Acute Effects of Navy Bean Powder, Lentil Powder and Chickpea Powder on Postprandial Glycaemic Response and

SUBJECTIVE APPETITE IN HEALTHY YOUNG MEN

By

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A thesis submitted in conformity with the requirements

for the degree of Master of Science

Department of Nutritional Sciences

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Acute Effects of Navy Bean Powder, Lentil Powder and Chickpea Powder on Postprandial Glycaemic Response and Subjective Appetite in Healthy Young Men.

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Master of Science

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2012

ABSTRACT

In order to examine the effects of industry processed pulse powder (navy bean, lentil and chickpea) on postprandial glycaemic response (BG) and subjective appetite (App) before and after a subsequent meal, three randomized, within-subject experiments on healthy young men were conducted. In experiment 1, all navy bean treatments reduced BG at 30 min and navy bean powder suppressed pre-meal App compared to whole wheat flour. In experiment 2, all lentil treatments reduced pre-meal BG compared to whole wheat flour. However, no App differences were observed. In experiment 3, all chickpea treatments reduced pre-meal BG compared to whole wheat flour. However, no App differences were observed. Therefore, navy bean powder, lentil powder and chickpea powder maintain their low GI and satiating effects, regardless of processing. Pulse powder can be used as a value-added food ingredient to moderate glycaemic response and increase satiety.

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LIST OF ABBREVIATIONS

ANOVA Analysis of variance

BG Blood glucose

BMI Body mass index

CCK Cholecystokinin

FI Food intake

GI Glycemic Index

GLP-1 Glucagon-like peptide-1

Kcal Kilocalories

Min Minute

PYY Peptide YY (3-36)

SCFA Short chain fatty acids

SEM Standard error of the mean

VAS Visual analogue scale

CHAPTER 1

INTRODUCTION

1 INTRODUCTION

Obesity is caused by chronic energy imbalance where energy intake exceeds energy expenditure. Particularly in the Western world, increased availability of inexpensive, high energy density and fast foods (1) combined with sedentary lifestyles (2) have been associated with increased incidence of obesity. Increased consumption of more satiating and low energy density foods such as pulses, and increased physical activity may help to prevent weight gain. The beneficial effects of various pulse foods on regulation of body weight, appetite and postprandial blood glucose response are well documented.

Canada is the second largest pulse producer in the world producing 4 to 4.5 million tones of pulses per year; however, Canadians consume only 1% of the production (3-5). Fewer than 15% of Canadian adults reported consuming pulses in 2004 (6). Thus, these nutrient-dense, widely available, inexpensive foods are underutilized in Canada. Study reported that reasons for the low intake of pulses include lack of awareness of their health benefits, unfamiliarity with using and preparing pulses, and concerns with lengthy cooking time and negative side effects (7). Therefore, pulse consumption may be increased through public education on the benefits of pulses, promotion of recipes using pulses, development of ready-to-eat pulse products, and incorporation of pulses into commonly consumed foods.

Precooked, ground pulse powders have been developed to increase ease of incorporation of pulses into a diverse range of foods. These pulse powders require little cooking time and can be easily added to different food matrixes, such as pastas, breads, cookies, energy bars and soups. However, unlike whole pulses, the health benefits of pulse powders have not been demonstrated. Cooking and grinding affect starch, fiber, protein and phytochemicals of food and disrupt the cell wall structure, possibly altering the access of digestive enzymes to cell contents. The objective of this study was to determine whether commercial food processing of pulses into a powder form alters their beneficial effects on appetite and blood glucose regulation. To answer this question, we conducted three experiments to compare the acute effects of whole and powdered forms of navy beans, lentils and chickpeas on the glycaemic response and subjective appetite in healthy young men.

CHAPTER 2

LITERATURE REVIEW

2 LITERATURE REVIEW

In order to provide background for the research conducted, this review addresses 1) the relationship between pulse consumption and weight management, satiety and glycaemic response as reported in observational and experimental studies; 2) nutritional components of pulses associated with their health benefits, and possible mechanism of action; 3) the effect of processing on pulse composition and its health benefits.

2.1 Overview of Pulses

A legume is a plant (or a fruit of these specific plants) in the botanical family Leguminosae (or Fabaceae), which includes alfalfa, clover, peas, beans, lentils, chickpeas, soybeans, and peanuts. Pulses differ from other legumes in that they are exclusively harvested for their dry grain, whereas green beans and green peas are vegetable crops; soybeans and peanuts are harvested for oil extraction; and clovers and alfalfa are used for sowing (8). Pulses include dry peas, dry beans, lentils and chickpeas (3).

Pulses are nutrient dense foods with low energy density. They are high in complex and slowly digestible carbohydrates; are high in dietary fiber; are an excellent source of protein; and are low in fat, cholesterol and sodium. Pulses also contain important bioactive components, including phytochemicals (tannins, phenolic acids, flavonoids and phytic acids) and enzyme inhibitors (trypsin inhibitors and alpha-amylase inhibitors). Furthermore, they also are a good source of folate, iron, phosphorous, magnesium, potassium, calcium and zinc. Pulses are gluten free and have low glycaemic indexes, and thus are suitable for gluten sensitive individuals or diabetic patients (9). In addition, pulses are also affordable. Drewnowski (10) reported that beans (a subgroup of pulses) were among the top 5 classes of foods having the highest micronutrient to price ratio.

Various pulses differ in seed size, colour and nutritional composition. Navy beans, belong to common beans (*Phaseolus Vulgaris*) and are also known as white pea beans or haricot beans, and are traditionally used in baked beans (11). Navy beans have the highest level of dietary fiber among pulses (12). One serving (175 ml, 133 g) of boiled navy beans contains 185 calories, 34.6 g carbohydrates, 0.9 g fat, 14.0 g dietary fiber and 11.0 g protein (12). Lentils (*Lens culinaris*) are often consumed with grains and together they constitute a complete protein meal (13). Lentils have the highest level of protein in the pulse category (approximately 30% of calories are from protein). One serving (175 ml, 144 g) of boiled lentils contains 168 calories, 29.0 g carbohydrates, 0.6 g fat, 11.4 g dietary fiber and 13.0 g protein (12). Chickpeas (*Cicer arietinum*) are commonly consumed in salad, soup, stew, and processed into dips called hummus. Chickpeas have a higher percentage of energy from fat compared to other pulses (14% vs. 5%). One serving (175 ml, 120 g) of boiled Kabuli chickpeas contains 196 calories, 32.9 g carbohydrates, 3.1 g fat

(10% saturated fat), 9.0 g dietary fiber and 10.7 g protein (12). The composition of pulses will be further discussed in detail in later sections.

2.2 Pulses, Weight Management, Satiety and Glycaemic Response

The benefits of consumption of legume-rich diets by diabetic and hyperlipidemic patients has been recognized since 1976 (14, 15). Studies assessing the effects of pulse consumption on weight management, satiety and glycaemic regulation are discussed below.

2.2.1 Pulse Consumption and Body Weight

Epidemiological surveys have shown that pulse consumption is negatively associated with body weight. Based on the National Health and Nutrition Examination Survey (NHANES) 1999-2002, adult bean consumers not only have higher intakes of dietary fiber, potassium, magnesium, iron and copper, but they also have lower body weight and smaller waist circumference (16); bean consumers in adolescents aged 12-19 years also have lower body weight and smaller waist circumference (17); bean consumers in children aged 4-12 years have a trend towards reduced risk of being overweight (17). Moreover, populations whose diets are rich in pulses have lower BMIs compared to populations who followed other dietary patterns (18-21). However, cause and effect cannot be determined from observational studies. From the results of these studies, it can be concluded that consumption of greater amount of pulses is associated with lower BMI. Pulse consumption may be part of a healthy lifestyle that prevents weight gain.

Experimental studies are necessary to determine whether pulses have independent effects on body weight. In general, most randomized controlled trials reported that with energy restricted diets (30% energy deficit), pulse consumers (3-4 cups of pulses/week for 6-8 weeks) experienced significantly more weight loss compared to non-pulse consumers, possibly due to the high fiber, high slowly digestible carbohydrate and low fat content of pulses (22-24). Our laboratory also conducted two studies to examine the relationship between pulses consumption and body weight in overweight and obese subjects. In the first study, overweight and obese adults consumed 5 cups of canned navy beans per week for four weeks and experienced waist circumference reduction compared to baseline (25). In the second study, overweight and obese adults were

randomized to groups to either consume 5 cups of pulses (beans, lentils, chickpeas and yellow peas) per week or to follow a calorie-restricted diet (reduced by 500 kcal/day) for 8 weeks. Participants in both groups had significant but similar reductions in body weight, waist circumference, systolic blood pressure, HbA1c and fasting blood glucose. Therefore, consuming 5 cups of pulses per week had similar effects on weight loss and symptoms of metabolic syndrome as following an energy-restricted diet (26).

2.2.2 Pulse Consumption, Satiety and Glycaemic Response

Several short-term studies have demonstrated that pulse consumption can reduce hunger, regulate glycaemic response and suppress food intake 2-6 hours after meal consumption. If consumed frequently, the effect of pulses on these factors may help individuals with weight management and glycaemic control.

When pulses are added to a meal, they increase the feeling of fullness. Pai et al. (27) compared the satiety rating after six isocaloric (250 kcal) meals in healthy young adults. They found that the pulse-rice combination meal had a higher satiety rating compared to 3 wheat-based meals and a rice-based meal for up to 120 min after consumption. When the energy and macronutrient composition of the diets were controlled, healthy volunteers consuming chickpea salad as part of lunch at 12 pm and red bean salad as part of dinner at 6 pm, reported increased satiating feeling throughout the afternoon up to 11 pm, compared to consuming white rice as part of lunch and instant mashed potatoes as part of dinner (28). Leathwood and Pollet (29) found that an isocaloric serving (300 kcal) of shepherd's pie made with bean flakes suppressed healthy volunteers' feeling of hunger at 180 min and 240 min compared to shepherd's pie made with potato. On the contrary, Holt (30) reported subjective appetite of volunteers after consuming 38 isocaloric (240 kcal) foods over 120 min and found that potato was the most satiating food, with a satiety index higher than lentils and baked beans. Possible explanations for the contrasting findings of Leathwood and Pollet's study and Holt's study could be due to varied preparation methods and recipes among the treatments. Furthermore, Holt noted that satiety was positively correlated with protein and fiber, and negatively correlated with fat content of the food (30).

These short term studies support that pulses are satiating, as they are high in protein and fiber and low in fat.

Another metabolic benefit of consuming pulses is attenuated glycaemic response. Jenkins *et al.* (31) observed that mean blood glucose peak and area under the curve (AUC) over 2 hours in non-diabetic subjects were 45% lower after consuming portions of seven boiled pulses (butter beans, haricot beans, kidney beans, blackeye peas, chickpeas, marrowfat peas and lentils) containing 50 g carbohydrates than after consuming a comparable portion of 24 starchy foods from grains, cereals, pasta, breakfast cereals, biscuits and tuberous vegetables. This research group conducted another study in which healthy non-diabetic volunteers were given 62 common foods with 50 g of carbohydrates. Blood glucose AUC over 2 hours after eating boiled or baked beans and lentils were 65% lower compared to glucose, over 50% lower than vegetables, breakfast cereals, biscuits and cookies, and 40% lower than fruits (32). As shown in the recent updated international GI table, boiled pulses have much lower GIs than potatoes, wheat bread, rice, breakfast cereal, fruits, root vegetables, juice, bakery products (Glycaemic Index is used to estimate blood glucose response after consuming 1 g of available carbohydrate in a food with reference to glucose or white bread) (33).

Our laboratory investigated the short-term (2-6 hours) effects of consuming pulses on blood glucose response, satiety and food intake in healthy young men in several studies (36-39). In the first two studies, consumption of navy beans (containing 50 g of available carbohydrates) led to lower blood glucose net AUC over 2 hours compared to glucose or white bread. However, the recipes used for preparation of the navy beans influenced this effect. Navy beans in tomato sauce, but not navy beans in maple syrup or with pork and molasses, suppressed blood glucose response compared to white bread, due to the addition sugar in the maple syrup and pork and molasses recipes (34).

In the third study, compared to white bread, consumption of an isocaloric serving (300 kcal) of canned lentils, chickpeas and yellow peas (tomato sauce recipe) reduced blood glucose response immediately after consumption and for up to 2 hours (34). In order to investigate the effect of pulses on blood glucose response to a second meal, participants in the fourth study consumed a fixed pizza meal 2 hours after consuming 300 kcal of lentils, chickpea, yellow peas or navy

beans (tomato sauce recipe). Compared to white bread, lentils and chickpeas but not yellow peas or navy beans, suppressed blood glucose response right after the pizza meal and also at 15 min later (35).

To further assess the effects of pulses when consumed as part of a meal, our group carried out a fifth study in which pulses were consumed with macaroni and tomato sauce as a mixed meal (36). Consumption of an isocaloric serving (600 kcal) of mixed meal with lentils, yellow peas and chickpeas lowered blood glucose immediately following consumption compared to macaroni and cheese. In addition, the lentil meal and yellow pea meal, but not the chickpea meal, led to lower subjective appetite AUC over 260 min and less food intake at the pizza meal, compared to macaroni and cheese (36).

In order to mimic real life meal consumption, in the sixth study, meals of chickpeas, lentils, navy beans or yellow peas with macaroni and tomato were served *ad libitum*, followed by an *ad libitum* pizza meal 4 hours later. Macaroni and tomato sauce with chickpeas, lentils and navy beans reduced pre-pizza meal BG AUC (0-260 min) compared to macaroni and tomato sauce alone. In addition, macaroni and tomato sauce containing lentils suppressed accumulative food intake compared to macaroni with tomato sauce alone (37).

This series of studies demonstrate that consuming pulses alone or as part of a meal has short-term benefits on blood glucose response, appetite and subsequent food intake. However, the magnitude of the benefit is dependent on processing methods, recipes and pulse type.

Overall, pulses have been shown to reduce the risk of obesity in epidemiological studies, assist with weight loss if consumed often over 6-8 weeks in long term studies, reduce glycaemic response and increase satiety up to 6 hours after consumption in short term studies.

2.3 Pulse Components

The starches, fiber, oligosaccharides, protein and phytochemicals of pulses contribute to their effects to delay digestion and absorption, lower blood glucose response and prolong feelings of satiety. The effect of commercial processing and home preparation of pulses alters these components, and thus may also affect biological response to consuming pulses.

2.3.1 Pulse Starches

Starches account for 22-45% of the pulse (by dry weight) (38). Pulse starches are slowly digestible or resistant to digestion, thus producing an attenuated postprandial glucose response (31, 39, 40). The slow digestibility of pulse starches is attributed to the nature of starches, enzyme inhibitors and the interaction of starches with protein and antinutrients, such as phenolic compounds and phytic acid (41).

The higher amount of amylose in pulse starches contributes to their slow digestibility. Pulse starch is 30-40% amylose, while cereal and rube starches contain about 25-30% amylose (40). Amylose is a linear polymer containing several hundred glucose units whereas amylopectin is composed of several thousand glucose units, with a long linear chain and branches every 24 to 30 glucose units (42). Therefore, amylopectin has a larger surface area than amylose for α -amylase to attack (40). As demonstrated in animal study, starches high in amylopectin were digested more quickly and completely than starches high in amylose (43). Also, amylose is abundant with hydrogen bonds, it is more prone to retrogradation and forms more resistant starch than amylopectin (42).

Resistant starch is defined as "the sum of starch and products of starch hydrolysis not absorbed in the small intestine of healthy individuals" (44). Because resistant starch is not digested and therefore not absorbed as glucose in the small intestine, it delays glycaemic response compared to digestible starch. Blood glucose peak after a high digestible starch meal (3.07±0.29 mmol/L) is nine times greater than after a high resistant starch meal (0.36±0.13 mmol/L) (45). Resistant starch also behaves similarly to soluble fiber: it slows gastric emptying, reduces glucose and fat absorption (45), and is fermented in the colon and produces short chain fatty acids (SCFA). In

addition, resistant starch may be useful for weight management since its energy value, including energy from fermentation, is half the value of digestible starch (46).

Much of the resistant starch in raw pulses is destroyed after heat treatment, but some is reformed during retrogradation. Different processing methods alter the amount of resistant starch in beans. For example, retrograded resistant starch content was higher in boiled beans (2.65-2.79%) than in autoclaved beans (1.62-1.94%) (47). Thus as processing affects retrograded starch content of pulses, it may also affect biological responses to consuming pulses.

2.3.2 Pulse Fiber

Pulses are high in fiber. Half a cup of boiled pulses contains 7 g of fiber on average (42). Many studies have reported that dietary fiber can increase satiety, reduce energy intake and lower blood glucose response (48) (49) (50).Raben *et al.* (48) reported increased feelings of fullness following a high fiber meal with pea fiber compared to a low fiber meal. Howarth *et al.* (49) reported that when meals were served *ad libitum*, volunteers reduced energy intake by 10% when consuming a high fiber diet compared to a low fiber diet. Joan *et al.* (50) reported fiber lowered postprandial blood glucose responses to a high carbohydrate meal in healthy individuals. Jenkins *et al.* (50) also reported that adding a purified fiber product to wholemeal bread reduced the blood glucose AUC over 3 hours to 51% in diabetic individuals.

Fiber regulates satiety signals and blood glucose throughout the course of digestion: when food enters the stomach, fiber's high water-holding capacity adds bulk to the food and increases gastric distension. When food moves to the small intestine, viscous soluble fiber slows gastric emptying (49). In the small intestine, fiber affects the release of gut hormones, glucagon-like peptide-1 (GLP-1), cholecystokinin (CCK) and Peptide YY (PYY), and increases the feeling of satiety (51-54). Fiber also reduces small intestinal absorption of minerals (55) and fat (56). When food reaches the colon, soluble fiber ferments and positively affects colon health through the production of SCFAs. SCFAs are oxidized and used for energy in preference to glucose, possibly lead to a stable glucose pattern over time (57, 58). Furthermore, SCFAs from the fermentation of

fiber are found to increase satiety (59), by increasing the expression of GLP-1 precursor proglucagon mRNA(53), leading to increased levels of GLP-1 hormone.

2.3.3 Oligosaccharides

The oligosaccharides found in pulses are responsible for the negative side effects of pulse consumption. They are difficult to digest and the undigested portion is fermented in the colon, resulting in bloating and gas. However, they also act as prebiotics and change the gut microflora by promoting the growth of beneficial bacteria (60). Lean and obese individuals have different gut microflora profiles, as the microflora in obese individuals extract more energy from the products of colonic fermentation (61). Therefore, by changing the gut microflora, oligosaccharides may play a role in regulating energy availability from fermentation in the colon.

2.3.4 Pulse Protein

Protein accounts for 17-35% of pulse composition (by dry weight) and is the most satiating macronutrient (62-64). Pulse proteins have been shown to affect appetite suppression and blood glucose control. Chris *et al.* (65) observed that a meal with 20 g of pea protein significantly reduced food intake 30 min later compared to a meal without pea protein in young male subjects. Blood glucose responses before and after the subsequent meal at two different time points (30 min and 120 min) were also suppressed by pea protein (65). Protein regulates blood glucose and satiety by triggering insulin (66) and anorexic hormones GLP-1 (67). Pepsin-derived peptides from country beans have also been found to stimulate the secretion of the anorexic hormone, CCK (68).

Protein also affects blood glucose and satiety by binding to starch to form a complex and slowing starch digestion. Protein-starch complexes have been found in potato starch (69) and legume protein isolates (70). Anderson *et al.* (71) observed increased carbohydrate malabsoprtion in white breads compared to gluten-free breads. They suggested that the protein-starch complex in

the white bread restricted the accessibility of the starch to digestive enzymes and consequently reduced digestibility.

Pulses contain protein-based protease inhibitors and amylase inhibitors that may also affect the biological responses to consuming pulses. Pulses have higher amounts of trypsin inhibitors compared to other plants, but proper cooking deactivates most of the inhibitors, and they do not appear to have an effect on weight loss (72). In contrast, α -amylase inhibitors found in pulses reduce starch digestion and absorption and have been shown to affect weight management (73). Alpha-amylase inhibitors from navy beans caused hypoglycemia and reduced growth rate in rats (74). Clinical studies also found that isolated α -amylase inhibitors lowered postprandial blood glucose levels (75). However, similar to trypsin inhibitors, most a-amylase inhibitors are inactivated during processing, such as boiling and autoclaving, although low levels of activity may remain (75).

