14. What is your relationship to this child? (Mark one)

O Mother
O Father
O Grandparent
O Childcare provider
O Other (please specify_____)

15. Are you currently pregnant?

O Yes
O No

Your Information

16. What is your current employment status? (Mark one)

O I chose not to work in order to be home with my child/children
O Unemployed and looking for work
O I work part-time
O I worked full-time
O Other, please tell us _____

17. What is your highest level of education? (Mark one)

- O Some high school
- O High school diploma or GED
- O Some college
- O 2 year degree (Associate's degree)
- O 4 year degree (Bachelor's degree)
- O Graduate Master's Degree
- O Graduate Doctoral Degree

18. What is your total household income? (Mark one)

- O Less than $35,000/year
- O $35,000 - $41,999/year
- O $42,000 - $51,999/year
- O $52,000 - $58,999/year
- O $59,000 - $73,999/year
- O Over $74,000/ year

19. Which group best describes you? (Mark one)

- O White
- O Hispanic or Latino
- O Black or African American
- O American Indian or Alaska Native
- O Asian or Asian American
- O Hawaiian or North Pacific Islander
- O Other (please specify): _____

20. What is your current marital status? (Mark one)

- O Single
- O Divorced
- O Separated
- O Married or in a committed relationship

21. What is your current weight? _____ Pounds *or* _____ kg
(if you are pregnant, was your pre-pregnancy weight)

22. What is your height? _____ Feet _____ Inches *or* _____ cm

Thank you for completing the questionnaire!! Please contact Dr. Ramsay if you have questions

Appendix D

Tortilla Recipe

Ingredients:
Flour 100 150 200
Salt, non-iodized 1.5 2.25 3
Baking powder 2 3 4
Shortening 7 10.5 14
Water, warm 56 84 112

Instructions:

1. Press 320oF, 2 sec, currently using thinnest setting
2. Bake ~350oF 50s 1st side, 40s 2nd side, 20s/side, 12s/side
3. Mix shortening into dry ingredients prior to adding water
4. 4 minute knead (2 min with paddle), 5 minute RT rest in bag
5. Cut into 40g dough balls
6. 30 minute proof in cabinet, then press & bake

Appendix E

Bread and Tortilla Preparation Protocol

Bread can be prepared one day (up to 36 hrs) prior to the tasting activity. Tasting activities take place M, T, W, F from 9:00 am- 2:00 pm. Bread and tortilla samples must be ready on M, T, W at 8:00 am to accommodate travel to Lewiston.

Estimated time: Forty-five minutes to one hour

Supplies:

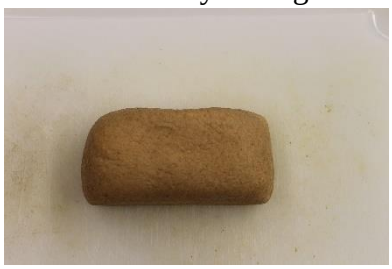
- Bread knife (serrated, silver handle)
- 6 cutting boards
- 1 circle cookie cutter
- 1 pizza cutter
- 2 hard white wheat tortillas
- 2 hard red wheat tortillas
- 1 loaf hard white wheat bread dyed
- 1 loaf hard red wheat bread
- 1 loaf hard red wheat bread dyed
- 1 loaf hard white wheat bread
- Food Storage & Bread Bag (1 gallon)
- 2 oz. cups, labeled
- Wheat 1 bin
- Wheat 2 bin

Instructions:

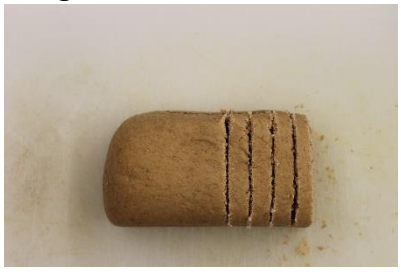
1. Place each type of bread and/or tortilla on a separate cutting board.
2. <u>To cut tortillas:</u> Place one hard white wheat tortilla on the cutting board. Punch out the center of the tortilla using the circle cookie cutter.

3. Place center to the side. Using the pizza cutter, slice tortilla (ring shape, with discarded center) into ¼ inch triangles.