There is recent interest in preparing pulse extracts using methods to retain the activities of α -amylase inhibitors for use in weight loss treatment (76-80). In those studies, initially overweight or obese participants were randomized to take pills or powder containing amylase inhibitors (also referred to as "starch blocker"), or placebo for 1-3 times a day for 4-12 weeks. In all studies, greater weight loss was observed in the treatment groups compared with the placebo group, although in some studies the difference was not statistical significant (76-80). Thus, pulse extracts containing active α -amylase inhibitors have the potential to be used as part of the treatment of obesity. Furthermore, these studies demonstrate the importance of processing methods as they can significantly affect α -amylase inhibitor activities and consequently affect starch digestibility.

2.3.5 Pulse Phytochemicals

Pulses contain a number of phenolic compounds, such as tannins, phenolic acids and flavonoids (73). Small red beans, red kidney beans, pinto beans and black beans are among the top 20 foods that contain the highest antioxidant levels (81), which is related in part to the amount of phenolic compounds in the foods (82). These compounds are known for their anticarcinogenic,

antimutagenic and antimicrobial properties (82). They also chelate metals such as iron and zinc and inhibit carbohydrate and protein digestive enzymes (83). Certain phenolic compounds can interfere with glucose transporters in enterocytes during carbohydrate uptake (84, 85) and precipitate proteins during protein digestion (86). Pulses are also one of the primary sources of phytic acid in the diet (87). Phytic acid is indigestible to humans and forms insoluble complexes with zinc, iron, calcium and magnesium. Yooh *et al.* (88) showed that unleavened white bread with added phytic acid decreased starch digestion rate *in vitro* as well as delayed postprandial glycaemic response in humans compared to plain unleavened white bread. Therefore, high level of phenolic compounds and phytic acid may also contribute to the attenuated glycaemic response and increased satiety of pulses.

2.4 Pulse Processing

Development of processed pulse powders is one approach to increasing the consumption of pulses, as pulse powders are more easily incorporated into commercial food products. As discussed above, however, processing pulses alters a variety of nutritional and antinutritional components, which may impact digestion and absorption, and ultimately the biological response to consuming pulses. The two main procedures of processing pulses in powders are cooking and grinding.

2.4.1 Cooking

Cooking methods for pulse processing includes autoclaving or canning, boiling, roasting, steaming and micronization.

Canned pulses are the most common pulses consumed in the Western world and the biological responses to canned pulses have been compared with home cooked pulses in numerous studies. Wong *et al.* (89) reported that starch from canned beans was hydrolyzed much faster than starch from either boiled or baked home-cooked beans. Traianedes and O'Dea (90) reported that the postprandial glucose and insulin responses to home-cooked baked beans were flatter and more attenuated compared to the responses to canned baked beans. The significant increase of starch digestibility in canned products is largely caused by autoclaving in the canning process. The high temperature and pressure environment (15psi for 121°C) accelerates the penetration of water into starch granules and increases the degree of starch gelatinization, thereby making the starch more accessible to digestive enzymes. Studies have shown that autoclaving significantly increased starch digestive rate *in vitro* (90) and postprandial glycaemic and insulin responses *in vivo* (91).

Micronization is a process used in the production of cooked, flaked cereals and instantized pulse products (92). In this process, grains are exposed to electromagnetic radiation in the wavelength region of 1.8–3.4 μm (93) for 2-3 min (94). These infrared waves cause molecules to vibrate at 60,000-150,000 MHz, therefore producing a rapid internal heat (93). The temperature can reach approximate 140°C and all starches are gelatinized (93). Then the grains are either dried as

whole micronized grains, or flaked and milled into powders (93). Micronized pulse flakes and powders significantly shortened cooking time later on compared to raw pulse flakes and powders (93, 95).

The moisture content during cooking is another important factor that influences starch digestibility of pulses. Greevani and Theophilus (96) observed a decrease in starch digestibility *in vitro* and *in vivo* in rats of pigeon peas, chickpeas, black lentils and mung beans after roasting the pulses for 10 min compared to boiling for 30 min and pressure cooking for 10 min. Siljestrom and colleagues (97) suggested that the formation of non-starch linkages under a low-moisture environment during cooking restricts the enzymatic access of starch, thus reducing digestibility. Similarly, Khattab *et al.* (98) found that protein digestibility of cowpeas, peas and kidney beans *in vitro* was significantly improved after boiling (35-45 min), microwave cooking (15-20 min), and pressure cooking (20 min) but was slightly decreased after roasting (15-20 min) and micronization (2.5-3 min). On the other hand, the tannins and phytic acid, which lower carbohydrate and protein digestibility, were deactivated to a greater degree by boiling and autoclaving compared to roasting and micronization of cowpeas, peas and kidney beans (98, 99). Boiling and autoclaving cowpeas, peas and kidney also caused a greater loss of oligosaccharides compared to roasting and micronization, due to the hydrolysis of oligosaccharides and the formation of di- and mono-saccharides (98-100).

2.4.2 Grinding

Breaking whole pulses to small particles through grinding or pureeing in a blender increases starch accessibility and digestibility. Wursch *et al.* (101) reported that the *in vitro* starch digestion rate of pureed cooked kidney beans was increased compared to whole cooked kidney beans. They observed that the starch granules in cooked beans were highly gelatinized, but the total swelling of starch granules was hampered by the rigidity of the cell walls. In contrast, some of the cell walls in the cooked and then pureed beans were broken and starch granules were released from the cells, thus the accessibility of starch to the enzymes was increased, which is likely responsible for increased digestibility. Increased starch digestibility was also reported for pureed red lentils and yellow peas *in vitro* (101). However, pureeing lentils did not increase glucose response in healthy human volunteers (102). A close relationship between *in vitro* starch

hydrolysis rate and postprandial glucose responses, which has been shown in cereals, is not evident with lentils (103).

Whether pulses are ground into smaller particles before or after cooking is another factor that influences starch digestibility. In the same study, Wursch *et al.* (101) found kidney beans, lentils and yellow peas that were ground after cooking (GAC) have higher starch digestibility than kidney beans, lentils and yellow peas that were ground before cooking (GBC) *in vitro*. Kon *et al.* (104) also found that the rate of starch hydrolysis was higher with GBC small white beans compared to the GAC beans. Furthermore, Tovar *et al.* (91) found that GBC red kidney beans produced higher glycaemic response compared to GAC red kidney beans since GBC pulses contain mostly free starches while GAC pulses contain a large number of intact cells with enclosed starch granules (91).

Therefore, grinding pulses to smaller particles increases its starch digestibility; and grinding before cooking, compare to grinding after cooking, also promotes starch digestibility.

2.4.3 Other Factors

Cooking temperature, cooking duration and alkaline treatment during soaking preparation also positively affect digestibility of starch in pulses (90, 105). In addition, combining multiple cooking and processing methods, as described by Jenkins *et al.* (105) and Tovar *et al.* (91), may increase the digestion rate of food compared to a single cooking method alone. These studies are further discussed below.

2.4.4 Studies on pulse powders and blood glucose response

The impact of method of cooking and processing of pulses on glucose response has been a topic of interest for many years. Jenkins *et al.* (102) conducted a study to assess the effect of processing of lentils on subsequent glucose response in eight healthy volunteers. The lentils were: a) boiled for 20 min (20min lentils); b) boiled for 20 min and pureed (pureed lentils); c) boiled

for 1h (1h lentils); and d) boiled for 20 min, pureed, dried for 12 hours at 250°F and then ground into a powder (12h lentils). 1h lentils and pureed lentils resulted in a similar glucose response as 20min lentils. However, 12h lentils resulted in significantly increased glucose responses at 15, 30 and 45 min following consumption compared to 20min lentils. The authors suggested the "slow release" property of lentils was altered by prolonged dry heat but not milling or refining. The 12 hours dry heat may have altered the nature of starch, the relationship between starch and fiber, and the relationship between starch and protein, making starch more readily available for digestion (102).

Tovar *et al.* (91) compared the glycaemic responses of consumption of various preparation of kidney beans and lentils in normal weight subjects. The treatments were a) boiled, b) autoclaved red kidney beans, c) precooked flour (PCF), made by boiling, freeze-drying and milling kidney beans, d) free starch flour (FSF), made by milling, steam-cooking, freeze-drying and milling kidney beans, and e) FSF lentils. All treatments were controlled for starch, weight, were similar in protein, fat and caloric contents. Compared to autoclaved red kidney beans, FSF but not PCF had a faster and greater blood glucose peak because FSF contained free starch while PCF has cell-enclosed starch. Compared to red bean PCF, lentil PCF resulted in greater blood glucose and insulin peaks. This suggested that the effect of processing of legumes on digestibility and impact on blood glucose following consumption may differ for different legumes. Therefore, examining the effect of various processing methods on different pulses is necessary.

These studies investigated the impact of different laboratory processing procedures on pulses at a laboratory level; as such, the results may not be representative of the effects of industrial processing of legumes. For example, beans are soaked for 3 hours at 29°C, blanched for 6 min at 80°C and then canned for 73 min at 121°C to produce canned backed navy beans in Canada (34). Salts, acidulates, and/or alkalis (for example: EDTA, sodium bicarbonate, mixture of carbonates and phosphates) are added during soaking, blanching, and autoclaving to shorten the cooking time (106). Therefore, compared to laboratory processing procedures, industry processing of pulses is a more complicated procedure with high consistency end products. Although studies on laboratory processed pulses serve as a good reference to industrial processed pulses, studies on commercially processed pulses are necessary.

There have been a few studies investigating the effects of foods with added ready-to-use pulse powders on appetite and blood glucose control using foods made of whole wheat flour as a reference.

In the development of novel breads and pasta products, substituting part of the wheat flour with pulse flours increased the protein, dietary fiber and resistant starch content (107-111) Using *in vitro* starch digestibility data, Osorio-Díaz *et al.* (109) predicted that the glycaemic index of spaghetti containing 25% added chickpea flour would be lower than durum wheat-control pasta. Also, Goni and Valentin-Gamazo (110) observed that pasta with added 25% chickpea flour resulted in a lower glycaemic response in subjects compared to 100% wheat pasta.

In a single-blind crossover trial, Marinangeli et al. (111) investigated the glycaemic responses of healthy subjects to a variety of food items including banana bread and biscotti, made with whole yellow pea flour (WYPF) or whole wheat flour (WWF) as the primary ingredient; spaghetti made with 30% WYPF and 70% white flour; and 100% whole wheat spaghetti. Boiled yellow peas (BYP) and white bread (WB) were the positive and negative controls, respectively. Interestingly, the glycaemic response did not differ among WYPF banana bread, WYPF biscotti and BYP. When WYPF products (banana bread, biscotti and spaghetti) were compared individually to WWF products, only WYPF biscotti produced a lower postprandial glycaemic response compared to WWF biscotti. Since WYPF biscotti contained 1.5 g less carbohydrate than WWF biscotti, the lower glycaemic response cannot be attributed to the WYPF only. Several other confounding factors in this study were noted: 1) the total available carbohydrates in treatment foods was not controlled (ranged 51.1-53.2 g, with no nutritional information for BYP and WB); 2) the available carbohydrates sources differed greatly between treatments (e.g. in WYPF banana bread, available carbohydrates were from banana, sugar and yellow pea flour, whereas in BYP, available carbohydrates were from yellow pea only); 3) the weight of the treatments as presented in the nutritional composition table was less than the sum of macronutrients; and 4) the subject population was highly variable (aged 22-67 yrs; BMI 21-42 kg/m²). These factors may have contributed to the lack of observed statistical differences in the response to the foods containing yellow pea flour compared to foods containing wheat flour only, and the similarity of glucose response after consuming the whole yellow pea and yellow pea

flour treatments. This study suggests that the benefits of foods with added pulses powder depend on the pulse flour incorporation ratio and recipe.

In summary, the addition of commercially prepared ready-to-eat pulse flours to commercial food products such as breads, cookies, pastas and others is a desirable way of increasing consumption of one of Canada's healthy primary agricultural products. However, there is concern that the processing of pulses into these convenient, easy-to-use products may negatively impact the observed health benefits of pulses, including impact on blood glucose control and satiety. Published literature to date has not adequately addressed the question of how ready-to-use commercially prepared pulse powders affect glycaemic response and appetite control in comparison with whole pulses. A study to compare the effect of pulse powder and whole pulses on glycaemic response and satiety in a population with similar characteristics, using treatments that are prepared in the same manner with same amount of available carbohydrate is needed. Thus, the objective of the present study was to fill this gap.

2.5 Conclusion

The benefits of consuming pulses on the regulation of body weight, appetite and postprandial blood glucose response have been reported in numerous studies. These benefits are attributed to the high content of complex carbohydrates, fiber, protein and antinutrients in pulses. Although the effects of various types of cooking and processing of pulses and resultant in changes in macronutrients and micronutrients have been reported, few studies have examined commercially processing of pulse powders. Thus, it is unknown if processing of pulses to ready-to-use powders reduces their benefits on appetite and blood glucose regulation. To answer this question, we conducted a serious of studies to compare the acute effects of powdered pulses and whole pulses on glycaemic response and subjective appetite in healthy young men.

CHAPTER 3

HYPOTHSIS, OBJECTIVE, DESIGN

3.1 Hypothesis

It is hypothesized that powdered pulses, compared to whole pulses, are less effective for postprandial blood glucose and subjective appetite control.

3.2 Objective

The objective of the study is to compare the acute effects of powdered pulses and whole pulses on glycaemic response and subjective appetite in healthy young men.

Specific Objectives:

- 1. To compare the acute effects of powdered navy beans and whole navy beans on glycaemic response and subjective appetite in healthy young men.
- 2. To compare the acute effects of powdered lentils and whole lentils on glycaemic response and subjective appetite in healthy young men.
- 3. To compare the acute effects of powdered chickpeas and whole chickpeas on glycaemic response and subjective appetite in healthy young men.

3.3 Experimental Design

Three experiments were conducted in healthy young men. The study followed a within-subject, randomized, single blind and repeated-measure design in which healthy young men attended 4 sessions where they received whole canned pulses, or pureed canned pulses, or pulse powders or whole wheat flour in a randomized order. All treatments were controlled for available carbohydrate, sodium and weight. Navy beans, lentils and chickpeas were studied in experiments 1, 2 and 3, respectively. A fixed pizza meal (12 kcal/kg of body weight) was served at 120 min. Blood glucose (BG) and subjective appetite (SA) were measured at 0, 15, 30, 45, 60, 90, 120, 140, 155, 170, 185 and 200 minutes.

CHAPTER 4

MATERIALS AND METHODS

4.1 Measurements

4.1.1 Blood Glucose

Blood samples obtained by finger pricks were used to measure blood glucose. After cleaning their fingers with alcohol swabs, subjects pricked their fingers using a Monoinjector Lancet Device (Sherwood Medical, St Louis, MO, USA). The first blood drop was wiped off to prevent alcohol contamination and glucose in the second drop was measured using a handheld glucometer (Accu-Chek Compact, Roche Diagnostics Canada, Laval, QUE, Canada).

4.1.2 Subjective Appetite

Motivation to Eat questionnaires (Appendix I) were used to assess subjective appetite (112). The questionnaires contained four questions.

Q1. How strong is your desire to eat?

Q2. How hungry do you feel?

Q3. How full do you feel?

Q4. How much food do you think you could eat?

Each question was measured by a Visual Analog Scale (VAS), a 100 mm line affixed with opposing statements at each end. Subjects were asked to mark an "X" on the line to depict their feelings at a given moment. Scores were determined by measuring the distance from the left end to the intersection of the "X" (112).

Average appetite was calculated as:

Average Appetite = [Q1 + Q2 + (1 - Q3) + Q4] / 4

4.1.3 Thirst and Water Intake

Subjective thirst was measured using the VAS question: "How thirsty do you feel?" (Appendix I) (113). Water was given *ad libitum* with the fixed pizza meal at 120 min and water intake was measured by weighing the water before and after the meal.

4.1.4 Palatability

The palatability of each treatment as well as the fixed pizza meal were rated by VAS (Appendix I) (113). The questions for treatment included:

- Q1. How pleasant have you found the beverage/food?
- **Q2.** How tasty have you found the treatment?
- **Q3.** How did you like the texture of the treatment?

Average palatability of treatment was calculated as:

Average Palatability =
$$(Q1 + Q2 + Q3)/3$$

The question for the pizza meal was: "How pleasant have you found the beverage/food?" (Appendix I)

4.1.5 Physical Comfort

Physical comfort was measured by physical comfort VAS (Appendix I). The questionnaires contained five questions (114):

- Q1. Do you feel nauseous?
- **Q2.** Does your stomach hurt?

- Q3. How well do you feel?
- **Q4.** Do you feel like you have gas?
- **Q5.** Do you feel like you have diarrhea?

Average physical comfort was calculated as:

Average Physical Comfort =
$$[(1 - Q1) + (1 - Q2) + Q3 + (1 - Q4) + (1 - Q5)] / 5$$

4.1.6 Energy and Fatigue

Subjective energy and fatigue were measured using VAS questionnaires (Appendix I) (114). The questionnaires contained two questions:

- Q1. How energetic do you feel right now?
- **Q2.** How tired do you feel right now?

4.2 Study Participants

Healthy male subjects aged 18-30 years with a body mass index between 20 and 24.9 kg/m² (115) were recruited via advertisements placed around the University of Toronto St. George campus, local newspapers and on student websites (Appendix II). To reduce between-subject variation and the effects of potential confounders, subjects were excluded if they had diabetes mellitus or any other metabolic disorders; were taking medications; were dieting; were frequent breakfast skippers, were smokers; or participated in any other nutrition-related studies within 4 weeks before the study.

Sample size of 12 was determined based on power analysis using data from previous fixed-meal studies (34, 35) and was designed to detect a difference of 0.5 mmol/L in blood glucose between treatments at a power level of 0.8 ($\alpha = 0.05$).

4.3 Screening

During screening sessions, subjects' height and weight were measured. If the BMI fell within the qualifying range, subjects were instructed to complete the Baseline Information Questionnaire form (Appendix III), the Eating Habits Questionnaire form and Food Acceptability Questionnaire (Appendix III) to investigate if they had any metabolic disorders, took medications, were on a diet, ate breakfast routinely, had significant weight fluctuation in the past and did not have any aversion to the test foods. If they met all inclusion criteria, study information was explained to eligible subjects. Then subjects chose a time between 9:45 a.m. and 12:45 p.m. to start the session and the chosen time was consistent for all 4 sessions of the experiment. Subjects were asked to abstain from caffeine and alcohol, maintain a normal routine of physical activity and to consume a similar dinner the day before each session. Proper procedures for obtaining finger-prick blood samples to assess blood glucose levels were demonstrated. How to correctly fill Visual Analog Scale questionnaires was also explained. Subjects then read and signed the Information Sheet and Consent Form (Appendix III).

4.4 Treatments

There were four treatments in each of the three experiments: tomato sauce with (1) whole canned pulses, (2) pureed canned pulses, (3) pulse powders or (4) whole wheat flour (control). All treatments had the same amount of available carbohydrates (38.8 g) where 25 g is from the test ingredient and 13.8 g is from the tomato sauce. Sodium and treatment weight were standardized in each experiment (**Table 4.1-4.3**). All treatments were prepared the day before the sessions, sealed in airtight containers and stored in an experimental fridge. Before serving, the treatments were heated in a microware under high power for 1 minute and 30 seconds. Detailed treatment preparation and ingredient list can be found in Appendix IV.

TABLE 4.1 Experiment 1/ Navy bean: treatments nutritional facts

| WB^2 | PB | BP | WF |
|--------|--|--|---|
| 234.1 | 234.1 | 248.5 | 195.2 |
| 1.9 | 1.9 | 2.1 | 0.6 |
| 1.5 | 1.5 | 1.7 | 0.7 |
| 17.5 | 17.5 | 20.5 | 9.6 |
| 55.0 | 55.0 | 60.1 | 46.6 |
| 16.3 | 16.3 | 21.4 | 7.9 |
| 38.7 | 38.7 | 38.7 | 38.7 |
| 375.9 | 375.9 | 375.9 | 375.9 |
| 405.0 | 405.0 | 405.0 | 405.0 |
| | 234.1 1.9 1.5 17.5 55.0 16.3 38.7 375.9 | 234.1 234.1 1.9 1.9 1.5 1.5 17.5 17.5 55.0 55.0 16.3 16.3 38.7 38.7 375.9 375.9 | 234.1 234.1 248.5 1.9 1.9 2.1 1.5 1.5 1.7 17.5 17.5 20.5 55.0 55.0 60.1 16.3 16.3 21.4 38.7 38.7 38.7 375.9 375.9 375.9 |

¹Fiber is calculated as 0 kcal/g.