4. Repeat until entire ring of tortilla is cut into uniform ¼ inch triangles.
5. Repeat process for second hard white wheat tortilla. On a separate cutting board, repeat for both hard red wheat tortillas.
6. <u>To cut bread:</u> Place hard white wheat bread dyed length wise on the cutting board.



7. From left to right, slice bread using bread knife into ¼ inch slices.



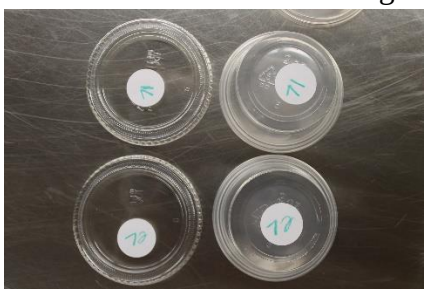
8. Discard ends of bread.

9. Cut crust (all sides) off of each slice of bread.

10. Cut each slice into 3 or 4 slices (¼ inch) length wise and 4 slices (¼ inch) width wise. This will create 12 to 16 ¼ inch cubes per slice. Repeat until entire loaf is in ¼ inch cubes.

11. Repeat for each type of bread, on separate cutting board.

12. <u>To portion sample cups:</u> Using pre-labeled cups and lids (see cup preparation protocol), place three ¼ inch hard white wheat triangles in each cup (with lid).



13. Continue until all hard white wheat tortillas are in cups.
    a. Blue- hard white wheat bread
    b. Pink- hard red wheat bread dyed
    c. Red- hard red wheat bread
    d. Teal- hard white wheat bread dyed
    e. Forest green- hard red wheat tortilla
    f. Bright green- hard white wheat tortilla

14. Repeat for hard red wheat tortillas.
15. Using pre-labeled cups and lids (see cup preparation protocol), place three ¼ inch hard white wheat bread dyed in each cup. Continue until all hard red wheat bread dyed cubes are in cups.

16. Repeat for hard red wheat bread, hard white wheat bread dyed, and hard white wheat bread.
17. Confirm correct placement of bread into correct cups.

18. <u>To organize cups for testing:</u> Stack all number 1 cups in the following order from bottom to top:

    a. Blue- hard white wheat bread
    b. Pink- hard red wheat bread dyed
    c. Red- hard red wheat bread
    d. Teal- hard white wheat bread dyed
    e. Forest green- hard red wheat tortilla
    f. Bright green- hard white wheat tortilla

19. Place the stack containing all of the same number of 6 different colors into a bread bag. Tie top of bread bag in lose knot. Place into Wheat bin 1.
20. Repeat for all numbered cups.
21. Place no more than 20 pre-labeled sample cups into Wheat bin 1.
22. Place no more than 20 pre-labeled and portioned sample cups into Wheat bin 2.
23. Run all supplies through dishwasher and clean work station.
24. Palace cookie cutter, pizza cutter, and bread knife (with sliver handle) in the bin at the back station
25. Thank you!

*Bread and tortilla preparation protocol was used for preparing bread and tortillas for cohorts 1 & 2.

**For larger portion size tasting activities, a larger container was used.

Appendix F

Tasting Protocol

Tasting Protocol (Really Yummy, Yummy, Just okay, Yucky, Really Yucky)

List of Study Foods to be purchased:

1. **Hard White Tortilla**
2. **Hard Red Tortilla**
3. **Hard White Dyed**
4. **Hard Red Wheat**
5. **Hard Red Dyed**
6. **Hard White Wheat**

Food items needed and details about specifics for purchasing:

Purchasing Food: All wheat flour was donated by Reuben McLean, Manager Quality/Audit Compliance, Grain Craft, 463 W. Hwy 26 Blackfoot, ID 83221, Ph: 208-785-2800 ext. 17, rmclean@graincraft.com, all yeast was donated by researcher Katherine O'Brian with the University of Idaho Wheat Quality Lab, katho@uidaho.edu, and all other ingredients were purchased from Safeway grocery market. All tortillas were donated by Gary Hou, Technical Director and Wheat Foods Specialist, Wheat Marketing Center, Inc., 1200 NW Naito Parkway, Suite 230 Portland, OR 97209, Ph: 503-295-0823, Fax 503-295-2735, ghou@wmcinc.org.

Preparing Bread: The crust from each loaf of bread was removed. All bread was cut into ¼ inch cubes to prevent choking. Three ¼ inch cubes were included in each cup. All cups should be covered and kept at room temperature at all times and placed on a tray as needed with each child.

Preparing Tortillas: All tortillas were cut into ¼ inch triangles to prevent choking. Three ¼ inch triangles need to be included in each cup. All cups should be covered and kept at room temperature at all times and placed on a tray as needed with each child.

Equipment:

Tasting Bags (1 per tester and 1 extra)

- Faces in stands (Yummy, Just Okay, Yucky)
- Pitcher of water (1/table)
- Small cup for Water
- Clip Board (1)
- Pen writing in indelible ink

- Stickers
- Hand Sanitizer
- Small spoons
- Assessment forms (paper)
- Protocol

Additional Equipment and Set-up Kit

- Assessment tablet for electronic data collection
- Trays - small (1 per tester)
- Gloves
- Paper towels
- Spray bottle of bleach and water solution (2 tablespoons of bleach diluted in 32 oz of water) or sanitation wipes.
- Trash can
- Cups (4) the number of study preferences (i.e. if you are testing 6 participants you would need 24 cups and lids)
- Lids (4 the number of taste preferences)
- Permanent Marker Sharpie
- Child tables (1) and chairs (2)

Setting up the testing environment:

Tasting should be preformed in an area where distractions to the child being tested are minimal. Use tables and chairs that are friendly and comfortable to the children and on their level.

Setting up the dining room:

The tasting activity will take place at different child care facilities in the greater Palouse Region & Inland Northwest. At each taste activity, one child sized table, and two chairs will be arranged in the room. All furniture arrangement and place settings will be consistent for each taste activity for all participants.

A. The child size table will be arranged in the same position (see figure 1).
B. The child size chairs in the same position (see figure 1)
C. Camera will be placed so that child's reactions and facial expressions can be captured.
D. Set up: napkins, spoon, water pitcher and small water cup(see figure3)

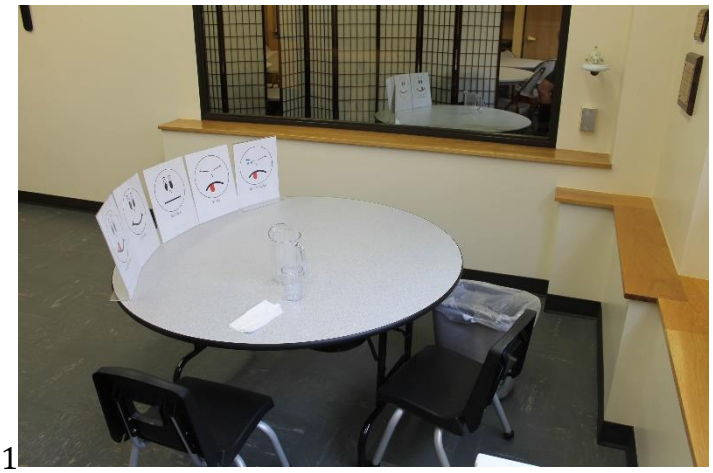


Figure 1

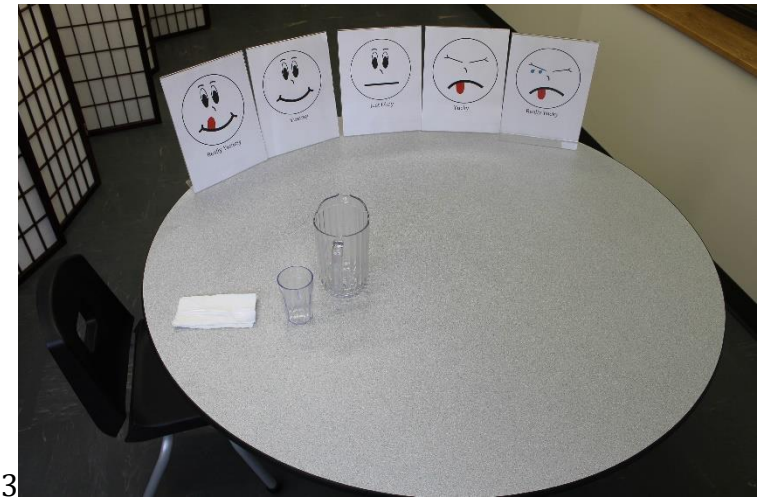


Figure 3

Item	Portion size	Number of portions on serving cup
Hard White Tortilla	¼ triangles	3
Hard Red Tortilla	¼ triangles	3
Hard White Dyed	¼ cubes	3
Hard Red Wheat	¼ cubes	3
Hard Red Dyed	¼ cubes	3
Hard White Wheat	¼ cubes	3

Presentation:

The tables will be set up with a water pitcher, disposable drinking cups, disposable napkins and spoon. The child will be presented with a tray with four disposable cups, each containing three ¼ inch cubes/triangles of each type of bread/tortilla per cup.