² WB, whole navy beans; PB, pureed navy beans; BP, navy bean powder; WF, whole wheat flour.

TABLE 4.2 Experiment 2/ Lentil: treatments nutritional facts

| Treatments | WL^2 | PL | LP | WF |
|------------------------------|--------|-------|-------|-------|
| Calories ¹ (kcal) | 225.5 | 225.5 | 220.7 | 201.1 |
| Ash (g) | 1.8 | 1.8 | 1.1 | 0.6 |
| Fat (g) | 0.9 | 0.9 | 0.8 | 0.9 |
| Protein (g) | 16.5 | 16.5 | 15.5 | 10.5 |
| Carbohydrates (g) | 50.0 | 50.0 | 49.0 | 46.6 |
| Dietary Fiber (g) | 11.3 | 11.3 | 10.3 | 7.9 |
| Available Carbohydrate (g) | 38.7 | 38.7 | 38.7 | 38.7 |
| Sodium (mg) | 566.3 | 566.3 | 566.3 | 566.3 |
| Weight (g) | 450.0 | 450.0 | 450.0 | 450.0 |
| | | | | |

Fiber is calculated as 0 kcal/g.

² WL, whole lentils; PL, pureed lentils; LP, lentil powder; WF, wheat flour.

TABLE 4.3 Experiment 3/ Chickpea: treatments nutritional facts

| Treatments | WC^2 | PC | CP | WF |
|------------------------------|--------|-------|-------|-------|
| Calories ¹ (kcal) | 240.3 | 240.3 | 245.3 | 201.1 |
| Ash (g) | 1.6 | 1.6 | 1.5 | 0.6 |
| Fat (g) | 3.6 | 3.6 | 3.5 | 0.9 |
| Protein (g) | 14.3 | 14.3 | 15.6 | 10.5 |
| Carbohydrates (g) | 49.2 | 49.2 | 48.3 | 46.6 |
| Dietary Fiber (g) | 10.5 | 10.5 | 9.6 | 7.9 |
| Available Carbohydrate (g) | 38.7 | 38.7 | 38.7 | 38.7 |
| Sodium (mg) | 566.3 | 566.3 | 566.3 | 566.3 |
| Weight (g) | 450.0 | 450.0 | 450.0 | 450.0 |
| | | | | |

Fiber is calculated as 0kcal/g.

² WC, whole chickpea; PC, pureed chickpea; CP, chickpea powder; WF, wheat flour.

4.5 Protocol

The University of Toronto Human Subjects Review Committee approved the study protocol. The protocol used in this study was similar to the one used in previous short-term studies in our laboratory (116, 117). Following a 10-12 hours overnight fasting, 4 hours before the scheduled session time, subjects consumed a standardized breakfast consisting of 26 g of Honey Nut Cheerios cereal (General Mills, Mississauga, Ontario), 250 mL of Beatrice 2% milk (Parmalat Canada, Toronto, Ontario) and 250 mL of Tropicana orange juice (Tropicana Products Inc., Bradenton, Florida) in 15 min at home. 500 mL of water (Canadian Springs, Toronto, Ontario) was provided and subjects were required to finish the bottle by 1 hour prior to their sessions.

Fifteen min before the sessions, subjects arrived at the lab and filled out a Sleep Habits and Stress Factors Questionnaire (Appendix I). If they reported significant deviations from their usual patterns, they were asked to reschedule. Then they completed baseline Subjective Appetite, Physical Comfort, Energy and Fatigue VAS (Appendix I) and their baseline blood samples were measured. If the blood glucose concentration was higher than 5.5 mmol/L, subjects were asked to reschedule.

Then subjects were given 15 min to consume the treatment along with 250 mL of filtered water in their entirety. Following treatment consumption, pleasantness, taste and texture of the treatments were measured using Palatability VAS (Appendix I). Motivation to eat, physical comfort, energy/fatigue and blood glucose were measured and repeated up to 120 min (at 15, 30, 45, 60, 90 and 120 min.).

In order to measure post-second meal glycaemic response and subjective appetite, at 120 min, subjects were asked to consume a fixed pizza meal (12 kcal/kg body weight, McCain Deep 'N Delicious; McCain Foods Ltd) with 500 ml of filtered water (which they could consume *ad libitum*) within 20 min. The palatability of pizza meal was measured immediately following the meal at 140 min (Appendix I), followed by blood glucose, motivation to eat, physical comfort, energy and fatigue measurements repeated until the end of the study session (140, 155, 170, 185 and 200 min.).

4.6 Data Analysis

All statistical analyses were conducted using the SAS version 9.2 (Statistical Analysis Systems, SAS Institute Inc., Cary, North Carolina) software suite. Two-way repeated measures analysis of variance (ANOVA) tests were used to test for treatment, time and treatment by time interaction effects on glycaemic response, subjective appetite, physical comfort, energy, tiredness and thirst. For variables with a significant treatment and/or interaction effect (p<0.05), one-way repeated-measures ANOVA and Tukey-Kramer's post-hoc test was used to determine between-treatment differences at individual time points. Treatment effects were tested via one-way repeated measured ANOVA on water intake, treatment palatability and pizza palatability. Correlation analyses among outcome measures were performed using the Pearson's Correlation Coefficient. All results are presented as mean ± standard error of the mean (SEM). Statistical significance was concluded with the P-value less than 0.05.

CHAPTER 5

RESULTS

5.1 Subject Characteristics

Seventeen subjects were recruited and completed experiment 1. The average BMI was 22.9 kg/m² and the average age was 22.1 yr (**Table 5.1**). Twelve subjects were recruited and completed experiment 2. The average BMI was 23.2 kg/m² and the average age was 22.2 yr (**Table 5.2**). Twelve subjects were recruited and completed experiment 3. The average BMI was 22.3 kg/m² and the average age was 23.6 yr (**Table 5.3**).

TABLE 5.1 Experiment 1/ Navy bean: Subject characteristics

| Subject No. | Age (y) | BMI ¹ (kg/m ²) | Weight(kg) | Height(cm) |
|----------------|------------|---------------------------------------|------------|------------|
| 1 | 24 | 22.91 | 72.6 | 178.0 |
| 2 | 21 | 20.92 | 62.6 | 173.0 |
| 3 | 20 | 21.84 | 64.6 | 172.0 |
| 4 | 22 | 23.22 | 68.7 | 172.0 |
| 5 | 21 | 20.50 | 53.8 | 162.0 |
| 6 | 19 | 21.74 | 66.2 | 174.5 |
| 7 | 18 | 23.61 | 72.3 | 175.0 |
| 8 | 20 | 24.73 | 81.9 | 182.0 |
| 9 | 29 | 21.99 | 64.3 | 171.0 |
| 10 | 20 | 24.02 | 82.2 | 185.0 |
| 11 | 28 | 22.33 | 68.4 | 175.0 |
| 12 | 22 | 24.39 | 76.4 | 177.0 |
| 13 | 20 | 24.88 | 76.2 | 175.0 |
| 14 | 23 | 23.57 | 86.0 | 191.0 |
| 15 | 18 | 22.66 | 78.4 | 186.0 |
| 16 | 23 | 21.74 | 72.4 | 182.5 |
| 17 | 28 | 23.55 | 71.3 | 174.0 |
| Mean | 22.12 | 22.86 | 71.66 | 176.76 |
| SE | 0.83 | 0.31 | 1.98 | 1.66 |

BMI = Body Mass Index

TABLE 5.2 Experiment 2/ Lentil: Subject characteristics

| Subject No. | Age (y) | BMI¹ (kg/m²) | Weight(Kg) | Height(cm) |
|----------------|------------|--------------|------------|------------|
| 1 | 21 | 23.80 | 72.9 | 175.0 |
| 2 | 24 | 21.68 | 75.8 | 187.0 |
| 3 | 20 | 23.17 | 73.4 | 178.0 |
| 4 | 27 | 22.37 | 77.4 | 186.0 |
| 5 | 21 | 24.00 | 81.7 | 184.5 |
| 6 | 29 | 23.87 | 73.1 | 175.0 |
| 7 | 22 | 22.93 | 74.3 | 180.0 |
| 8 | 21 | 23.81 | 76.3 | 179.0 |
| 9 | 21 | 24.83 | 77.8 | 177.0 |
| 10 | 18 | 24.03 | 71.1 | 172.0 |
| 11 | 20 | 20.38 | 66.4 | 180.5 |
| 12 | 22 | 23.71 | 72.2 | 174.5 |
| Mean | 22.17 | 23.22 | 74.37 | 179.04 |
| SE | 0.89 | 0.35 | 1.11 | 1.38 |

BMI = Body Mass Index

TABLE 5.3 Experiment 3/ Chickpea: Subject characteristics

| Subject No. | Age (y) | BMI¹ (kg/m²) | Weight(Kg) | Height(cm) |
|----------------|------------|--------------|------------|------------|
| 1 | 21 | 24.11 | 76.4 | 178.0 |
| 2 | 27 | 22.03 | 76.2 | 186.0 |
| 3 | 18 | 22.47 | 69.6 | 176.0 |
| 4 | 29 | 24.28 | 73.5 | 174.0 |
| 5 | 24 | 21.67 | 67.9 | 177.0 |
| 6 | 30 | 24.57 | 74.4 | 174.0 |
| 7 | 21 | 23.06 | 73.9 | 179.0 |
| 8 | 22 | 21.98 | 72.8 | 182.0 |
| 9 | 24 | 20.44 | 69.2 | 184.0 |
| 10 | 25 | 22.07 | 67.6 | 175.0 |
| 11 | 20 | 20.19 | 65.4 | 180.0 |
| 12 | 22 | 21.12 | 71.9 | 184.5 |
| Mean | 23.58 | 22.33 | 71.57 | 179.13 |
| SE^2 | 1.05 | 0.42 | 1.03 | 1.21 |

BMI = Body Mass Index

5.2 Blood Glucose Response

5.2.1 Experiment 1. Acute effects of navy bean powder on postprandial blood glucose and subjective appetite in healthy young men

a. Absolute Blood Glucose (BG)

Pre-meal mean BG (0, 15, 30, 45, 60, 90 and 120 min) was significantly affected by time (P<0.0001) and treatment by time interaction (P<0.0001), but not by treatment (P=0.2848) (**Table 5.4**). BG was lowest at 0 min and increased after treatment consumption until 30 min (**Figure 5.1A**). BG at 15 min was suppressed by whole navy beans (WB) (P=0.0038) and pureed navy beans (PB) (P=0.0167) compared to whole wheat flour (WF). BG peaked at 30 min and was suppressed by all navy bean treatments compared to WF (P_{WB}<0.0001, P_{PB}<0.0001, P_{PB}=0.0023). After the peak, BG following WF dropped more rapidly compared to all navy bean treatments. At 60 min, BG following WF was lower compared to whole navy beans (P=0.0278), which accounts for the treatment by time interaction (**Figure 5.1A**).

There was an effect of time (P<0.0001) and treatment (P=0.0173) but no effect of treatment by time interaction (P=0.2516) on post-meal BG (120, 140, 155, 170, 185 and 200 min) (**Table 5.4**). Over the entire post-meal period, the whole navy bean treatment suppressed BG compared to WF (P=0.0142), while pureed navy beans and navy bean powder led to intermediate BG. BG was lowest at 120 min, increased after pizza meal consumption and peaked at 155 min (**Figure 5.1A**). After the peak, BG declined over time without reaching baseline until 200 min.

b. Change from Baseline Blood Glucose (ΔBG)

Pre-meal mean ΔBG (15, 30, 45, 60, 90 and 120 min) was significantly affected by time (P<0.0001), treatment (P=0.0007) and their interaction (P<0.0001) (**Table 5.4**). Over the entire pre-meal period, ΔBG was suppressed following whole navy beans and navy bean powder compared to whole wheat flour (P_{WB}=0.0236, P_{BP}=0.0004). After treatment consumption, ΔBG after pureed navy beans, navy bean powder and whole wheat flour increased and peaked at 30 min, while ΔBG following whole navy beans peaked at 45 min (**Figure 5.1B**). ΔBG at 15 min

and 30 min was suppressed by all navy bean treatments compare to whole wheat flour (15 min: P_{WB} =0.001, P_{PB} =0.005, P_{BP} =0.049; 30 min: P_{WB} <0.0001, P_{PB} <0.0002, P_{BP} <0.0003). ΔBG at 45 min was suppressed by navy bean powder compare to pureed navy beans and whole wheat flour (P_{PB} =0.0169, P_{WF} =0.0219). After the peak, ΔBG declined overtime and was close to baseline at 90 and 120 min.

There was an effect of time (P<0.0001) and treatment (P = 0.002) but no effect of treatment by time interaction (P=0.5624) on post-meal Δ BG (140, 155, 170, 185 and 200 min) (**Table 5.4**). Over the entire post-meal period, whole and pureed navy bean treatment suppressed Δ BG compared to whole wheat flour (P_{WB}=0.0022, P_{PB}=0.0469), while navy bean powder led to intermediate Δ BG (**Figure 5.1B**). Mean Δ BG increased after pizza meal consumption and peaked at 155 min. After the peak, Δ BG declined over time until 200 min (**Figure 5.1B**).

c. Blood Glucose Net Area Under the Curve (AUC)

There was a treatment effect on pre-meal BG AUC (P=0.0286) where navy bean powder suppressed BG AUC compared to whole wheat flour (P=0.0163) (**Table 5.4**). Post-meal AUC was significantly affected by treatment (P=0.0170) where whole navy beans lowered BG AUC compared to whole wheat flour (P=0.0163). There were no other differences between any treatments on pre- or post-meal BG AUC.

TABLE 5.4 Experiments 1/Navy bean: Overall absolute blood glucose (BG), change from baseline blood glucose (Δ BG), blood glucose net area under the curve (BG AUC) for the pre- and post-meal periods¹

| | | Pre-meal ² | | Post-m | neal ³ |
|-----------------|-----------------|-----------------------|-------|----------------------|-------------------|
| | Treatment | Mean | SE | Mean | SE |
| BG ⁴ | WB ⁵ | 5.57 | 0.10 | 5.78 ^a | 0.10 |
| (mmol/L) | PB | 5.63 | 0.10 | 5.95 ^{ab} | 0.10 |
| | BP | 5.59 | 0.10 | 6.05 ab | 0.11 |
| | WF | 5.69 | 0.12 | 6.12 ^b | 0.12 |
| | P | 0.28 | 48 | 0.01 | 73 |
| ΔBG | WB | 0.97 ^a | 0.11 | 1.28 ^a | 0.10 |
| (mmol/L) | PB | 1.06 ^{ab} | 0.11 | 1.40 ^a | 0.10 |
| | BP | 0.89 ^a | 0.11 | 1.57 ^{ab} | 0.12 |
| | WF | 1.23 ^b | 0.14 | 1.71 ^b | 0.13 |
| | P | 0.00 | 07 | 0.00 | 21 |
| BG AUC | WB | 97.18 ^{ab} | 11.27 | 94.51 ^a | 10.07 |
| (mmol*min/L) | PB | 102.09 ab | 10.79 | 101.96 ab | 10.35 |
| | BP | 82.94 ^a | 11.43 | 114.49 ^{ab} | 11.51 |
| | WF | 115.99 ^b | 12.77 | 123.93 ^b | 15.35 |
| | P | 0.02 | 86 | 0.01 | 64 |

¹ All values are mean \pm SEM (n=17).

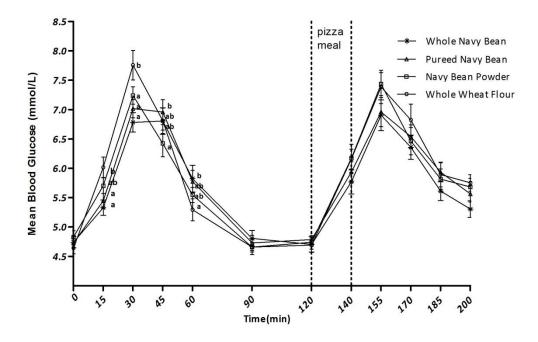
² Pre-meal values are means of all observations before the pizza meal: 0, 15, 30, 45, 60, 90 and 120 min.

³ Post-meal values are means of all observations after the pizza meal: 120, 140, 155, 170, 185 and 200 min.

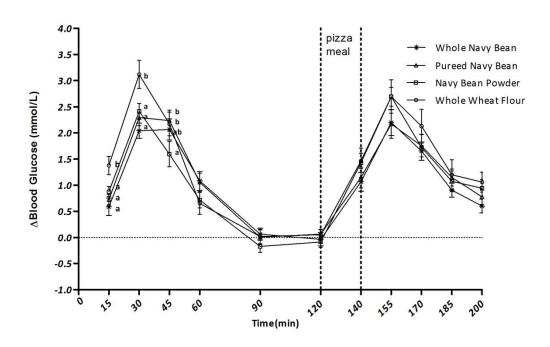
 $^{^4}$ Means in the same column with different superscript letters are significantly different from each other (two-way ANOVA, Tukey-Kramer post hoc test, P < 0.05).

⁵ WB, whole navy bean; PB, pureed navy bean; BP, navy bean powder; WF, whole wheat flour.

FIGURE 5.1 Experiment 1/ Navy bean: Effect of treatments on absolute blood glucose over time. A) blood glucose. B) change from baseline blood glucose. Means with different superscripts are significantly different at each measured time (one-way ANOVA, Tukey-Kramer post hoc test, P < 0.05). All values are mean \pm SEM.



B.



5.2.2 Experiment 2. Acute effects of lentil powder on postprandial blood glucose and subjective appetite in healthy young men

a. Absolute Blood Glucose (BG)

Pre-meal mean BG was significantly affected by time (P<0.0001), treatment (P=0.0001) and treatment by time interaction (P=0.0008) (**Table 5.5**). Over the entire pre-meal period, BG was lower following all lentil treatments compared to whole wheat flour (P_{WL} <0.0001, P_{PL} =0.0066, P_{LP} =0.0091). Pre-meal BG was lowest at 0 min and increased after treatment consumption (**Figure 5.2A**). BG at 15 min was suppressed by all lentil treatments compared to whole wheat flour (P_{WL} =0.0001, P_{PL} =0.0034, P_{LP} =0.0475). Among lentil treatments, BG at 15 min was lower following whole lentils compared to lentil powder (P=0.0389). BG peaked at 30 min and was suppressed by whole lentils compared to whole wheat flour (P=0.0272). After the peak, BG in all treatments declined over time. At 90 min, BG was suppressed by lentil powder compared to whole wheat flour (P=0.0096) (**Figure 5.2A**).

There was an effect of time (P<0.0001), but no treatment (P=0.6450) or treatment by time interaction (P=0.4384) on post-meal BG (**Table 5.5**). Mean BG increased after the pizza meal, peaked at 155 min, and declined over time without reaching baseline until 200 min, regardless of the treatments (**Figure 5.2B**).

b. Change from Baseline Blood Glucose (ΔBG)

Pre-meal mean ΔBG was significantly affected by time (P<0.0001), treatment (P=0.0083) and treatment by time interaction (P=0.0037) (**Table 5.5**). Over the entire pre-meal period, ΔBG was suppressed following whole lentils and lentil powder compared to whole wheat flour (P_{WL}=0.0094, P_{LP}=0.0273). A trend towards a smaller ΔBG after pureed lentils compared to whole wheat flour (P=0.0577) was also observed. ΔBG at 15 min and 30 min was suppressed by whole lentils compares to whole wheat flour (P₁₅=0.0059, P₃₀=0.0474) (**Figure 5.2B**). After the peak at 30 min, ΔBG after all treatments declined overtime and returned to baseline at 90 and 120 min.

Post-meal mean ΔBG was affected by time (P<0.0001), but not treatment (P=0.0851) or treatment by time interaction (P=0.5164) (**Table 5.5**). Mean ΔBG peaked at 155 min and declined over time until 200 min (**Figure 5.2B**).

c. Blood Glucose Net Area Under the Curve (AUC)

There was a treatment effect on pre-meal BG AUC (P=0.0210) (**Table 5.5**). Whole lentils and lentil powder lowered pre-meal BG AUC compared to whole wheat flour (P_{WL} =0.0379, P_{LP} =0.0250). There was no treatment effect on post-meal AUC (P=0.1055).

TABLE 5.5 Experiments 2/ Lentil: Overall absolute blood glucose (BG), change from baseline blood glucose (Δ BG), blood glucose net area under the curve (BG AUC) for the pre- and post-meal periods¹

| | | Pre-m | eal ² | Post-n | neal 3 |
|----------------------|-----------|----------------------|------------------|--------|--------|
| | Treatment | Mean | SE | Mean | SE |
| BG^4 | WL^5 | 5.54 ^a | 0.10 | 6.09 | 0.12 |
| (mmol/L) | PL | 5.64 ^a | 0.11 | 6.04 | 0.12 |
| | LP | 5.65 ^a | 0.12 | 6.09 | 0.13 |
| | WF | 5.93 ^b | 0.14 | 6.19 | 0.14 |
| | P | 0.00 | 01 | 0.6450 | |
| $\Delta \mathrm{BG}$ | WL^5 | 1.00 ^a | 0.11 | 1.45 | 0.14 |
| (mmol/L) | PL | 1.03 ^{ab} | 0.12 | 1.34 | 0.13 |
| | LP | 1.01 ^a | 0.13 | 1.54 | 0.15 |
| | WF | 1.27 ^b | 0.15 | 1.69 | 0.14 |
| | P | 0.00 | 83 | 0.0851 | |
| BG AUC | WL^5 | 97.06 ^a | 10.14 | 108.31 | 15.98 |
| (mmol*min/L) | PL | 102.44 ^{ab} | 8.37 | 94.23 | 10.44 |
| | LP | 95.75 ^a | 9.55 | 113.40 | 17.10 |
| | WF | 125.81 ^b | 14.95 | 125.50 | 16.44 |
| | P | 0.02 | 10 | 0.10 |)55 |

¹ All values are mean \pm SEM (n=12).

² Pre-meal values are means of all observations before pizza meal: 0, 15, 30, 45, 60, 90 and 120 min.

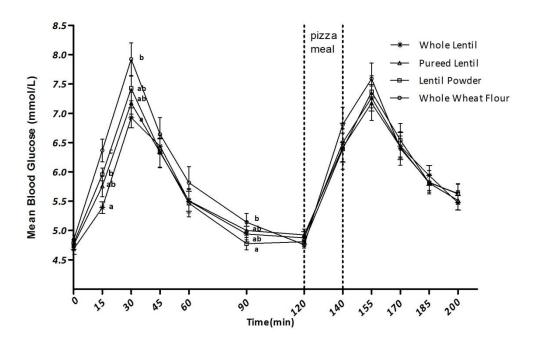
³ Post-meal values are means of all observations after pizza meal: 120, 140, 155, 170, 185 and 200 min.

 $^{^4}$ Means in the same column with different superscript letters are significantly different from each other (two-way ANOVA, Tukey-Kramer post hoc test, P < 0.05).

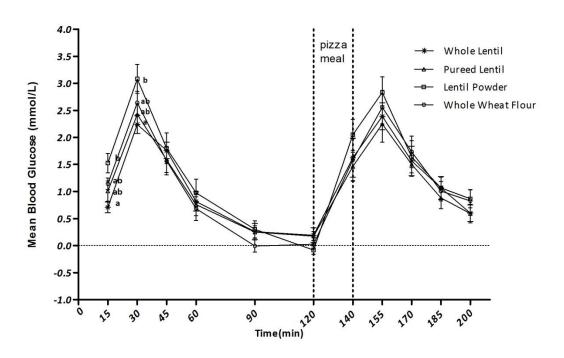
⁵ WL, whole lentil; PL, pureed lentil; LP, lentil powder; WF, wheat flour.

FIGURE 5.2 Experiment 2/ Lentil: Effect of treatments on absolute blood glucose over time. A) blood glucose. B) change from baseline blood glucose. Means with different superscripts are significantly different at each measured time (one-way ANOVA, Tukey-Kramer post hoc test, P < 0.05). All values are mean \pm SEM.

A.



B.



5.2.3 Experiment 3. Acute effects of chickpea powder on postprandial blood glucose and subjective appetite in healthy young men

a. Absolute Blood Glucose (BG)

Pre-meal mean BG was significantly affected by time (P<0.0001), treatment (P=0.0016) and treatment by time interaction (P<0.0001) (**Table 5.6**). Over the entire pre-meal period, BG was lower following all chickpea treatments compared to whole wheat flour (P_{WC} =0.0399, P_{PC} =0.0011, P_{CP} =0.0253). Pre-meal BG was lowest at 0 min and increased after treatment consumption (**Figure 5.3A**). BG at 15 min and 30 min was suppressed by whole and pureed chickpeas compare to whole wheat flour (15min: P_{WC} =0.0034, P_{PC} =0.0065; 30min: P_{WC} =0.0007, P_{PC} =0.0038). There was a trend towards lower BG at 15 min after chickpea powder compared to whole wheat flour (P=0.0572). After the peak, BG declined over time until 120 min (**Figure 5.3A**).

There was an effect of time (P<0.0001), but not treatment (P=0.6662) or treatment by time interaction (P=0.3617) on post-meal BG (**Table 5.6**). Mean BG was lowest at 120 min, increased after fixed pizza meal consumption and peaked at 155 min, regardless of treatment (**Figure 5.3A**). After the peak, BG declined over time, reaching baseline by 200 min.

b. Change from Baseline Blood Glucose (ΔBG)

Pre-meal mean ΔBG was significantly affected by time (P<0.0001) and treatment by time interaction (P<0.0001) but not by treatment (P=0.3042) (**Table 5.6**). ΔBG at 15 min was suppressed by whole and pureed chickpea treatments (P_{WC}=0.0044, P_{PL}=0.0258) compared to whole wheat flour (**Figure 5.3B**). ΔBG at 30 min was lower after the whole chickpea treatment compared to whole wheat flour (P_{WF}=0.0013) and chickpea powder (P_{LP}=0.0176), and was also lower after pureed chickpeas compared to whole wheat flour (Pw=0.0349). After peak ΔBG at 30 min, ΔBG declined overtime and returned to baseline at 90 and 120 min.

There was an effect of time (P<0.0001) but no effect of treatment (P=0.1503) or treatment by time interaction (P=0.5853) on post-meal mean Δ BG (**Table 5.6**). Mean Δ BG peaked at 155 min and declined over time until 200 min (**Figure 5.3B**).

c. Blood Glucose Net Area Under the Curve (AUC)

There was no effect of treatment on pre-meal BG AUC (P=0.5686) or post-meal BG AUC (P=0.1934) (Table 5.6).

TABLE 5.6 Experiment 3/ Chickpea: Overall absolute blood glucose (BG), change from baseline blood glucose (Δ BG), blood glucose net area under the curve (BG AUC) for the pre- and post-meal periods¹

| | | Pre-m | neal ² | Post-r | meal ³ |
|-----------------|-----------|-------------------|-------------------|--------|-------------------|
| | Treatment | Mean | SE | Mean | SE |
| BG^4 | WC^5 | 5.83 ^a | 0.11 | 6.11 | 0.12 |
| (mmol/L) | PC | 5.71 ^a | 0.12 | 6.08 | 0.14 |
| | CP | 5.80 ^a | 0.12 | 6.07 | 0.11 |
| | WF | 6.04 ^b | 0.14 | 6.18 | 0.12 |
| | P | 0.00 | 016 | 0.6662 | |
| ΔBG | WC^5 | 1.08 | 0.12 | 1.23 | 0.13 |
| (mmol/L) | PC | 1.05 | 0.12 | 1.49 | 0.16 |
| | CP | 1.13 | 0.12 | 1.48 | 0.11 |
| | WF | 1.25 | 0.15 | 1.76 | 0.12 |
| | P | 0.30 |)42 | 0.1503 | |
| BG AUC | WC^5 | 112.19 | 11.43 | 91.56 | 14.77 |
| (mmol*min/L) | PC | 105.50 | 7.95 | 111.63 | 19.89 |
| | CP | 110.88 | 10.48 | 108.00 | 12.26 |
| | WF | 122.25 | 12.65 | 127.35 | 13.01 |
| | P | 0.56 | 586 | 0.19 | 934 |

All values are mean \pm SEM (n=12).

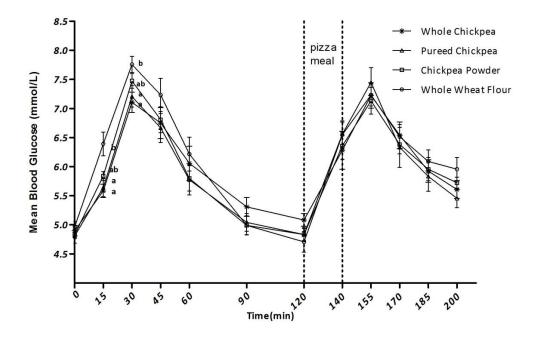
² Pre-meal values are means of all observations before pizza meal: 0, 15, 30, 45, 60, 90 and 120 min.

³ Post-meal values are means of all observations after pizza meal: 120, 140, 155, 170, 185 and 200 min.

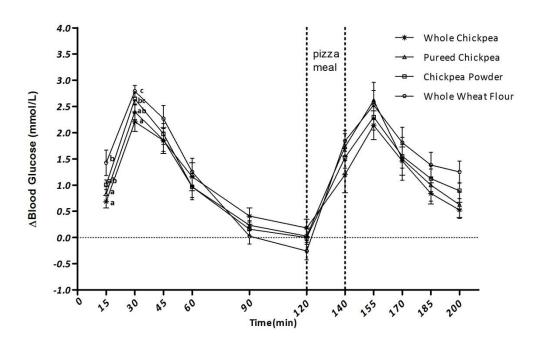
⁴ Means in the same column with different superscript letters are significantly different from each other (two-way ANOVA, Tukey-Kramer post hoc pizza, P < 0.05).

⁵ WC, whole chickpea; PC, pureed chickpea; CP, chickpea powder; WF, wheat flour.

FIGURE 5.3 Experiment 3/ Chickpea: Effect of treatments on absolute blood glucose over time. A) blood glucose. B) change from baseline blood glucose. Means with different superscripts are significantly different at each measured time (one-way ANOVA, Tukey-Kramer post hoc test, P < 0.05). All values are mean \pm SEM.



B.



5.3 Subjective Appetite

5.3.1 Experiment 1/Navy Bean

a. Subjective Appetite (App)

Pre-meal mean App was significantly affected by time (P<0.0001) and treatment (P=0.0314), but not by their interaction (P=0.1912) (**Table 5.7**). Over the entire pre-meal period, App was lower following navy bean powder compared to whole wheat flour (P=0.0293). Pre-meal App was highest at 0 min and decreased after treatment consumption and gradually rose until the pizza meal (**Figure 5.4A**). Tukey-Kramer's post-hoc test did not reveal any differences between treatments at any time points.

Post-meal mean App was significantly affected by time (P<0.0001), but not by treatment (P=0.2643) or their interaction (P=0.08325) (**Table 5.7**). Mean App declined after pizza meal consumption, reached its lowest level at 140 min and gradually increased until 200 min, regardless of treatments (**Figure 5.4A**). There was no difference between any of the treatments at any time.

b. Change from Baseline Subjective Appetite (Δ App)

Pre-meal Δ App was significantly affected by time (P<0.0001) but not treatment (P=0.7316) or their interaction (P=0.2196) (**Table 5.7**). Δ App was lowest at 15 min and gradually increased until 120 min, regardless of the treatments (**Figure 5.4B**).

Post-meal Δ App was significantly affected by time (P<0.0001) and treatment (P=0.0283) but not their interaction (P=0.8134) (**Table 5.7**). Although there was a significant treatment effect, Tukey-Kramer's post-hoc test did not reveal any differences between treatments. Mean Δ App declined after fixed pizza meal consumption, reached its lowest level at 140 min and gradually increased until 200 min, regardless of treatments (**Figure 5.4B**). There was no difference between any of the treatments at any time.

c. Subjective Appetite Area Under the Curve (App AUC)

There was no treatment effect on pre-meal App AUC (P=0.5633) or post-meal App AUC (P=0.0565) (Table 5.7).

TABLE 5.7 Experiment 1/ Navy bean: Overall mean subjective appetite score (App), change from baseline appetite score (Δ App), appetite net area under the curve (App AUC) for the pre- and post-meal periods¹

| | | Pre-m | eal ² | Post-r | neal ³ |
|------------------|-----------------|--------------------|------------------|---------|-------------------|
| | Treatment | Mean | SE | Mean | SE |
| App ⁴ | WB ⁵ | 38.40 ab | 2.07 | 27.19 | 1.73 |
| (mm) | PB | 40.81 ab | 2.18 | 27.66 | 2.10 |
| | BP | 37.32 ^a | 2.06 | 29.66 | 1.77 |
| | WF | 45.79 ^b | 2.21 | 31.65 | 2.09 |
| | P | 0.03 | 14 | 0.26 | 543 |
| ΔΑpp | WB | -34.33 | 2.73 | -26.63 | 2.01 |
| (mm) | PB | -30.91 | 2.24 | -36.05 | 2.30 |
| | BP | -35.83 | 2.23 | -23.75 | 1.80 |
| | WF | -32.21 | 2.43 | -33.62 | 2.71 |
| | P | 0.73 | 16 | 0.02 | 283 |
| App AUC | WB | -3591.3 | 619.34 | -1848.8 | 282.27 |
| (mm*min) | PB | -3203.1 | 470.83 | -2418.1 | 290.13 |
| | BP | -3848.1 | 521.71 | -1683.1 | 235.32 |
| | WF | -3194.2 | 434.33 | -2272.0 | 363.14 |
| | P | 0.56 | 83 | 0.07 | 741 |

¹ All values are mean \pm SEM (n=17)

² Pre-meal values are means of all observations before pizza meal: 0, 15, 30, 45, 60, 90 and 120 min.

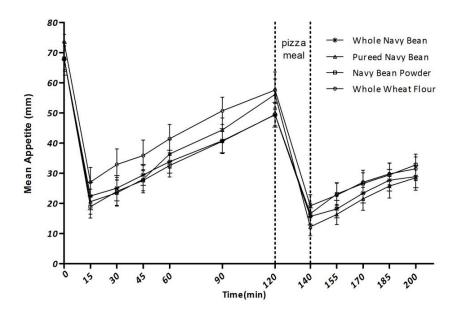
³ Post-meal values are means of all observations after pizza meal: 120, 140, 155, 170, 185 and 200 min.

 $^{^4}$ Means in the same column with different superscript letters are significantly different from each other (two-way ANOVA, Tukey-Kramer post hoc test, P < 0.05).

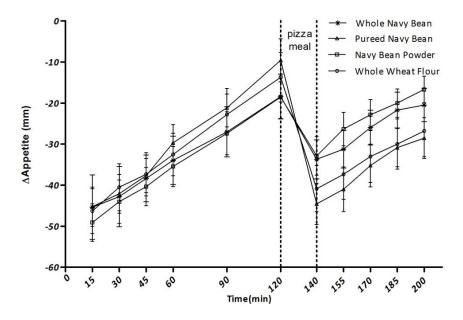
⁵ WB, whole navy bean; PB, pureed navy bean; BP, navy bean powder; WF, whole wheat flour.

Figure 5.4. Experiment 1/ Navy bean: Effect of treatments on subjective appetite score over time. A) subjective appetite score. B) change from baseline subjective appetite. All values are mean \pm SEM.

A.



B.



5.3.2 Experiment 2/ Lentil

a. Subjective Appetite (App)

Pre-meal mean App was significantly affected by time (P<0.0001), but not by treatment (P=0.3237) or their interaction (P=0.9024) (**Table 5.8**). Pre-meal App was highest at 0 min, decreased after treatment consumption and gradually rose up to the pizza meal (**Figure 5.5A**). There was an effect of time (P<0.0001), but no treatment (P=0.8305) or treatment by time interaction (P=0.7675) on post-meal App (**Table 5.8**). Mean App declined over time after fixed pizza meal consumption and increased until 200 min regardless of treatment (**Figure 5.5A**). There was no difference between any of the treatments at any time pre meal or post meal.

b. Change from Baseline Subjective Appetite (ΔApp)

Pre-meal Δ App was significantly affected by time (P=0.0039) but not treatment (P=0.8075) or their interaction (P=0.5112) (**Table 5.8**). Δ App was lowest at 15 min and gradually increased until 120 min, regardless of treatment (**Figure 5.5B**).

Post-meal Δ App was significantly affected by time (P<0.0001) and treatment (P=0.0434) but not their interaction (P=0.8450) (**Table 5.8**). Although there was a significant treatment effect, Tukey-Kramer's post-hoc test did not reveal any differences between treatments (**Figure 5.5B**). Mean Δ App declined after fixed pizza meal consumption, reached its lowest level at 140 min and gradually increased until 200 min, regardless of treatment.

c. Subjective Appetite Area Under the Curve (App AUC)

There was no treatment effect on pre-meal App AUC (P=0.0842) or post-meal App AUC (P=0.8698) (**Table 5.8**).

TABLE 5.8 Experiment 2/ Lentil: Overall mean subjective appetite score (App), change from baseline appetite score (Δ App), appetite net area under the curve (App AUC) for the pre- and post-meal periods 1

| | | Pre-m | neal ² | Post-n | neal ³ |
|--------------|-----------------|----------|-------------------|----------|-------------------|
| | Treatment | Mean | SE | Mean | SE |
| App | WL^4 | 50.48 | 2.06 | 30.50 | 2.64 |
| (mm) | PL | 54.54 | 2.40 | 33.17 | 2.59 |
| | LP | 48.77 | 2.46 | 28.90 | 2.62 |
| | WF | 52.31 | 2.25 | 32.35 | 2.67 |
| | P | 0.27 | 773 | 0.3237 | |
| ΔApp | WL^4 | -18.83 | 2.61 | -41.23 | 3.30 |
| (mm) | PL | -26.44 | 2.92 | -40.88 | 2.55 |
| | LP | -29.09 | 2.88 | -44.11 | 3.08 |
| | WF | -23.78 | 2.94 | -38.23 | 2.49 |
| | P | 0.80 |)75 | 0.0851 | |
| App AUC | WL^4 | -1923.28 | 518.76 | -2819.01 | 462.4 |
| (mm*min) | PL | -2742.81 | 668.66 | -2916.46 | 377.3 |
| | LP | -3127.81 | 648.29 | -3002.55 | 456.2 |
| | WF | -2590.31 | 767.81 | -2669.85 | 361.6 |
| | P | 0.08 | 342 | 0.85 | 598 |

All values are mean \pm SEM (n=12)

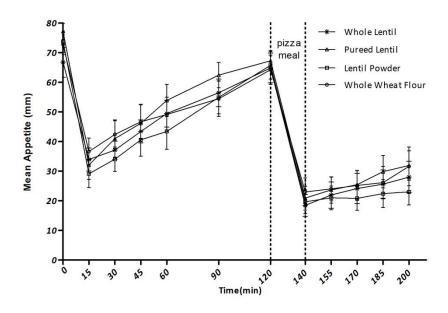
² Pre-meal values are means of all observations before pizza meal: 0, 15, 30, 45, 60, 90 and 120 min.

³ Post-meal values are means of all observations after pizza meal: 120, 140, 155, 170, 185 and 200 min.

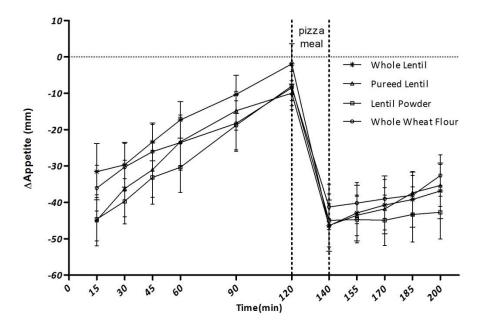
⁴ WL, whole lentil; PL, pureed lentil; LP, lentil powder; WF, wheat flour.

FIGURE 5.5 Experiment 2/ Lentil: Effect of treatments on subjective appetite score over time. A) subjective appetite score. B) change from baseline subjective appetite. All values are mean \pm SEM.

A.



B.



5.3.3 Experiment 3/ Chickpea

a. Subjective Appetite (App)

Pre-meal mean App was significantly affected by time (P<0.0001), but not treatment (P=0.5552) or treatment by time interaction (P=0.8913) (**Table 5.9**). Pre-meal App was highest at 0 min, decreased after treatment consumption and gradually rose until pizza meal (**Figure 5.6A**). There was an effect of time (P<0.0001), but no effect of treatment (P=0.3009) or treatment by time interaction (P=0.5394) on post-meal App (**Table 5.9**). Mean App declined because of pizza consumption and inclined after 140 min until the end of the study, regardless of treatment (**Figure 5.6A**). There was no difference between any of the treatments at any time.

b. Change from Baseline Subjective Appetite (ΔApp)

Pre-meal Δ App was significantly affected by time (P<0.0001) but not treatment (P=0.4033) or their interaction (P=0.8826) (**Table 5.9**). Δ App was lowest at 15 min and gradually increased until 120 min, regardless of treatment (**Figure 5.6B**).