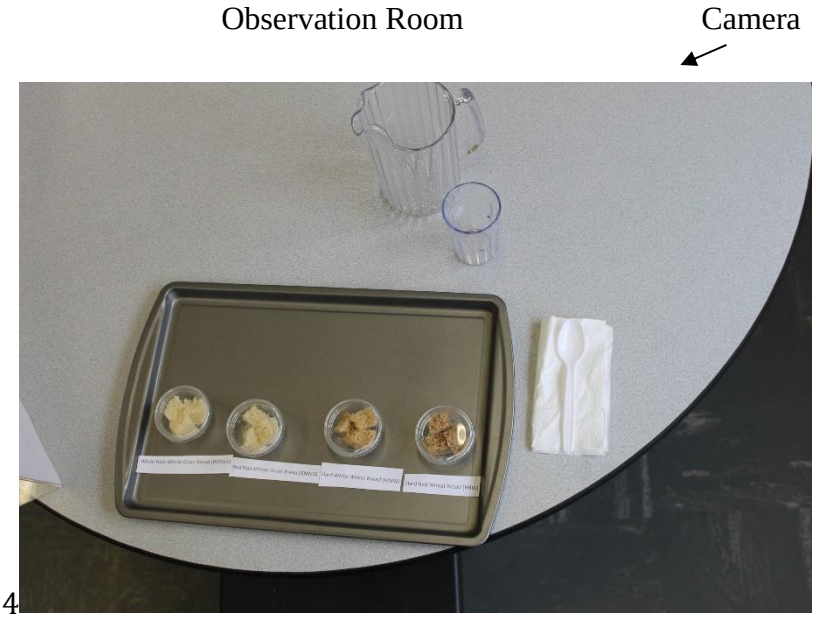


Figure 4

(**From left to right: White Non-Whole Grain, Red Non-Whole Grain, Hard White Wheat, Hard Red Wheat**)

Serving the Snack:

A. Portion control in serving dishes. Portion control of bread for child will include three ¼ inch cubes weighing approximately XXXX.
   a. *Weigh each 1/4 inch bread cube*. Prior to presenting bread in the dining area, the bread cubes in each serving dish will be weighed and recorded.
   b. Record data on the spreadsheet and tablet
B. Portion control in serving dishes. Portion control of tortilla for child will include three ¼ inch triangle weighing approximately XXXX.
C. *Weigh each 1/4 inch bread cube and tortilla triangle*. Prior to presenting bread in the dining area, the bread cubes and tortilla triangle in each serving dish will be weighed and recorded.
D. Record data on the spreadsheet and tablet
E. Place all cups in the same order on the serving cart (see figure 4).
   a. Serve food by bringing the serving cart into the testing area and placing the cups on the table in the SAME ORDER (left to right: HWW, HRW, HWT, HRT) and SAME LOCATION (see figure 4).
   b. Each cup of bread/tortilla will be presented in the same order for each child (left to right: HWW, HRW, HWT, HRT).
F. Conduct Snack Activity

<u>Snack Activity</u>:

Child Code:_____ Date:_____ Time of snack: _____
Exposure #:_____

Ask *"Would you like to help us with the project today?"*
Child assent given? YES ☐ NO ☐

- **Obtain child assent. If the child does not give assent, reschedule for another day.**

Offer the child water and say:

"You can pour yourself some water if you'd like." (point to pitch of water and cup)

Does the child have any allergies?: (circle) YES NO

- **ALWAYS CHECK FOR ALLERGIES BEFORE YOU BEGIN TESTING A CHILD!**
- Hunger Dolls give children a visual to assess their own hunger. Huger is a cofactor for how much bread/tortillas a child consumes.