Post-meal Δ App was not affected by time (P=0.1926), treatment (P=0.2201) or their interaction (P=0.6978) (**Table 5.9**). Mean Δ App declined after pizza meal consumption reached its lowest level at 140 min and gradually increased until 200 min, regardless of treatment (**Figure 5.6B**).

c. Subjective Appetite Area Under the Curve (App AUC)

There was no treatment effect on pre-meal (P=0.3154) or post-meal App AUC (P=0.3390) (Table 5.9).

TABLE 5.9 Experiment 1/ Navy bean: Overall mean subjective appetite score (App), change from baseline appetite score (Δ App), appetite net area under the curve (App AUC) for the pre- and post-meal periods¹

| | | Pre-meal ² | | Post-n | neal ³ |
|----------|-----------------|-----------------------|--------|----------|-------------------|
| | Treatment | Mean | SE | Mean | SE |
| App | WC^4 | 48.93 | 1.99 | 33.35 | 2.33 |
| (mm) | PC | 53.05 | 2.12 | 34.60 | 2.50 |
| | CP | 48.13 | 2.25 | 31.71 | 2.38 |
| | WF | 49.28 | 2.28 | 34.58 | 2.37 |
| | P | 0.55 | 552 | 0.30 | 009 |
| ΔΑρρ | WC^4 | -18.56 | 2.36 | -37.71 | 2.40 |
| (mm) | PC | -15.44 | 2.65 | -40.43 | 2.64 |
| | CP | -22.40 | 2.24 | -37.05 | 2.52 |
| | WF | -22.59 | 2.01 | -33.78 | 2.54 |
| | P | 0.40 | 033 | 0.22 | 201 |
| App AUC | WC^4 | -1977.35 | 507.01 | -2671.31 | 334.12 |
| (mm*min) | PC | -1645.94 | 617.26 | -2857.97 | 408.51 |
| | CP | -2452.82 | 461.51 | -2600.47 | 378.27 |
| | WF | -2425.32 | 395.23 | -2377.56 | 377.64 |
| | P | 0.3390 | | 0.55 | 536 |

¹ All values are mean \pm SEM (n=12)

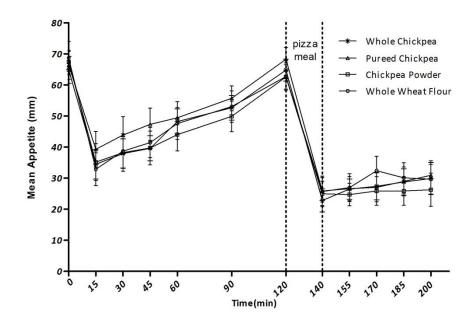
² Pre-meal values are means of all observations before pizza meal: 0, 15, 30, 45, 60, 90 and 120 min.

³ Post-meal values are means of all observations after pizza meal: 120, 140, 155, 170, 185 and 200 min.

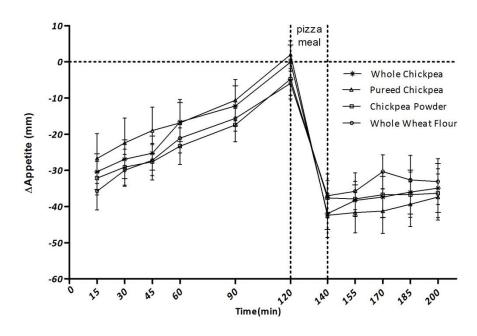
⁴ WC, whole chickpea; PC, pureed chickpea; CP, chickpea powder; WF, wheat flour.

FIGURE 5.6 Experiment 3/ Chickpea: Effect of treatments on subjective appetite score over time. A) subjective appetite score. B) change from baseline subjective appetite. All values are mean \pm SEM.

A.



B.



5.4 Thirst and Water Intake

5.4.1 Experiment 1/ Navy Bean

Overall subjective thirst was affected by time (P<0.0001), but not treatment (P=0.1790) or time by treatment interaction (P=0.2681) (**Table 5.10a**). Subjective thirst was highest at 0 min, decreased after treatment and water consumption at 15 min (**Figure 5.7**). After 15 min, thirst score gradually rose with until the pizza meal. Thirst score decreased after pizza and water consumption at 140 min and gradually increased until the end of the study (**Figure 5.7**).

Treatment did not affect water intake at the pizza meal (P=0.5633) (**Table 5.10b**).

TABLE 5.10a Experiment 1/ Navy bean: Overall mean thirst¹

| | Thirst (mm) | | | |
|-----------|-------------|------|--|--|
| Treatment | Mean | SE | | |
| WB^2 | 36.26 | 1.62 | | |
| PB | 37.77 | 1.72 | | |
| BP | 35.02 | 1.65 | | |
| WF | 30.98 1.66 | | | |
| P | 0.2681 | | | |

¹ All values are mean \pm SEM (n=1 $\overline{7}$).

TABLE 5.10b Experiment 1/ Navy bean: Water intake¹

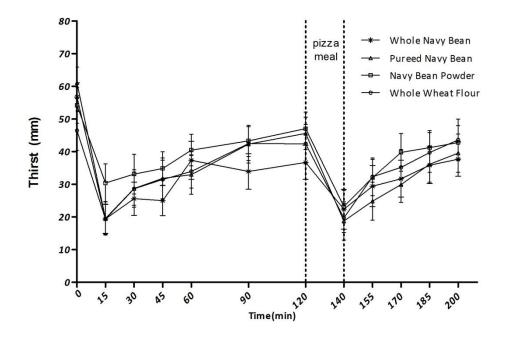
| | Water Intake (ml) | | | |
|-----------|-------------------|-------|--|--|
| Treatment | Mean | SE | | |
| WB^2 | 368.90 | 28.31 | | |
| PB | 332.06 | 23.68 | | |
| BP | 343.31 | 27.23 | | |
| WF | 342.57 | 34.77 | | |
| P | 0.5 | 633 | | |

¹ All values are mean \pm SEM (n=1 $\overline{7}$).

² WB, whole navy bean; PB, pureed navy bean; BP, navy bean powder; WF, whole wheat flour.

² WB, whole navy bean; PB, pureed navy bean; BP, navy bean powder; WF, whole wheat flour.

FIGURE 5.7 Experiment 1/ Navy bean: Effect of treatments on thirst over time. All values are mean \pm SEM



5.4.2 Experiment 2/ Lentil

Overall subjective thirst was affected by time (P<0.0001), but not treatment (P=0.7498) or time by treatment interaction (P=0.3863) (**Table 5.11a**). Subjective thirst decreased after treatment and water consumption and gradually rose until the pizza meal (**Figure 5.8**). Subjective thirst score decreased after pizza and water consumption at 140 min and gradually increased until the end of the study (**Figure 5.8**).

Treatment did not affect water intake at the pizza meal (P=0.8600) (Table 5.11b).

TABLE 5.11a Experiment 2/ Lentil: Overall mean thirst¹

| | Thirst (mm) | | | |
|-----------|-------------|------|--|--|
| Treatment | Mean | SE | | |
| WL^5 | 39.37 | 1.87 | | |
| PL | 38.85 | 2.06 | | |
| LP | 36.12 | 2.04 | | |
| WF | 39.44 | 2.00 | | |
| Р | 0. 7498 | | | |

¹ All values are mean \pm SEM (n=12).

TABLE 5.11b Experiment 2/ Lentil: Water intake¹

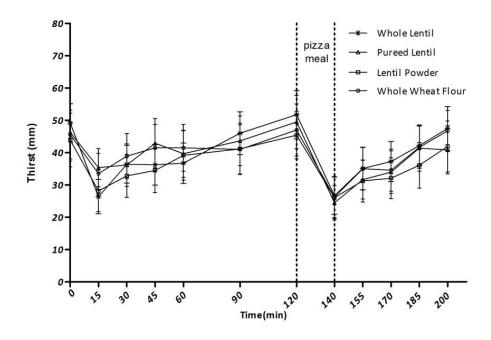
| | Water Intake ² (ml) | | | |
|-----------|--------------------------------|-------|--|--|
| Treatment | Mean | SE | | |
| WL^5 | 337.78 | 36.40 | | |
| PL | 350.93 | 39.43 | | |
| LP | 341.33 | 33.19 | | |
| WF | 323.37 | 33.16 | | |
| P | 0.86 | 500 | | |

¹ All values are mean \pm SEM (n=12).

² WL, whole lentil; PL, pureed lentil; LP, lentil powder; WF, wheat flour.

²WL, whole lentil; PL, pureed lentil; LP, lentil powder; WF, wheat flour.

FIGURE 5.8 Experiment 2/ Lentil: Effect of treatments on thirst over time. All values are $mean \pm SEM$



5.4.3 Expriment 3/ Chickpea

Overall subjective thirst was affected by time (P<0.0001), but not treatment (P=0.3445) or time by treatment interaction (P=0.7829) (**Table 5.12a**). Subjective thirst decreased after treatment and water consumption and gradually rose with until the pizza meal (**Figure 5.9**). Thirst score decreased after pizza and water consumption at 140 min and gradually increased until the end of the study (**Figure 5.9**).

Treatment did not affect water intake at the pizza meal (P=0.5180) (Table 5.12b).

TABLE 5.12a Experiment 3/ Chickpea: Overall mean thirst¹

| | Thirst (mm) | | | | | |
|-----------|-------------|------|--|--|--|--|
| Treatment | Mean SE | | | | | |
| WC^2 | 36.10 | 2.30 | | | | |
| PC | 36.55 | 2.43 | | | | |
| CP | 34.74 | 2.41 | | | | |
| WF | 35.01 2.29 | | | | | |
| P | 0.3445 | | | | | |

¹ All values are mean \pm SEM (n=12).

TABLE 5.12b Experiment 3/ Chickpea: Water intake¹

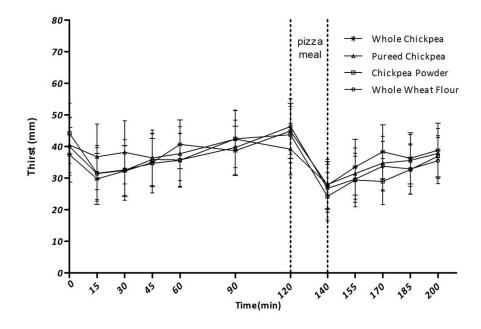
| | Water Intake ² (ml) | | | |
|-----------------|--------------------------------|-------|--|--|
| Treatment | Mean SE | | | |
| WC ⁵ | 351.87 | 41.43 | | |
| PC | 345.23 41.53 | | | |
| CP | 321.02 40.98 | | | |
| WF | 327.42 | 33.13 | | |
| P | 0.5180 | | | |

¹ All values are mean \pm SEM (n=12).

² WC, whole chickpea; PC, pureed chickpea; CP, chickpea powder; WF, wheat flour.

² WC, whole chickpea; PC, pureed chickpea; CP, chickpea powder; WF, wheat flour.

FIGURE 5.9 Experiment 3/ Chickpea: Effect of treatments on thirst over time. All values are mean \pm SEM



5.5 Palatability

5.5.1 Experiment 1/ Navy Bean

Palatability was different among treatments (P=0.0087) (**Table 5.13a**). Subjects found the whole and pureed navy bean treatments to be more palatable than navy bean powder treatment (P_{WB} =0.0147, P_{PB} =0.0154). However, the treatments did not affect the palatability of the pizza meal (P=0.6111) (**Table 5.13a**).

The pleasantness of the treatments varied (P=0.0045) (**Table 5.13b**). Subjects found that the whole and pureed navy bean treatments were more pleasant compared to navy bean powder treatment (P_{WB} =0.0411, P_{PB} =0.0031). Tastiness was not different among the treatments (P=0.1061), but the texture was different (P=0.0087). Subject found that whole navy beans had a better texture compared to navy bean powder (P=0.0068).

TABLE 5.13a Experiment 1/ Navy bean: Treatment and pizza palatability ¹

| | Treatment | | | Pizza | | |
|---------------------------|-----------|---------------------|------|-------|------|--|
| | Treatment | Mean | SE | Mean | SE | |
| Palatability ² | WB | 55.53 ^b | 5.29 | 79.12 | 2.82 | |
| (mm) PB | PB | 55.96 ^b | 5.70 | 80.71 | 2.80 | |
| | BP | 38.98 ^a | 6.55 | 78.41 | 3.27 | |
| | WF | 49.51 ^{ab} | 5.30 | 91.12 | 2.64 | |
| | P | 0.0087 | | 0.10 | 061 | |

¹ All values are mean \pm SEM (n=17).

TABLE 5.13b Experiment 1/ Navy bean: Treatment palatability breakdown¹

| | Pleasar | Pleasant ² (mm) | | mm) Taste ² (mm) | | ² (mm) |
|-----------|--------------------|----------------------------|-------|-----------------------------|--------------------|-------------------|
| Treatment | Mean | SE | Mean | SE | Mean | SE |
| WB^3 | 57.35 ^b | 5.64 | 56.18 | 5.76 | 53.06 ^b | 6.01 |
| PB | 63.47 ^b | 6.30 | 56.12 | 5.70 | 48.29 ^b | 6.51 |
| BP | 42.24 ^a | 7.56 | 44.18 | 5.77 | 30.53 ^a | 7.66 |
| WF | 55.59 ab | 5.67 | 52.76 | 5.00 | 40.18 ab | 7.37 |
| Р | 0.0045 | | 0.10 | 061 | 0.00 | 87 |

¹ All values are mean \pm SEM (n=17).

 $^{^{2}}$ Means in the same column with different superscript letters are significantly different from each other (two-way ANOVA, Tukey-Kramer post hoc test, P < 0.05).

³ WB, whole navy bean; PB, pureed navy bean; BP, navy bean powder; WF, whole wheat flour.

 $^{^{2}}$ Means in the same column with different superscript letters are significantly different from each other (two-way ANOVA, Tukey-Kramer post hoc test, P < 0.05).

³ WB, whole navy bean; PB, pureed navy bean; BP, navy bean powder; WF, whole wheat flour.

5.5.2 Experiment 2/ Lentil

Palatability of the lentil treatments was different (P=0.0054). Whole and pureed lentils had a higher palatability score compared to whole wheat flour (P_{WL} =0.0039, P_{PL} =0.0342) (**Table 5.14a**). There was no treatment effect on the palatability of the pizza meal (P=0.9924).

Pleasantness, taste and texture were different among treatments ($P_{pleasantness}$ =0.0175, P_{taste} =0.0088, $P_{texture}$ =0.0062) (**Table 5.14b**). Subject found the whole lentil treatment to be more pleasant compared to whole wheat flour (P=0.0108); whole and pureed lentil treatments were rated tastier compared to whole wheat flour (P_{WL} =0.0164, P_{PL} =0.0108), and whole lentils had a better texture compared to lentil powder and whole wheat flour (P_{LP} =0.0220, P_{WF} =0.0048).

TABLE 5.14a Experiment 2/ Lentil: Treatment and pizza palatability¹

| | | Treatment | | Pizza | | |
|---------------------------|-----------|--------------------|------|-------|------|--|
| | Treatment | Mean | SE | Mean | SE | |
| Palatability ² | WL^3 | 57.79 ^a | 5.53 | 73.08 | 5.34 | |
| (mm) | PL | 45.67 ^a | 7.19 | 74.08 | 4.19 | |
| | LP | 40.00^{ab} | 6.93 | 74.42 | 4.8 | |
| | WF | 33.19 ^b | 7.76 | 74.33 | 4.07 | |
| | P | 0.0054 | | 0.99 | 924 | |

¹ All values are mean \pm SEM (n=12).

TABLE 5.14b Experiment 2/ Lentil: Treatment palatability breakdown¹

| | Pleasant ² (mm) | | Taste ² (1 | Taste ² (mm) | | Texture ² (mm) | |
|-----------|----------------------------|------|-----------------------|-------------------------|---------------------|---------------------------|--|
| Treatment | Mean | SE | Mean | SE | Mean | SE | |
| WL^3 | 58.36 ^b | 6.17 | 56.45 ^b | 6.90 | 58.55 ^b | 4.68 | |
| PL | 47.92 ^{ab} | 8.17 | 50.25 ^b | 7.53 | 38.83 ^{ab} | 6.45 | |
| LP | 43.42 ^{ab} | 7.30 | 43.67 ^{ab} | 7.53 | 32.92 ^a | 7.09 | |
| WF | 35.25 ^a | 8.36 | 35.83 ^a | 8.64 | 28.50 ^a | 7.17 | |
| P | 0.01 | 75 | 0.008 | 38 | 0.00 | 62 | |

¹ All values are mean \pm SEM (n=12).

 $^{^{2}}$ Means in the same column with different superscript letters are significantly different from each other (two-way ANOVA, Tukey-Kramer post hoc test, P < 0.05).

³ WL, whole lentil; PL, pureed lentil; LP, lentil powder; WF, wheat flour.

 $^{^{2}}$ Means in the same column with different superscript letters are significantly different from each other (two-way ANOVA, Tukey-Kramer post hoc test, P < 0.05).

³ WL, whole lentil; PL, pureed lentil; LP, lentil powder; WF, wheat flour.

5.5.3 Experiment 3/ Chickpea

There was no difference in treatment palatability (P=0.0919) or pizza meal palatability (P=0.2724) among treatments (**Table 5.15a**).

The pleasantness, taste and texture of the treatments were not different ($P_{pleasantness}$ =0.0847, P_{taste} =0.1186, $P_{texture}$ =0.1629) (**Table 5.15b**).

TABLE 5.15a Experiment 3/ Chickpea: Palatability of the treatment and pizza¹

| | | Trea | tment | Pizza | | |
|--------------|-----------|--------|-------|-------|------|--|
| | Treatment | Mean | SE | Mean | SE | |
| Palatability | WC^2 | 58.69 | 7.58 | 73.00 | 4.81 | |
| (mm) | PC | 58.64 | 6.01 | 75.67 | 4.58 | |
| | CP | 47.08 | 7.47 | 75.00 | 4.70 | |
| | WF | 49.44 | 7.52 | 77.58 | 4.09 | |
| | P | 0.0919 | | 0.2 | 724 | |

¹ All values are mean \pm SEM (n=12).

TABLE 5.15b Experiment 3/ Chickpea: Treatment palatability breakdown¹

| | Pleasant(mm) | | Taste(| (mm) | Textur | Texture(mm) | |
|-----------|--------------|------|--------|--------|--------|-------------|--|
| Treatment | Mean | SE | Mean | SE | Mean | SE | |
| WC^2 | 61.67 | 7.34 | 55.5 | 7.91 | 58.92 | 8.04 | |
| PC | 59.58 | 6.68 | 62.33 | 6.04 | 54.00 | 6.78 | |
| CP | 47.83 | 7.52 | 49.00 | 7.51 | 44.42 | 7.99 | |
| WF | 51.42 | 7.78 | 52.42 | 8.24 | 44.50 | 7.42 | |
| P | 0.0847 | | 0.11 | 0.1186 | | 0.1629 | |

 $^{^{\}mathsf{T}}$ All values are mean \pm SEM (n=12).

² WC, whole chickpea; PC, pureed chickpea; CP, chickpea powder; WF, wheat flour.

² WC, whole chickpea; PC, pureed chickpea; CP, chickpea powder; WF, wheat flour.

5.6. Physical Comfort, Energy and Tiredness

5.6.1 Experiment 1/Navy Bean

Overall physical comfort was not affected by time (P=0.3089), treatment (P=0.4646) or treatment by time interaction (P=0.4798) (**Table 5.16**).

Overall energy level was affected by time (P=0.0011), but not by treatment (P=0.3730) or treatment by time interaction (P=0.5743) (**Table 5.16**). Energy level was constant from 0-120 min, and then rose slightly immediately following the pizza meal.

Overall tiredness was affected by time (P=0.0032), but not treatment (P=0.0573) or treatment by time interaction (P=0.8128) (**Table 5.16**). Tiredness was constant from 0-120 min., and then dropped slightly immediately following the pizza meal.