Hunger Dolls

FULL	NEUTRAL	HUNGRY
		

Match the sex of the doll with the sex of the child i.e. his/hers

Tell the child:

"*I have some friends I'd like you to meet. Do you want to meet them?*"

Pull out the hunger dolls one at a time, beginning with the hungry hunger doll, followed by neutral hunger doll and end of full hunger doll. Say:

"This one is very hungry. See how his/her tummy is very empty? That means he/she is very hungry."

"This one is not really hungry and not really full. See how his/her tummy is not really full and not really empty? That means that he/she is not really hungry and not really full."

"This one is really full. See how his/her tummy is filled all the way up? That means he/she is really full."

Check for understanding: Mix the dolls up and ask:

- Hand the pointer finger to the child.

"Can you show me which one of my friends is really hungry?"
Wait for the child to point to the hungry doll.
"Yes, you can point to this doll if your tummy is feeling really hungry."

"Can you show me which one of my friends in not really hungry and not really full?"
Wait for the child to point to the neutral doll.
"Yes, you can point to this doll if your tummy is not really hungry and not really full."

"Can you show me which one of my friends is really full?"
Wait for the child to point to the full doll.
"Yes, you can point to this doll if your tummy feeling really full."

Wait for the child to correctly identify each doll, and then ask:
"Can you tell me how hungry you're feeling right now?"

- Take the pointer finger from the child.

- Each child is asked to rate their preference for 3-bite samples of four types of bread. The bread items will include:

Foods

1. Hard White Tortilla
2. Hard Red Tortilla
3. Hard White Dyed
4. Hard Red Wheat
5. Hard Red Dyed
6. Hard White Wheat

- **ALWAYS CHECK FOR ALLERGIES BEFORE YOU BEGIN TESTING A CHILD!**

- The containers of all samples will be placed on a tray table out of the child's line of sight to prevent distraction.

  Tell the child:

"I'd like to ask you to play the tasting game. In the tasting game, you get to taste some foods and then tell me how they taste to you."

- [Present the three cartoon pictures, talking about each one, one at a time]
- Match the sex of the doll with the sex of the child i.e. his/her

Yummy Just OK Yucky

"You will use 3 pictures to tell me how you think the foods taste."

- Pick up the **Yummy** face and show it to the child. Tell the child:

*"This is a picture of a **Yummy** face. See how he/she is smiling? That means he/she likes it and he/she thinks that it's yummy."*

- Place the **Yummy** face, face down on the table.

- Pick up the **Yucky** face and show it to the child. Tell the child:

*"This picture is of a **Yucky** face. See how he/she is frowning? That means he/she doesn't like it and he/she thinks that it's yucky."*

- Place the **Yucky** face, face down on the table.

- Pick up the **Just Okay** face and show it to the child. Tell the child:

*"This is the **Just OK** face. See how he/she isn't smiling and isn't frowning? That means he/she thinks it's **Just OK.** It's not yummy or yucky."*

- Place the **Just Ok** face, face down on the table.

*"So this is the **Yummy** face, he/she's smiling (*stand picture up on table*), this is the yucky face, he/she's frowning (*stand picture up on table*), and this is the **Just OK** face, he/she's not frowning or smiling (*stand picture up on table*)."*

<u>Check for understanding:</u>

"See how he/she is smiling?" (Point to the **Yummy** face) *What's the name of this face?"* (The child should say, "The yummy face.)

"See how he/she is frowning?" (Point to the **Yucky** face) *What's the name of this face?"* (The child should say, "The yucky face.)

"See how he/she isn't smiling or frowning?" (Point to the **Just Ok** face) *What's the name of this face?"* (The child should say, "The Just Ok face.)

- Hand the pointer finger to the child and ask:

*"If you tried a food and liked it, which face would you point to?" (*The child should point to the **Yummy** face)

If the child does not point to **Yummy** face say, *"If you tried a food and like it and thought it was yummy, which face would you point to?" (*The child should point to the **Yummy** face)

"If you tried a food and didn't like it, which face would you point to?" (The child should point to the **Yucky** face)

If the child does not point to **Yucky** face say, *"If you tried a food and like it and thought it was yucky, which face would you point to?" (*The child should point to the **Yucky** face)

*"If you tried a food and thought it was **Just Ok**, which face would you point to?"* (The child should point to the **Just Ok** face)

"Great! Now we are ready to play the tasting game! Let's try some bread and tortillas."