TABLE 5.16 Experiment 1/ Navy Bean: Overall mean comfort, energy, and tiredness¹

| Comfort (mm) | | (mm) | Energy | (mm) | Tiredness (mm) | |
|--------------|--------|------|--------|------|----------------|------|
| Treatment | Mean | SE | Mean | SE | Mean | SE |
| WB^2 | 90.99 | 1.07 | 60.20 | 1.96 | 39.57 | 1.96 |
| PB | 89.29 | 1.26 | 62.48 | 1.96 | 32.04 | 2.26 |
| BP | 88.31 | 1.45 | 63.84 | 2.01 | 36.04 | 2.07 |
| WF | 90.52 | 0.92 | 64.35 | 1.95 | 33.26 | 1.96 |
| P | 0.4646 | | 0.373 | 30 | 0.0573 | |

¹ All values are mean \pm SEM (n=17).

² WB, whole navy bean; PB, pureed navy bean; BP, navy bean powder; WF, whole wheat flour.

5.6.2 Experiment 2/ Lentil

Overall physical comfort was affected by time (P=0.0007) and treatment (P=0.0049), but not their interaction (P=0.5920) (**Table 5.17**). Despite the time effect, comfort level remained high and stable throughout the session. Subjects experienced a slightly higher comfort level after pureed lentils and whole wheat flour compared to whole lentils (P_{LP} =0.0067, P_{WF} =0.0169). However, the different between the highest treatment (PL) and the lowest treatment (WL) was only 2.4 mm (**Table 5.17**).

Overall energy level was affected by time (P=0.0005), but not by treatment (P=0.3083) or treatment by time interaction (P= 0.1033) (**Table 5.17**). Energy level was constant from 0-120 min, and then rose slightly immediately following the pizza meal.

Overall tiredness was affected by time (P=0.0268), but not by treatment (P=0.5323) or treatment by time interaction (P=0.5242) (**Table 5.17**). Tiredness was constant from 0-120 min, and then dropped slightly immediately following the pizza meal.

TABLE 5.17 Experiment 2/ Lentil: Overall mean comfort, energy, and tiredness¹

| | Comfort (mm) | | Energ | Energy (mm) | | ss (mm) | |
|-----------|---------------------|------|-------|-------------|-------|---------|--|
| Treatment | Mean | SE | Mean | SE | Mean | SE | |
| WL^2 | 86.91 ^a | 1.40 | 52.06 | 1.99 | 47.93 | 2.09 | |
| PL | 89.34 ^b | 1.13 | 55.58 | 2.52 | 40.94 | 2.55 | |
| LP | 88.31 ^{ab} | 1.17 | 55.22 | 2.50 | 43.21 | 2.41 | |
| WF | 88.77 ^b | 1.17 | 56.58 | 2.33 | 44.06 | 2.41 | |
| P | 0.0007 | | 0.3 | 0.3083 | | 0.5323 | |

All values are mean \pm SEM (n=12).

 $^{^{2}}$ Means in the same column with different superscript letters are significantly different from each other (two-way ANOVA, Tukey-Kramer post hoc test, P < 0.05).

³ WL, whole lentil; PL, pureed lentil; LP, lentil powder; WF, wheat flour.

5.6.3 Experiment 3/ Chickpea

Overall physical comfort was affected by time (P=0.0491), but not by treatment (P=0.4239) or treatment by time interaction (P=0.9070) (**Table 5.18**). Despite the time effect, comfort level remained high and stable throughout the session.

Overall energy level was affected by time (P=0.0107), but not treatment (P=0.7474) or treatment by time interaction (P=0.2542) (**Table 5.18**). Energy level was constant from 0-120 min, and then rose slightly immediately following the pizza meal.

Overall tiredness was affected by time (P=0.0005), but not treatment (P=0.7084) or treatment by time interaction (P= 0.7308) (**Table 5.18**). Tiredness was constant from 0-120 min, and then dropped slightly immediately following the pizza meal.

TABLE 5.18 Experiment 3/ Chickpea: Overall mean comfort, energy, and tiredness¹

| | Comfort (mm) | | Energy | y (mm) | Tiredne | Tiredness (mm) | |
|-----------|--------------|------|--------|--------|---------|----------------|--|
| Treatment | Mean | SE | Mean | SE | Mean | SE | |
| WC^2 | 88.62 | 1.21 | 56.97 | 2.75 | 35.15 | 3.06 | |
| PC | 89.36 | 1.09 | 58.83 | 2.49 | 38.33 | 2.96 | |
| CP | 89.41 | 1.10 | 58.63 | 2.79 | 34.13 | 2.98 | |
| WF | 88.89 | 1.23 | 58.85 | 2.90 | 34.68 | 3.19 | |
| P | 0.4239 | | 0.7 | 0.7474 | | 0.7084 | |

¹ All values are mean \pm SEM (n=12).

² WC, whole chickpea; PC, pureed chickpea; CP, chickpea powder; WF, wheat flour.

5.7 Correlations between Dependent Measures

5.7.1 Experiment 1/ Navy Bean

In Experiment 1, pre-meal App AUC was negatively correlated with pre-meal Δ BG (r=-0.25252, P=0.0378) and positively correlated with the tastiness of the treatment (r=0.28022, P=0.0206) and pre-meal thirst (r= 0.24078, P=0.0479) (**Table 5.19**). Pre-meal mean App was positively correlated with the pleasantness (r=0.43955, P=0.0002), taste(r=0.49734, P<0.0001), texture (r=0.38049, P=0.0014) and palatability(r=0.47374, P<0.0001) of the treatments. Pre-meal mean Δ App was negatively correlated with pre-meal BG AUC (r=-0.27223, P=0.0247) and Δ BG (r=-0.32686, P=0.0065) and positively correlated with the pleasantness (r=0.23965, P=0.0490) and taste (r=0.29003, P=0.0164) of the treatment, and pre-meal thirst level (r=0.28093, P=0.0203)

TABLE 5.19 Experiment 1/ Navy bean: Pearson correlations between pre-meal mean App, pre-meal mean Δ App, pre-meal App AUC and dependent measurements¹

| | App AUC ³ | | App ² | | ΔApp^2 | |
|------------------------|----------------------|--------|----------------------|--------|----------------|--------|
| Correlated Variables | r | P | r | P | R | P |
| Treatment Energy | -0.20160 | 0.0992 | -0.01660 | 0.8931 | -0.19800 | 0.1055 |
| BG AUC ³ | -0.20367 | 0.0957 | -0.03366 | 0.7853 | -0.27223 | 0.0247 |
| BG^2 | -0.0882 | 0.4745 | 0.14141 | 0.2500 | -0.08718 | 0.4796 |
| ΔBG^2 | -0.25252 | 0.0378 | -0.04894 | 0.6919 | -0.32686 | 0.0065 |
| Treatment Pleasant | 0.2309 | 0.0582 | 0.43955 | 0.0002 | 0.23965 | 0.0490 |
| Treatment Taste | 0.28022 | 0.0206 | 0.49734 | <.0001 | 0.29003 | 0.0164 |
| Treatment Texture | 0.11274 | 0.3600 | 0.38049 | 0.0014 | 0.13726 | 0.2643 |
| Treatment Palatability | 0.21972 | 0.0718 | 0.47374 | <.0001 | 0.23596 | 0.0527 |
| Tiredness ² | -0.04453 | 0.7184 | -0.07444 | 0.5463 | -0.06137 | 0.6191 |
| Energy ² | 0.10405 | 0.3984 | 0.0415 | 0.7368 | 0.10647 | 0.3875 |
| Comfort ² | 0.00457 | 0.9705 | -0.1059 | 0.3901 | -0.05503 | 0.6558 |
| Thirst ² | 0.24078 | 0.0479 | 0.14228 | 0.2471 | 0.28093 | 0.0203 |

^TAll values are mean \pm SEM (n=17).

² Values are the means of pre-meal measurements.

 $^{^{3}}$ Values are the pre-meal area under the curve.

5.7.2 Experiment 2/ Lentil

In Experiment 2, pre-meal App AUC was correlated with pre-meal mean energy level (r=0.33347, P=0.0205) (**Table 5.20**). Pre-meal mean App was negatively correlated with pre-meal mean thirst (r=-0.31076, P=0.0316). Pre-meal mean Δ App was correlated with pre-meal energy level (r=-0.34436, P=0.0165).

TABLE 5.20 Experiment 2/ Lentil: Pearson correlations between pre-meal mean App, pre-meal mean Δ App, pre-meal App AUC and dependent measurements¹

| | App AUC ³ | | Apj | p ² | ΔApp^2 | |
|------------------------|----------------------|--------|----------|----------------|----------------|--------|
| Correlated Variables | r | P | r | P | r | P |
| Treatment Energy | 0.00316 | 0.9830 | 0.15080 | 0.3063 | 0.01456 | 0.9217 |
| BG AUC ³ | -0.15148 | 0.3041 | -0.04704 | 0.7509 | -0.13411 | 0.3635 |
| BG^2 | -0.1521 | 0.3021 | -0.08806 | 0.5517 | -0.13103 | 0.3747 |
| $\Delta \mathrm{BG}^2$ | -0.194 | 0.1864 | -0.0495 | 0.7383 | -0.18747 | 0.2020 |
| Treatment Pleasant | 0.12231 | 0.4128 | 0.22456 | 0.1291 | 0.12146 | 0.4161 |
| Treatment Taste | -0.03731 | 0.8034 | 0.0327 | 0.8273 | -0.03752 | 0.8023 |
| Treatment Texture | -0.03064 | 0.8380 | -0.09498 | 0.5254 | -0.02811 | 0.8512 |
| Treatment Palatability | 0.02028 | 0.8924 | 0.0609 | 0.6842 | 0.02073 | 0.8900 |
| Tiredness ² | -0.03126 | 0.8329 | 0.17887 | 0.2238 | -0.0544 | 0.7134 |
| Energy ² | 0.33347 | 0.0205 | 0.06128 | 0.6790 | 0.34436 | 0.0165 |
| Comfort ² | 0.18014 | 0.2205 | -0.00955 | 0.9486 | 0.17942 | 0.2224 |
| Thirst ² | -0.22787 | 0.1193 | -0.31076 | 0.0316 | -0.2352 | 0.1076 |

¹ All values are mean \pm SEM (n=12).

² Values are the means of pre-meal measurements.

³ Values are the pre-meal area under the curve.

5.7.3 Experiment 3/ Chickpea

In Experiment 3, pre-meal App AUC was positively correlated with pre-meal mean comfort (r=0.32132, P=0.0356) (**Table 5.21**). Pre-meal mean App was negatively correlated with pre-meal mean tiredness (r=0.56998, P<0.0001), and positively correlated with pre-meal mean energy level (r=0.5219, P=0.0003), comfort level (r=0.44796, P=0.0026) and thirst (r=0.45546, P=0.0021). Pre-meal mean Δ App was positively correlated with pre-meal comfort level (r=0.3291, P=0.0312).

TABLE 5.21 Experiment 3/ Chickpea: Pearson correlation between pre-meal mean App, pre-meal mean Δ App, pre-meal App AUC and dependent measurements¹

| | App AUC ³ | | App | App ² | | pp^2 |
|------------------------|----------------------|--------|----------|------------------|----------|--------|
| Correlated Variables | r | P | r | P | r | Р |
| Treatment Energy | 0.08367 | 0.5718 | 0.01575 | 0.9154 | 0.09347 | 0.5275 |
| BG AUC ³ | 0.04395 | 0.7796 | 0.04166 | 0.7908 | 0.01977 | 0.8999 |
| BG^2 | 0.03283 | 0.8344 | -0.08254 | 0.5988 | 0.01461 | 0.9259 |
| CBG^2 | 0.01513 | 0.9233 | -0.00193 | 0.9902 | -0.01038 | 0.9473 |
| Treatment Pleasant | -0.28795 | 0.0611 | 0.09526 | 0.5434 | -0.269 | 0.0811 |
| Treatment Taste | -0.26336 | 0.0879 | 0.02982 | 0.8494 | -0.2449 | 0.1135 |
| Treatment Texture | -0.2682 | 0.0821 | -0.07899 | 0.6146 | -0.247 | 0.1103 |
| Treatment Palatability | -0.28567 | 0.0633 | 0.01546 | 0.9216 | -0.26523 | 0.0856 |
| Tiredness ² | -0.10589 | 0.4992 | -0.56998 | <.0001 | -0.12132 | 0.4383 |
| Energy ² | -0.00128 | 0.9935 | 0.5219 | 0.0003 | 0.00563 | 0.9714 |
| Comfort ² | 0.32132 | 0.0356 | 0.44796 | 0.0026 | 0.3291 | 0.0312 |
| Thirst ² | 0.02301 | 0.8836 | 0.45546 | 0.0021 | 0.03183 | 0.8394 |

All values are mean \pm SEM (n=12).

² Values are the means of pre-meal measurements.

³ Values are the pre-meal area under the curve.

CHAPTER 6

DISCUSSION

6 DISCUSSION

The present findings do not support the hypothesis that powdered pulses are less effective than canned whole pulses at suppressing postprandial blood glucose and subjective appetite. No differences in blood glucose response and subjective appetite scores were observed between pulse treatments varying in processing type (whole canned pulses, pureed canned pulses and pulse powder), indicating that lentil, navy bean and chickpea powders can similarly contribute to the regulation of short-term glycaemia and appetite.

Industrial processing of navy beans, lentils and chickpeas to powdered forms did not negatively influence the known beneficial effects of canned pulses on blood glucose control (34-37). The pre-cooked pulse powders resulted in similar blood glucose responses compared with whole and pureed canned pulses, which were all significantly lower than that for the whole wheat flour control. Likewise, Tovar and colleagues (91) observed that red kidney bean flour made in a laboratory setting by boiling, freeze-drying and milling whole red kidney beans led to comparable glycaemic responses as the whole autoclaved red kidney bean treatment, similar to canned beans in the present study. The results also show that micronization, as used in producing lentil and chickpea powder, is an ideal method for pulse powder production. Micronization does not diminish the positive effect of pulses on blood glucose and appetite control while maintaining their macronutrient profile. Compared to boiling, microwave cooking and pressure cooking, micronized pulses have a greater amount of tannins, phytic acid and oligosaccharides (118). These factors contribute to decreased carbohydrate digestibility in micronized pulses, which may also contribute to the observed low glycaemic responses after lentil powder and chickpea powder in present study (118).

However, Jenkins and colleagues (102) observed that lentil powder prepared in a laboratory setting by boiling lentils for 20 min, drying for 12 hours at 250°F and then grinding into a powder, produced a significantly greater glycaemic response compared to boiled whole lentils. The discrepancy with the current study could be due to their prolonged cooking time. Cooking time of pulses is positively correlated with starch hydrolysis rate (90). The 12 hours dry heat treatment of the lentil powder may have broken down some of the resistant starch and increased the degree of starch gelatinization, therefore making starch granules more readily available for

digestion. This prolonged heat may have also denatured all proteins in the lentil powder. Protein binds to starch and forms a complex which restricts the accessibility of enzymes to starch granules. Acid denaturation of protein increases the accessibility of α -amylase to the starch granules by breaking these bonds. Without protein blocking access to starch, α -amylase is able to attack starch granule and therefore increase starch digestibility. This prolonged heat may also have deactivated all α -amylase inhibitors found in lentils (102). In contrast, industrial processing methods are much faster as they are optimized to provide just enough time to soften the pulses and achieve the best textural characteristics. In the current study, the lentil and chickpea powders were produced via micronization, a process involving only 2-3 min cooking by ultraviolet light exposure (34, 94).

It is now clear that cooking first and then grinding, as used to produce pulse powders in the present study, does not change the low glycaemic property of the cooked pulses. As shown previously (102) and also in the present study, pureeing whole pulses does not increase their subsequent glycaemic response. As observed under microscopy, pureeing after cooking does increase cell wall destruction in the pulses, but a large number of cells remain intact (101). Even though the accessibility of digestive enzymes to starch granules increases slightly, it is unlikely the magnitude of this increase would trigger a significant increase of postprandial BG response (102, 103). However, pulse powders that are dry milled before cooking contain mostly free starches (91). Therefore, the enzyme accessibility increases significantly, leading to higher blood glucose responses compared to whole pulses or pulse powders that have been cooked prior to grinding.

Blood glucose suppression following the pizza meal was only observed after whole navy beans compared to whole wheat flour, but not after pureed navy beans and navy bean powder in experiment 1. This suggests that decreased particle size weakened navy bean's ability to suppress blood glucose response after a second meal. The seed tissue architecture plays a important role in maintaining prolonged glucose absorption of intact pulses (91). Post-meal blood glucose following lentil and chickpea treatments was marginally lower compared to whole wheat flour; however, these differences were not statistically significant with a sample size of twelve. A sample size of twelve is sufficient to detect a mean blood glucose difference of ± 0.5 mmol/L with a power of 0.8 and $\alpha = 0.05$. However, the difference in post-meal glycaemic responses

between the lentil and chickpea treatments and the whole wheat control ranged from 0.1-0.4 mmol/L. Thus, it is possible that these differences would reach significance with a higher subject number. Another note is that the amount of available carbohydrate in the treatments of the present study is small (37.8 g in total with 25 g from pulses or wheat) relative to studies that have detected second meal effects of pulses (102-112 g) (119). The duration between meals (2 hr) is also shorter compared to other studies investigated second meal effect (4-5 hr between breakfast and lunch or 10-12 hr between dinner and breakfast), making post-meal glycaemic effects more difficult to detect (120).

Pre-meal subjective appetite was suppressed by navy bean powder compared to whole wheat flour. It is likely the comparatively high amount of fiber and protein in the navy bean powder treatment suppressed appetite. The navy bean powder treatment used in the current study has 10.9 g more protein and 13.5 g more fiber than the whole wheat flour control. Studies have shown that satiety is positively correlated with the protein and fiber, and negatively correlated with the fat content of foods (30). Protein suppresses food intake (65) by triggering the satiety-inducing hormones insulin (66) and GLP-1 (67). Fiber regulates short-term satiety by adding bulk to the food thus increasing gastric distention (49), slowing gastric emptying (49), delaying macronutrient digestion in the duodenum by gelling (121) and triggering the release of the anorexic gut hormones GLP-1, CCK and PYY (51-54).

Another factor that may contribute to the suppression of appetite after navy bean powder is treatment palatability. In experiment 1, subjective appetite was positively correlated with treatment palatability, as well as its contributing factors: treatment pleasantness, taste and texture. Since foods with a lower palatability suppress subsequent hunger and desire to eat more than highly palatable foods (122), the low palatability of navy bean powder treatment compared to other treatments may have contributed to the suppressed appetite after its consumption. However, treatment palatability alone cannot explain the differences in subjective appetite. Whole wheat flour was rated the second lowest in treatment palatability, but had the least appetite suppression effect. The high fiber and high protein content in the navy bean powder treatment, as discussed earlier, is likely the biggest contributing factor. It is notable that the unpleasant granular texture of the powders did not affect the palatability of lentil and chickpea powder treatments. It is

encouraging to see that micronization, a low cost, fast and highly efficient technique, also preserved the taste of pulses.

Appetite scores after lentil powder and chickpea powder were the lowest within their respective experiments, but the differences were not statistically significant. The sample size of twelve was chosen to provide sufficient power to detect a glycaemic response (the primary dependent measure) effect size of 0.5 mmol/L. Subjective appetite scores are inherently more variable and thus the current experiments may not have sufficient power to detect a modest difference in subjective appetite ratings. It may also be because the pulse treatments were compared to a whole wheat flour treatment as a control, which may already be quite satiating. Indeed, all treatments suppressed subjective appetite for the two hours following consumption.

It is possible that the different energy content of the treatments may have confounded the subjective appetite results since the treatments were controlled for available carbohydrate but not energy content. However, Pearson's correlation analysis shows no correlation between the energy content of the treatments and mean subjective appetite before the pizza meal. Measurement of food intake at an *ad libitum* pizza meal after consumption of energy-matched pulse treatments is needed in order to isolate the effect of the pulse treatments on appetite control.

Thirst and water intake were measured since they are possible confounders in the measurement of subjective appetite. Feelings of thirst are often confused with hunger, leading to changes in reported appetite that cannot be isolated from those of thirst (123). Therefore, the sodium content of the treatments was matched to control in order to minimize differences in thirst between treatments. Pre-meal thirst was positively correlated with pre-meal App AUC and Δ App in experiment 1, negatively correlated with pre-meal App in experiment 2, and positively correlated with pre-meal App in experiment 3. However, neither thirst nor water intake were affected by any of the treatments and thus were unlikely to be confounding factors in the present study.