- Take the pointer finger from the child

If the child is not able to point to the correct faces, re-teach the task and ask them to identify the faces again. If the child is not able to identify faces correctly after two attempts, reschedule for another day.

<u>After a clear understanding of the faces has been established:</u>

- Place all the bread and tortillas on a tray and show them to the child. Ask the child to select a bread/tortilla to try.

Say, *"Please take one cup to try."*

- If the child takes the bread or tortilla out of the cup, but doesn't take the cup from the tray, encourage him or her to take the whole cup.

- Note the Order in which the child chose the bread/tortilla by assigning a 1 to the first bread/tortilla the child chooses to try, and do so consecutively until the last cup the child tries is number 4.

- If the child refuses, encourage the child once. When encouraging a child, you can say something like:

- Indicate all behaviors the child displayed by marking an X under the behavior(s).

"Go ahead and take a bite, and if you want you can spit it back out."

- If a child refuses first three (bread/tortillas), the last bread/tortilla cup handled can be given an Order (1). The rest will only get marked as Refusal.

- After the child takes a bite, say to the child:

"Put the cup in front of the face that tells me how it tastes to you."

- You may need to prompt them with: *"Is the bread/tortilla Yummy? Yucky? Or Just Ok"* **Always use a blank face expression when prompting them.**

- After they place it in front of a face, ask them for the name of the face.

"What face is that?"

- Wait for child's response. You may need to prompt them with: *"Is the bread/tortilla Yummy? Yucky? Or Just Ok"* **Always use a blank face expression when prompting them.**
- And then verify by asking:

"*How does it taste to you?"* You may need to prompt by asking, *"Does that mean that you think its Yummy, Yucky, or Just Okay?"*

- Make sure to record the appropriate **rating and order** under the appropriate **face** in line with the appropriate **bread or tortilla.**

- If more than one bread/tortilal is placed in front of a face (Yummy, Yucky, Just Okay) ask the child:

"Which of these two breads did you think was the yummiest?"

- Mark a 1 under "Rank" for the bread the child signifies as the yummiest.
- Mark a 2 under "Rank" for the bread the child *did NOT* signify as the yummiest i.e. the second yummiest of the two breads.
- If the child placed breads in front of different faces, leave the "Rank" column empty.
- If more than one tortilla is placed in front of a face (Yummy, Yucky, or Just Okay) ask the child:

"Which of these two breads did you think was the yummiest?"

- Mark a 1 under "Rank" for the tortilla the child signifies as the yummiest.
- Mark a 2 under "Rank" for the tortilla the child *did NOT* signify as the yummiest i.e. the second yummiest of the two tortillas.
- If the child placed tortillas in front of different faces, leave the "Rank" column empty.

"*Great! Maybe we can play it again sometime"*

Pre/Post measures only:

"Can we go next door and measure your height and weight? It's pretty neat if you want try."

Take the child to be measured:

- Have the child remove their shoes
- Have them stand in front of the tape measurer with their heels pressed against the wall.
- Make sure they are not leaning against the wall.
- Have the child look straight ahead, at the stickers on the wall in front of them. (make sure their chin is down).
- Measure their height accurately by placing a clipboard on their head. ASK FIRST
    - After you get the measurement point to the tape measure to show them how tall they are.
- Bring the child to the scale and have them stand still on it so that you can accurately gather their weight.

Finishing Activity:

I. From the observation room, closing of the activity will be noted and server will be informed to collect the food items.
II. After activities, all cups and serving dishes will be gathered onto the serving cart and brought back into the Food Laboratory.

Reminder: *The importance of writing notes after an assessment cannot be overemphasized.* If any part of the assessment seems a bit unusual to you, take the time to write a note before you start again with another child.

NOTES:

DEFINITIONS:

DO NOT CHOOSE A FACE FOR THE FOLLOWING:

Refusal: not interacting with the **food** in anyway – not touching, tasting, smelling, licking, etc. **If a child picks up the cup but does not touch the food it is a refusal**. If they pick up the cup but do not display any other behaviors, you can **mark an order, but not a face**. Each type of bread is separate from the next and the test should continue on as long as the child is willing or able regardless if they refuse previous bread or not.