It is surprising that there were no differences between any of the treatments in terms of physical comfort, given that the high level of fiber and oligosaccharides in pulses may cause discomfort, as previously reported (124). It is suggested that if pulses are consumed in a small amount, as seen in present experiment (0.9-1 cup), the gas and bloating symptoms caused by the

fermentation of fiber and oligosaccharides in the large intestine is small (124). This is also likely due to the relatively short 200 min study period which may not be long enough for fermentation to take place (125).

Navy bean powder contains more protein and fiber compared to whole navy beans, whereas lentil powder and chickpea powder have comparable amount of protein and fiber to their respective whole pulses. Unfortunately, the specifics of the production procedure used in the creation of the navy bean powder are a trade secret and thus not available. The navy beans used to produce navy bean powder may be grown in different areas than the canned navy beans. This could also contribute to the protein and fiber differences since pulse composition is known to vary by cultivar (98, 118). On another hand, the similarity of macronutrient profile between whole lentils/chickpeas and lentil/chickpea powders indicates that industry powder processing can preserved most, if not all, of the nutrients in the pulses.

In conclusion, commercial processing of pulses to a powder form does not alter their beneficial effects on blood glucose and appetite regulation. Pulse powders, therefore, could be used as value-added ingredients in home cooking and functional foods that will improve glycaemic and appetite control. The development of such foods will also promote consumption of pulses in convenience foods among individuals who normally avoid them due to taste or perceived inconvenience. The use of pulse flours in this manner may also replace other, less nutritious ingredients such as wheat, corn and rice flours.

CHAPTER 7

FUTURE DIRECTIONS

7 FUTURE DIRECTIONS

The results from this study shows that commercial processing of pulses to a powder form does not negatively impact their effects on glycaemic response and appetite control in healthy young men. The following section discusses some of the limitations of the study and how they may be addressed in the future.

Due to the strict inclusion criteria in the present study aimed to maximize participant homogeneity, the study population did not represent the whole population and thus results may not be applied to the population as whole. Age, BMI, sex and metabolic disorders can affect physiological responses to dietary components (126). The blood glucose and appetite suppression effects of powdered navy beans, lentils and chickpeas observed in the present study are particularly encouraging for individuals with diabetes mellitus. Type-2 diabetes risk is strongly associated with obesity (127) and therefore the combined effects of suppressed glycaemia and appetite of pulse powders would be especially beneficial. Further studies are needed to determine if the benefits of pulse powders are applicable to those with type-2 diabetes.

Because the current study is focused on glycaemic response before and after a fixed meal, food intake was not measured as this would confound post-meal glycaemia. To determine if the pulse powders lead to suppression of food intake, a similar study utilizing an *ad libitum* amount of pulse powders as well as an ad libitum pizza meal could be conducted. This would accurately represent a real-world scenario where food is rarely artificially limited. Measurement of gut hormones such as insulin, GLP-1, CCK, PYY and ghrelin could be measured to investigate whether processing pulses to a powder eliminate their positive effects on food intake regulation and help elucidate their mechanism of action.

The treatments in present study were prepared by cooking pulse powders with minimal added ingredients to reduce confounding factors. Future studies to investigate the effects of pulse powders as part of a food matrix on glycaemic response and appetite control is needed. Whether their health benefits after being incorporated into foods are maintained, and whether these effects are dose depend are important for the development of functional foods containing pulse powders aimed at reducing blood glucoses and suppressing food intake.

CHAPTER 8

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CHAPTER 9

APPENDICES

APPENDIX I: Session Forms

Motivation to eat and Thirst

Palatability

Physical Comfort

Energy and Fatigue

Sleep Habits and Stress Factors Questionnaire

Recent Food Intake and Activity Questionnaire

| ID: _ | | | |
|-------|---|-----------------------|------------|
| DAT | TE: | | |
| SES | SION: | | |
| | *** | | |
| | | d Analogue Scales | |
| | Motivati | ion to Eat and Thirst | |
| | | | |
| | se questions relate to your "motivation Il "x" across the horizontal line at the | | |
| 1. | How strong is your desire to eat? | • | - |
| | VERY | | VERY |
| | weak | | strong |
| | | | C |
| 2. | How hungry do you feel? | | |
| | NOT | | As hungry |
| | hungry | as I have | |
| | at all | | ever felt |
| 3. | How full do you feel? | | |
| | NOT | | |
| | full | | VERY |
| | at all | | full |
| 4. | How much food do you think you | could eat? | |
| | NOTHING | | A LARGE |
| | | | amount |
| | | | |
| 5. | How thirsty do you feel? | | |
| | NOT | | As thirsty |
| | thirsty — | | as I have |
| | at all | | ever felt |

| ID: | |
|------------------------------------|--|
| DATE: | |
| SESSION: | |
| | al Analogue Scales ability: Treatment |
| | of the beverage/food you just consumed. Please rate ne horizontal line at the point which best reflects your |
| 1. How pleasant have you found t | the beverage/food? |
| NOT | VERY |
| at all pleasant | pleasant |
| 2. How tasty have you found the | treatment? |
| NOT | VERY |
| at all | tasty |
| tasty | |
| 3. How did you like the texture of | f the treatment? |
| NOT | VERY |
| at all | much |

| ID: | |
|---|---|
| DATE: | |
| SESSION: | |
| | nalogue Scales bility: Pizza |
| · · · · · · · · · · · · · · · · · · · | he beverage/food you just consumed. Please rate orizontal line at the point which best reflects you |
| How pleasant have you found the beverage/ | food? |
| NOT | VERY |
| at all | pleasant |
| pleasant | 1 |
| | |

| ID: | | |
|-------|--|-----------|
| DATE | E: | |
| SESSI | ION: | <u>es</u> |
| | questions relate to your "motivation to eat" at this tin "x" across the horizontal line at the point which best | , , , |
| 1. | Do you feel nauseous? | |
| | NOT at all | VERY much |
| 2. | Does your stomach hurt? | |
| | NOT at all | VERY much |
| 3. | How well do you feel? | |
| | NOT well at all | VERY well |
| 4. | Do you feel like you have gas? | |
| | NOT at all | VERY much |
| 5. | Do you feel like you have diarrhea? | |
| | NOT at all | VERY much |

| ID: | | |
|--------|---|---|
| DATE: | | |
| SESSIO | N: | |
| | Visual Anal Energy an | |
| | estions relate to your energy level and fatigue at the horizontal line at the point which best reflects you | his time. Please rate yourself by placing a small "x" pur present feelings. |
| 1. | How energetic do you feel right now? | |
| No | OT | VERY |
| at | all | energetic |
| 2. | How tired do you feel right now? | |
| No | OT | VERY |
| at | all | tired |

| | : ATE: ESSION: | | | | |
|--|--------------------------------------|---|--|--|------|
| | Sleep Ha | abits and Stres | s Factors Questionn | aire | |
| 1. | Did you have a normal night' | s sleep last | Past 24 hrs: | □ YES | □NO |
| | mgnt: | | If yes, please des | scribe briefly: | |
| | □ YES | □ NO | | | |
| 2. | How many hours of sleep did | I you have? | 7. Are you under any unusual stress? (i.e. exams, reports, work deadlines, personal) | | |
| 3. | At what time did you go to be night? | ed last | Today: Past 24 hrs: | □ YES □ YES | □ NO |
| | mg.w. | | If yes, please describe briefly: | | |
| 4. At what time did you wake up this morning? | | 8. Have you becactivity, unus within the pa | sual to your norm | | |
| 5. | Recount your activity since w | vaking up: | | □ YES | □NO |
| TI | ME ACTIVITY | | If yes, please des | scribe briefly: | |
| | | | 9. Have you had other than wa | d anything to eat ater for the past 1 | |
| 6. Are you experiencing any feelings of illness or discomfort other than those | | | □ YES | □ NO | |
| | from hunger? | | If yes, please des | scribe briefly: | |
| То | oday: | □NO | | | |

| ID: | |
|--|-------------------------|
| DATE: | |
| SESSION: | |
| Recent Food Intake and Activity Questionnair | <u>e</u> |
| At what time did you have dinner? | |
| Please describe your dinner last night (list all food and drink and give as size): | - |
| The following three questions relate to your food intake, activity and hours. Please rate yourself by placing a small "x" across the horizontal best reflects your present feelings. | stress over the last 24 |
| How would you describe your food intake over the past 24 hours? | |
| Much LESS than usual | Much MORE than usual |
| How would you describe your level of activity over the last 24 hours? Much LESS | Much MORE than usual |
| How would you describe your level of stress over the last 24 hours? | |
| Much LESS | Much MORE |
| than usual | than usual |

APPENDIX II: Advertisement



University of Toronto Department of Nutritional Sciences

Male Participants Needed For Nutritional Study!

Requirements: age 18-30 years, non-smoking *Involves:* Four sessions over 4 weeks

\$ Compensation and Food are provided **\$**

Please contact us at: appetite.studies@gmail.com

APPENDIX III: Screening forms

Baseline Information Questionnaire

Eating Habits Questionnaire

Food Acceptability Questionnaire

Information Sheet and Consent Form

Baseline Information Questionnaire

(NOTE: After you are recruited for the study, you will be assigned an ID# which will be used on your forms and data throughtout the study.)

| NAME: | | AGE: | |
|--|-----------------|---------------------------------------|---------------------------------------|
| ADDRESS: | | | |
| PHONE #: () | E-MAIL: | | · · · · · · · · · · · · · · · · · · · |
| HEIGHT: | WEIGHT: _ | · · · · · · · · · · · · · · · · · · · | BMI: |
| Participation in Athletics/Exercise: | | | |
| ACTIVITY HO | OW OFTEN? | НО | W LONG? (HOURS) |
| | | | |
| Do you usually eat breakfast? | | □ YES | □ NO |
| If YES, what do you usually eat? _ | | | |
| <u>Health Status:</u> | | | |
| Do you have diabetes? | | □ YES | □NO |
| Do you have any other major disease or | condition? | □ YES | □NO |
| If YES, please specify: | | | |
| Are you taking any medications? | | □ YES | □NO |
| If YES, please specify: | | | |
| Do you have reactions to any foods? | | □ YES | □NO |
| If YES, please specify: | | | |
| Are you on a special diet? | | □ YES | □NO |
| If YES, please specify: | | | |
| Have you recently lost or gained weight? | | □ YES | □NO |
| If YES, please specify: | | | |
| Do you smoke? | | □ YES | □NO |
| How many alcoholic beverages do you co | onsume per day? | | Per week? |

Eating Habits Questionnaire

Choose the appropriate answer to best describe your personal situation.

| 1. | How often are you dieting? | | |
|-----|--|--|--|
| | Never rarely sometimes often always | | |
| 2. | What is the maximum amount of weight (in pounds) that you have ever lost within one month? | | |
| | 1 - 4 5 - 9 10 - 14 15 - 19 20+ | | |
| 3. | What is your maximum weight gain within one week? | | |
| | 0-11.1-22.1-33.1-55.1+ | | |
| 4. | In a typical week, how much does your weight fluctuate? | | |
| | 0-1 1.1-2 2.1-3 3.1-5 5.1+ | | |
| 5. | Would a weight fluctuation of 5lbs affect the way you live your life? | | |
| | Not at all slightly moderately very much | | |
| 6. | Do you eat sensibly in front of others and splurge alone? | | |
| | Never rarely often always | | |
| 7. | Do you give too much time and thought to food? | | |
| | Never rarely often always | | |
| 8. | Do you have feelings of guilt after overeating? | | |
| | Never rarely often always | | |
| 9. | How conscious are you of what you are eating? | | |
| | Not at all slightly moderately extremely | | |
| 10. | How many pounds over your desired weight were you at your maximum weight? | | |
| | 0-1 2 - 5 6 - 10 11 - 20 21+ | | |

Food Acceptability Questionnaire

Please indicate with a rating between 1 and 10 how much you enjoy the following foods (1 = not at all, 10 = very much) and how often you eat them (never, daily, weekly, monthly).

| | Enjoyment? | How often? |
|--|--|--|
| 1. Pasta | | |
| 2. Rice | | |
| 3. Potatoes (mashed, roasted) | | · |
| 4. French fries | | · |
| 5. Pizza | | |
| 6. Bread, bagels, dinner rolls | | |
| 7. Sandwiches, subs | | |
| 8. Cereal | | |
| 9. Cake, donuts, cookies | | |
| 10. Protein/breakfast shakes | | |
| At the end of each session, you will be provided that you will enjoy, we ask that you rank the preferences (i.e. 1st, 2nd, 3rd choice) in the space of pizza, then do not rank it but instead place an " | following pizzas accorprovided. If you do N | ording to your <u>persona</u> OT like a particular typ |
| Pepperoni (cheese, pepperoni) | | |
| Deluxe (cheese, pepperoni, peppers, mushrooms) | | |
| Three-cheese (mozzarella, cheddar, parmesan) | | |

Information Sheet and Consent Form



Department of Nutritional Sciences FitzGerald Building, 150 College Street, 3rd Floor Toronto, ON M5S 3E2 CANADA

The Effects of Whole and Powdered Lentils on Glycaemic Response and Satiety in Young Men

Information Sheet and Consent Form

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Funding Source:

Funding for this project is provided by Agriculture and Agri-Food Canada and Saskatchewan Pulse Growers.

Background and Purpose of Research:

Lentils have been shown to have a beneficial effect on blood sugar and appetite (less hunger) control compared to other foods. However, whether these health benefits remain when lentils are made into powder form for use in other food products is currently unclear.

The purpose of this study is to determine the effect of lentils flour on appetite and blood sugar responses.

This study will have 20 participants.

Invitation to Participate:

You are being invited to take part in this study. If you chose to take part, you will be asked to eat a treatment of lentils or wheat flour four times (four sessions). At each session, your appetite and blood sugar will be measured after eating the treatment. Each session will take up to 3.5 hours of your time.

Eligibility:

To participate in this study you must be a healthy male and between the ages of 18 and 30. You must be a nonsmoker and you cannot be taking any medications. Please indicate if you are taking any supplements. You will not be able to participate if you have allergies to beans, wheat products or tomatoes. The study will take place in the Department of Nutritional Sciences, Room 334, 331 and 331A, FitzGerald Building, 150 College Street, Toronto, ON.

Procedure:

To find out if you can take part in this study, you will be asked to fill out questionnaires, which ask questions about your age, if you smoke, exercise, your health, if you are on any medications and your eating habits. Your height and weight will be measured.

If you can take part, you will be asked to fill out questionnaires about the foods you like. You will be scheduled to meet with us for four sessions.

You will be asked to eat a standard breakfast on the day of the session following a 11 hour fast (no eating for 11 hours before eating breakfast). We will give you the standard breakfast (cereal, milk and orange juice) the day before the session.

You will be asked to arrive at the FitzGerald Building between 9:45 a.m. and 12:45 p.m., 3 ³/₄ hours after eating breakfast. Please do not eat between breakfast and meeting with us. You will be asked to stick to your normal routine, including exercise and to eat a similar meal the night before each session. You can drink water up to one hour before meeting with us.

At each session you will be asked to eat a treatment, give blood samples and to complete questionnaires at the times outlined in the table below. The treatment will consist of commercially whole lentils, pureed lentils, lentils flour or wheat flour. 12 times during each session, for a total of 48 times over the whole study, you will be asked to provide a small drop of blood by finger prick. Blood will be sampled before eating the treatment (0 min) and then at 15, 30, 45, 60, 90,120,140.155,170,185,200 min after eating the treatment. You will be asked to fill out visual analog scale (VAS) questionnaires measuring your appetite and physical comfort as well as the palatability (pleasantness) of the treatment. Each session will last up to 3.5 hours.

Time and Activity Schedule for Each Session Example

| Time | Activity | |
|---------------|---|--|
| 7:00 | Consumption of breakfast | |
| 10:45 | Arrive at the laboratory | |
| 10:50 | Fill in Sleep, Stress, and VAS questionnaires and take first blood | |
| | sample | |
| 11:00 - 11:15 | Eat the treatment (0 min). | |
| 11:15 – 1:00 | Blood sampling and VAS questionnaires at 15, 30, 45, 60, 90 and 120 | |
| | min | |
| 1:00-1:20 | Pizza served and eaten at 120 minutes | |
| 1:20-2:20 | Blood sampling and VAS questionnaires at 140, 155, 170, 185 and | |
| | 200 minutes | |

VAS: Visual analogue scale

Notice: This is just an example. You can choose the staring time between 9:45 a.m. and 12:45 p.m

Voluntary Participation and Early Withdrawal:

It is hoped that you will finish all four sessions. However, you may choose to stop being in the study at anytime without any problems.

Early Termination:

Not applicable.

Risks:

All of the foods and beverages (water) that you will be asked to consume are prepared hygienically in the kitchen and present minimal risk.

After the overnight fast you may feel faint or dizzy, however the risk of this is minimal.

The risks and discomfort will come from the blood sampling procedure. A total of 12 finger pricks will be conducted per session. Great care will be taken when taking your finger prick blood samples. The investigator will help you. To make sure that you are not exposed to another person's needle, we will ask you to sit away from other study participants. You will put a needle into the finger prick gun before taking each blood sample and then put it into the safety container. You will swab your finger with alcohol before and after each finger prick and will use a new sterile needle each time.

Some discomfort will be felt as a result of a sharp momentary pain caused as the needle enters the skin. However, because the lancet needle is very small the pain felt is usually less than you might feel from skin puncture during vaccination or if a blood sample is taken by a needle inserted in a vein. You will be provided with your own finger prick gun for the entire study.

There is very little risk of infection. Before the finger is pricked the area is cleaned with an alcohol swab. There might be slight bruising under the skin, but this will be minimized by applying pressure after the finger is pricked and blood sugar is measured.

There is always a possibility that you may become ill following consumption of food, but this is very unlikely. All treatments are freshly prepared at the time of your session.

You may experience flatulence (passing gas) and feelings of gastrointestinal discomfort (bloating) from the treatments if they are high in fiber. This hardly ever happens and there is no health risk linked with these effects.

Benefits:

You will not benefit directly from taking part in this study. You will be shown your blood sugar results and if they are not normal you will be told and advised to talk to your doctor. The foods and drinks (water) will be provided for free.

Confidentiality and Privacy:

Confidentiality will be respected and no information that shows your identity will be released or published without your permission unless required by law. Your name, personal information and signed consent form will be kept in a locked filing cabinet in the investigator's office. Your results will not be kept in the same place as your name. Your results will be recorded on data sheets and in computer records that have an ID number for identification, but will not include your name. Your results, identified only by an ID number, will be made available to the study sponsor if requested. Only study investigators will have access to your individual results.

Publication of Results:

The results of the study may be presented at scientific meetings and published in a scientific journal. If the results are published, only average and not individual values will be reported.

Possible Commercialization of Findings:

This study is preliminary. Once these products are tested more widely in future studies, results may lead to commercialization of a product, new product formulation, changes in the labeling of a product and/or changes in the marketing of a product; you will not share in any way from the possible gains or money made by commercial application of findings.

Alternative Treatment/ Therapy:

Not applicable.

New Findings:

If anything is found during the course of this research which may change your decision to continue, you will be told about it.

Compensation:

You will be paid \$40 per session. You will also be given \$6 per session for travel (bus, subway). If you withdraw from the study before finishing or asked to withdraw, you will be paid for the sessions you have already finished.

Injury Statement:

If you begin to feel sick following participation in the study, please seek medical advice as soon as possible. We will provide your medical specialist with information about the food you have consumed during the session, so take our phone number with you.

Rights of Subjects:

Before agreeing to take part in this research study, it is important that you read and understand your role as described here in this study information sheet and consent form. You waive no legal rights by taking part in this study. If you have any questions or concerns about your rights as a participant you can contact the Ethics Review Office at ethics.review@utoronto.ca or call 416-946-3273.

If you have any questions after you read through this information please do not hesitate to ask the investigators for further clarification.

Dissemination of findings:

A summary of results will be made available for you to pick up after the study is done.

Copy of informed consent for participant:

You are given a copy of this informed consent to keep for your own records.

Consent:

I acknowledge that the research study described above has been explained to me and that any questions that I have asked have been answered to my satisfaction. I have been informed of the alternatives to participation in this study, including the right not to participate and the right to withdraw. As well, the potential risks, harms and discomforts have been explained to me. I understand that I will receive compensation for my time spent participating in the study.

As part of my participation in this study, I understand that I may come in contact with other study participants because our session times overlap. I agree to keep anything I learn about other participants confidential and know that other participants have agreed to do the same for me.