Touching: is the process of physically touching the bread. You cannot mark a face if all they do is touch it.

AT THIS POINT, YOU CAN WRITE AN ORDER, AND A FACE:

Smelling: is the process of inhaling odor with your nasal passage (nose)

Licking: is the action of passing the tongue over a surface.

Spitting: is when the child has placed it in their mouth and then taken it back out again.

Swallowing: is the process of passing food into the mouth, down the pharynx, and into the esophagus.

Between Testing Children

- Double check your recording sheet or tablet. Did you record:
  - Child ID
  - Assessor initials
  - Date and Time
  - Does EVERY bread have an order, rating, and behavior OR is "refused" marked

- Throw out the tasting bread and cups. Any cup that is opened, regardless of whether or not the child touched the bread, must be disposed of. Bread can only

be reused if the lid is not removed from the cup AND the bread has not been sitting at room temperature for more than 60 minutes. (texture has not been changed)
- Wipe down trays and testing station with bleach and water solution.
- Prepare a tasting tray for the next child.

Taking down the testing stations:

- Clean up station. Wipe down table and chairs with water and bleach solution. Wipe trays and faces with water/bleach solution after final testing.

- Clean up any spilled bread.

- Dispose of all cups, lids, water bottles, etc. Pack up all supplies in identified tubs and bags. Check for any extra bread/tortillas or trays not used and keep at room temperature (in a.m.), or dispose of left overs (in p.m.).

Appendix G

Child Code:_____ Date:_____ Time of snack: _____
Exposure #:_____ Cup#_____

Ask *"Would you like to help us with the project today?"*
Child assent given? YES ☐ NO ☐

Does the child have any allergies?: (circle) YES NO
Height_____ inches Weight_____ pounds

CHILD'S HUNGER

Ask the child "Can you tell me how hungry you are feeling right now?"

FULL – Say to the child "you can point to this doll if your stomach is feeling full." 	**NEUTRAL** – Say to the child, "you can point to this doll if you are not really hungry, but you're not really full either." 	**HUNGRY** – Say to the child, "you can point to this doll if your stomach is feeling hungry." 

*Denote hunger with an "X" in the corresponding box.
**If not understood after two attempts, put a line through the hunger doll boxes and move on to activity.

Child Code:_____ Exposure #:_____

Food	Rating						Behavior					
	YUMMY 		JUST OKAY 		YUCKY 		Refused	Touched	Smell	Licked	Spit	Swallow
Hard White Tortilla												
Hard Red Tortilla												
Hard White Dyed												
Hard Red Wheat												
Hard Red Dyed												
Hard White Wheat												

*** When rating: Place ORDER of selected food as 1,2,3,etc.**

****When more than one food has the same ranking: Place RANK of food as 1= most preferred, 2= second most preferred, etc.**

NOTES

Appendix H

Type of Bread or Tortilla*

Date	Container ID	Full Container Weight + Lid	Post-Consumption Container + Waste + Lid	Child ID	Exposure #

Date	Container ID	Full Container Weight + Lid	Post-Consumption Container + Waste + Lid	Child ID	Exposure #

*A separate sheet was used for each type of wheat portion i.e. HWWT, HRWT, HWWD, HRW, HRWD, HWW

Appendix I

PROP Test Protocol- Administration

To determine if children are bitter tasters a 6-n-propylthiouracil (PROP) test will be conducted. For PROP testing, diluted water that is sipped and spit out.

Supplies:

- 60 mL plastic cup
- 10 mL PROP
- Concentration = 0.56 mmol/L of PROP
- Large 32 oz spitting cup

Instructions:

1. Present child with 60 mL plastic cup containing 10mL PROP solution and distilled water
2. Ask child to rinse mouth with distilled water prior to sipping the PROP solution.
3. Instruct child to spit all liquids (distilled water, and PROP solution) into large cup
4. Allow child to taste solution.
5. Ask child "Do you taste anything?"
    a. If child says NO ask: "Did it taste like water?"
        i. If they say no and that it tasted like water they are classified as nontasters
    b. If child says YES that it was yucky or bad they are classified as tasters
    c. If child says "I don't know what it tasted like" or provided an ambiguous response, they are to be tested at a later time.
6. Observe and record facial expressions to support verbal responses, identify ambiguous responses, or conflicting responses.