I hereby agree and give my authorized consent to participate in the study and to treat confidential information in a restrictive manner as described above. I have been given a copy of the consent form to keep for my own records.

| Participant Name | Signature | Date |
|-------------------|-----------|------|
| Witness Name | Signature | Date |
| Investigator Name | Signature | Date |

APPENDIX IV

List of Ingredients

Treatment Preparation Method

List of Ingredients

Robin Hood® All Purpose Whole Wheat Flour (2Kg)

- Ingredients: Whole Wheat Flour, Amylase, Ascorbic Acid, Azodicarbonamide
- ©/TMMC/®/MD Smucker Foods of Canada Co. Markham, ON L3R OP3

| Amount/Serving | |
|---------------------------|-----|
| Per ½ cup. (30g) | |
| Calories (kcal) | 110 |
| Fat (g) | 0.5 |
| - Saturated Fat (g) | 0.2 |
| - Trans Fat (g) | 0 |
| Cholesterol (mg) | 0 |
| Sodium (mg) | 5 |
| Carbohydrates (g) | 22 |
| - Total Dietary Fiber (g) | 3 |
| - Sugars (g) | 0 |
| Protein (g) | 4 |

Hunt's ® Tomato Paste – Original (156 ml)

- Ingredients: tomatoes
- Prepared under license for ConAgra Foods Canada Inc., ConAgra Canada Inc., Mississauga, Ontario, Canada L4V 1W5

| Amount/Serving | |
|---------------------------|----|
| Per 2 tbsp. (30 ml) | |
| Calories (kcal) | 20 |
| Fat (g) | 0 |
| - Saturated Fat (g) | 0 |
| - Trans Fat (g) | 0 |
| Cholesterol (mg) | 0 |
| Sodium (mg) | 20 |
| Carbohydrates (g) | 4 |
| - Total Dietary Fiber (g) | 1 |
| - Sugars (g) | 3 |
| Protein (g) | 1 |

ReaLemon® Lemon Juice from Concentrate (125.0 ml)

- Ingredients: water, concentrated lemon juice, sulphites, lemon oil
- Trademark of: Cadbury Beverages Delaware Inc., product of USA
- Authorised User Cadbury Beverages Canada Inc. Cadbury Canada Inc., Mississauga, Ontario, L5R 3L7

| Amount/Serving | |
|---------------------------|---|
| Per 1 tsp. (5 ml) | |
| Calories (kcal) | 0 |
| Fat (g) | 0 |
| - Saturated Fat (g) | 0 |
| - Trans Fat (g) | 0 |
| Cholesterol (mg) | 0 |
| Sodium (mg) | 0 |
| Carbohydrates (g) | 0 |
| - Total Dietary Fiber (g) | 0 |
| - Sugars (g) | 0 |
| Protein (g) | 0 |

McCormick® No Salt Added Garlic & Herb Seasoning (77 g)

- Ingredients: dehydrated vegetables (garlic, onion, parsley, celery), spice (including red pepper), dried orange peel
- Imported by: McCormick Canada London, Canada N6A 4Z2

| Amount/Serving | |
|---------------------------|-----|
| Per 2 ml (1 g) | |
| Calories (kcal) | 0 |
| Fat (g) | 0 |
| - Saturated Fat (g) | 0 |
| - Trans Fat (g) | 0 |
| Cholesterol (mg) | 0 |
| Sodium (mg) | 0 |
| Carbohydrates (g) | 1 |
| - Total Dietary Fiber (g) | 0 |
| - Sugars (g) | 0 |
| Protein (g) | 0.1 |

Equality® Basil Leaves (42g)

- Ingredients: Basil Leaves
- Imported for The Great Atlantic & Pacific Company of Canada LTD., Toronto, Canada, M5W 1A6

| Amount/Serving | |
|-------------------------------|-----|
| Per $\frac{1}{2}$ tsp. (0.5g) | |
| Calories (kcal) | 0 |
| Fat (g) | 0 |
| - Saturated Fat (g) | 0 |
| - Trans Fat (g) | 0 |
| Cholesterol (mg) | 0 |
| Sodium (mg) | 0 |
| Carbohydrates (g) | 0 |
| - Total Dietary Fiber (g) | 0 |
| - Sugars (g) | 0 |
| Protein (g) | 0.1 |

Equality® Ground Black Pepper (145g)

- Ingredients: Ground Black Pepper
- Imported for The Great Atlantic & Pacific Company of Canada LTD., Toronto, Canada, M5W 1A6

Equality® Parsley Flakes (14g)

- Ingredients: Dehydrated Parsley
- Imported for The Great Atlantic & Pacific Company of Canada LTD., Toronto, Canada, M5W 1A6

| Amount/Serving | |
|---------------------------|-----|
| Per ½ tsp. (0.5g) | |
| Calories (kcal) | 0 |
| Fat (g) | 0 |
| - Saturated Fat (g) | 0 |
| - Trans Fat (g) | 0 |
| Cholesterol (mg) | 0 |
| Sodium (mg) | 0 |
| Carbohydrates (g) | 0 |
| - Total Dietary Fiber (g) | 0 |
| - Sugars (g) | 0 |
| Protein (g) | 0.1 |

Equality® Garlic Powder (155g)

- Ingredients: Dehydrated Garlic, Canola Oil
- Imported for The Great Atlantic & Pacific Company of Canada LTD., Toronto, Canada, M5W 1A6

| Amount/Serving | |
|-------------------------------|-----|
| Per $\frac{1}{4}$ tsp. (0.5g) | |
| Calories (kcal) | 0 |
| Fat (g) | 0 |
| - Saturated Fat (g) | 0 |
| - Trans Fat (g) | 0 |
| Cholesterol (mg) | 0 |
| Sodium (mg) | 0 |
| Carbohydrates (g) | 0 |
| - Total Dietary Fiber (g) | 0 |
| - Sugars (g) | 0 |
| Protein (g) | 0.1 |

Selection® Chilli Powder (100g)

- Ingredients: Chilli Pepper, Paprika, Cumin, Salt, Oregano, Dehydrated garlic, Coriander, Silicon Dioxide, Ethoxyquin (antioxidant)
- Packed for Metro Brands, Montreal(Quebec),H1C 1V6,Toronto(Ontario) M9B 1B9

| Amount/Serving | |
|-------------------------------|-----|
| Per $\frac{1}{4}$ tsp. (0.5g) | |
| Calories (kcal) | 0 |
| Fat (g) | 0 |
| - Saturated Fat (g) | 0 |
| - Trans Fat (g) | 0 |
| Cholesterol (mg) | 0 |
| Sodium (mg) | 10 |
| Carbohydrates (g) | 0 |
| - Total Dietary Fiber (g) | 0 |
| - Sugars (g) | 0 |
| Protein (g) | 0.1 |

Redpath® Special fine Granulated Sugar (2kg)

- Ingredients: Pure Cane Sugar
- Prepared by Redpath® Sugar LTD. Toronto, Ontario, M5E 1A3

| Amount/Serving | |
|---------------------------|----|
| Per 1 tsp. (4g) | |
| Calories (kcal) | 15 |
| Fat (g) | 0 |
| - Saturated Fat (g) | 0 |
| - Trans Fat (g) | 0 |
| Cholesterol (mg) | 0 |
| Sodium (mg) | 0 |
| Carbohydrates (g) | 0 |
| - Total Dietary Fiber (g) | 0 |
| - Sugars (g) | 4 |
| Protein (g) | 0 |

Experiment 1 Navy Bean

FERMA® White Pea Beans (540ml)

- Ingredients: White Pea Beans, Water, Salt, Calcium Chloride
- Packed for Ferma® Import &Export LTD. Toronto, M6H 4C7, Montreal, Canada

| Amount/Serving | _ |
|---------------------------|-----|
| Per 125.0ml (100g) | |
| Calories (kcal) | 120 |
| Fat (g) | 0.4 |
| - Saturated Fat (g) | 0.1 |
| - Trans Fat (g) | 0 |
| Cholesterol (mg) | 0 |
| Sodium (mg) | 280 |
| Carbohydrates (g) | 21 |
| - Total Dietary Fiber (g) | 8 |
| - Sugars (g) | 0 |
| Protein (g) | 8 |

VegaFull ® Instant Dehydrated Whole Navy Bean Powder (10Kg)

- Ingredients: Cooked Navy Beans, Sugar
- Manufactured by ADM® Edible Bean Specialties, Inc.

| Amount/Serving | |
|---------------------------|-----|
| Per ½ cup (91g) | |
| Calories (kcal) | 110 |
| Fat (g) | 1 |
| - Saturated Fat (g) | 0 |
| - Trans Fat (g) | 0 |
| Cholesterol (mg) | 0 |
| Sodium (mg) | 10 |
| Carbohydrates (g) | 23 |
| - Total Dietary Fiber (g) | 9 |
| - Sugars (g) | 3 |
| Protein (g) | 09 |

Experiment 2 Lentil

Precooked Eston Lentil Powder (10kg)

- Ingredients: Eston Lentils
- Prepared by InfraReady® Products Ltd. Saskatoon, SK Canada

| Amount/Serving | |
|---------------------------|-------|
| Per 100g | |
| Calories (kcal) | 353 |
| Fat (g) | 1.06 |
| - Saturated Fat (g) | 0.156 |
| - Trans Fat (g) | 0 |
| Cholesterol (mg) | 0 |
| Sodium (mg) | 6 |
| Carbohydrates (g) | 60.08 |
| - Total Dietary Fiber (g) | 30.5 |
| - Sugars (g) | 2.03 |
| Protein (g) | 25.80 |

NuPak® Green Lentils (540mL)

- Ingredients: Green lentils, water, salt, calcium chloride, citric acid, disodium EDTA
- Packed under licence for Shah Trading Company Ltd. Scarborough, ON

| Amount/Serving | |
|---------------------------|-----|
| Per 250mL (190g) | |
| Calories (kcal) | 150 |
| Fat (g) | 0.4 |
| - Saturated Fat (g) | 0 |
| - Trans Fat (g) | 0 |
| Cholesterol (mg) | 0 |
| Sodium (mg) | 550 |
| Carbohydrates (g) | 26 |
| - Total Dietary Fiber (g) | 14 |
| - Sugars (g) | 0 |
| Protein (g) | 13 |

Experiment 3 Chickpea

Precooked Garbanzo Bean Powder (10kg)

- Ingredients: Garbanzo Beans (Chick Peas)
- Prepared by InfraReady® Products Ltd. Saskatoon, SK Canada

| Amount/Serving | |
|---------------------------|-------|
| Per 100g | |
| Calories (kcal) | 364 |
| Fat (g) | 6.04 |
| - Saturated Fat (g) | 0.626 |
| - Trans Fat (g) | 0 |
| Cholesterol (mg) | 0 |
| Sodium (mg) | 24 |
| Carbohydrates (g) | 60.65 |
| - Total Dietary Fiber (g) | 17.4 |
| - Sugars (g) | 10.70 |
| Protein (g) | 19.30 |

NuPak® Chick Peas (540mL)

- Ingredients: Chick peas, water, salt, disodium EDTA
- Packed under licence for Shah Trading Company Ltd. Scarborough, ON

| Amount/Serving | |
|---------------------------|-----|
| Per 250mL (190g) | |
| Calories (kcal) | 200 |
| Fat (g) | 3.5 |
| - Saturated Fat (g) | 0.4 |
| - Trans Fat (g) | 0 |
| Cholesterol (mg) | 0 |
| Sodium (mg) | 560 |
| Carbohydrates (g) | 34 |
| - Total Dietary Fiber (g) | 10 |
| - Sugars (g) | 0 |
| Protein (g) | 11 |

Treatment Preparation Method

Tomato Sauce:

Mix 125.0g no salt added tomato paste with ½ tsp. lemon juice, ½ tsp. garlic and herb seasoning, ½ tsp. basil, ½ tsp. black pepper, ¼ tsp. garlic powder, ½ tsp. parsley, ¼ tsp. chili pepper and ¼ tsp. sugar.

Experiment 1 Navy Beans

Treatment 1 Canned Whole Navy Beans:

Pour whole canned navy beans into a strainer, wash them in running water for 30 seconds and drain well.

Mix 188.0g of navy beans with 70.0 ml of filtered water.

Make the tomato sauce by combining 125.0g of tomato paste, spices, sugar, lemon juice and 110ml of water.

Cook the tomato sauce in an non-sticky cooking pan on medium heat until the liquid is boiled, about 3 min.

Add the whole navy beans to the pot, mix well, cover and cook until it is boiled again, about 2 min, turn to "2" and cook for 10 min, stir every 2 min.

After 10 min, transfer the treatment into an airtight container, measure the weight and add water to 405.0g. Cover, label and keep it in the fridge.

Treatment 2 Pureed Canned Navy Beans:

Pour whole canned navy beans into a strainer, wash them in running water for 30 seconds and drain well.

Mix 188.0g of navy beans with 70.0 ml of filtered water.

Blend the navy bean mixture in a food processor (KitchenAid Mod: KFP720OB2) for 60 seconds.

Make the tomato sauce by combining 125.0g of tomato paste, spices, sugar, lemon juice and 110ml of water.

Cook the tomato sauce in an un-sticky cooking pan on medium heat until the liquid is boiled, about 3 min.

Add the pureed navy bean mixture to the pot, mix well, cover and cook until it is boiled again, about 2 min, turn to low heat and cook for 10 min, stir every 2 min.

After 10 min, transfer the treatment into an airtight container, measure the weight and add water to 450.0g. Cover, label and keep it in the fridge.

Treatment 3 Navy Bean Powder:

Mix 62.5g of navy bean powder well with 195.5ml of water and 0.90g of Salt.

Make the tomato sauce by combining 125.0g of tomato paste, spices, sugar, lemon juice and 110ml of water.

Cook the tomato sauce in an un-sticky cooking pan on medium heat until the liquid is boiled, about 3 min.

Add the navy bean powder mixture to the pot, mix well, cover and cook until it is boiled again, about 2 min, turn to low heat and cook for 10 min, stir every 2 min.

After 10 min, transfer the treatment into an airtight container, measure the weight and add water to 450.0g. Cover, label and keep it in the fridge.

Whole Wheat Flour:

Mix 39.5g of whole wheat flour with 208.0ml of water and 0.94g of Salt.

Make the tomato sauce by combining 125.0g of tomato paste, spices, sugar, lemon juice and 110ml of water.

Cook the tomato sauce in an un-sticky cooking pan on medium heat until the liquid is boiled, about 3 min.

Add the whole wheat mixture to the pot, mix well, cover and cook until it is boiled again, about 2 min, turn to low heat and cook for 10 min, stir every 2 min.

After 10 min, transfer the treatment into an airtight container, measure the weight and add water to 450.0g. Cover, label and keep it in the fridge.

Experiment 2 Lentils

Treatment 1 Whole Canned Lentils:

Pour whole canned lentils into a strainer, wash them in running water for 30 seconds and drain well.

Mix 183.8g of lentils with 70.0 ml of filtered water.

Blend the chickpea mixture in a food processor (KitchenAid Mod: KFP720OB2) for 60 seconds. Make the tomato sauce by combining 125.0g of tomato paste, spices, sugar, lemon juice and 110ml of water.

Cook the tomato sauce in an un-sticky cooking pan on medium heat until the liquid is boiled, about 3 min.

Add the whole lentil mixture to the pot, mix well, cover and cook until it is boiled again, about 2 min, turn to low heat and cook for 10 min, stir every 2 min.

After 10 min, transfer the treatment into an airtight container, measure the weight and add water to 450.0g. Cover, label and keep it in the fridge.

Treatment 2 Pureed Canned Lentils:

Pour whole canned lentils into a strainer, wash them in running water for 30 seconds and drain well.

Mix 183.8g of lentils with 70.0 ml of filtered water.

Blend the chickpea mixture in a food processor (KitchenAid Mod: KFP720OB2) for 60 seconds. Make the tomato sauce by combining 125.0g of tomato paste, spices, sugar, lemon juice and 110.0 ml of water.

Cook the tomato sauce in an un-sticky cooking pan on medium heat until the liquid is boiled, about 3 min.

Add the pureed lentil mixture to the pot, mix well, cover and cook until it is boiled again, about 2 min, turn to low heat and cook for 10 min, stir every 2 min.

After 10 min, transfer the treatment into an airtight container, measure the weight and add water to 450.0g. Cover, label and keep it in the fridge.

Treatment 3 Lentil Powder:

Mix 47.7g lentil powder well with 204.9ml of water and 1.21 Salt.

Make the tomato sauce by combining 125.0g of tomato paste, spices, sugar, lemon juice and 110ml of water.

Cook the tomato sauce in an un-sticky cooking pan on medium heat until the liquid is boiled, about 3 min.

Add the lentil powder mixture to the pot, mix well, cover and cook until it is boiled again, about 2 min, turn to low heat and cook for 10 min, stir every 2 min.

After 10 min, transfer the treatment into an airtight container, measure the weight and add water to 450.0g. Cover, label and keep it in the fridge.

Treatment 4 Whole Wheat Flour:

Mix 39.4g of whole wheat flour with 213.2ml of water and 1.21g of Salt.

Make the tomato sauce by combining 125.0g of tomato paste, spices, sugar, lemon juice and 110ml of water.

Cook the tomato sauce in an un-sticky cooking pan on medium heat until the liquid is boiled, about 3 min.

Add the whole wheat mixture to the pot, mix well, cover and cook until it is boiled again, about 2 min, turn to low heat and cook for 10 min, stir every 2 min.

After 10 min, transfer the treatment into an airtight container, measure the weight and add water to 450.0g. Cover, label and keep it in the fridge.

Study 3 Chickpeas

Treatment 1 Whole Canned Chickpeas:

Pour whole canned chickpeas into a strainer, wash them in running water for 30 seconds and drain well.

Mix 148.8g of chickpeas with 104.7ml of filtered water and 0.31g of salt.

Make the tomato sauce by combining 125.0g of tomato paste, spices, sugar, lemon juice and 110.0 ml of water.

Cook the tomato sauce in an un-sticky cooking pan on medium heat until the liquid is boiled, about 3 min.

Add the whole chickpea mixture to the pot, mix well, cover and cook until it is boiled again, about 2 min, turn to low heat and cook for 10 min, stir every 2 min.

After 10 min, transfer the treatment into an airtight container, measure the weight and add water to 450.0g. Cover, label and keep it in the fridge.

Treatment 2 Pureed Canned Chickpea:

Pour whole canned chickpeas into a strainer, wash them in running water for 30 seconds and drain well.

Mix 148.8g of chickpeas with 104.7ml of filtered water and 0.31g of salt.

Blend the chickpea mixture in a food processor (KitchenAid Mod: KFP720OB2) for 60 seconds. Make the tomato sauce by combining 125.0g of tomato paste, spices, sugar, lemon juice and 110ml of water.

Cook the tomato sauce in an un-sticky cooking pan on medium heat until the liquid is boiled, about 3 min.

Add the pureed chickpea mixture to the pot, mix well, cover and cook until it is boiled again, about 2 min, turn to low heat and cook for 10 min, stir every 2 min.

After 10 min, transfer the treatment into an airtight container, measure the weight and add water to 450.0g. Cover, label and keep it in the fridge.

Treatment 3 Chickpea Powder:

Mix 49.7g of chickpea powder well with 202.9ml of water and 1.20g of Salt.

Make the tomato sauce by combining 125.0g of tomato paste, spices, sugar, lemon juice and 110.0 ml of water.

Cook the tomato sauce in an un-sticky cooking pan on medium heat until the liquid is boiled, about 3 min.

Add the chickpea powder mixture to the pot, mix well, cover and cook until it is boiled again, about 2 min, turn to low heat and cook for 10 min, stir every 2 min.

After 10 min, transfer the treatment into an airtight container, measure the weight and add water to 450.0g. Cover, label and keep it in the fridge.

Treatment 4 Whole Wheat Flour:

Mix 39.4g of whole wheat flour with 213.2ml of water and 1.21g of Salt.

Make the tomato sauce by combining 125.0g of tomato paste, spices, sugar, lemon juice and 110.0 ml of water.

Cook the tomato sauce in an un-sticky cooking pan on medium heat until the liquid is boiled, about 3 min.

Add the whole wheat mixture to the pot, mix well, cover and cook until it is boiled again, about 2 min, turn to low heat and cook for 10 min, stir every 2 min.

After 10 min, transfer the treatment into an airtight container, measure the weight and add water to 450.0g. Cover, label and keep it in the fridge.