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Stability of Selected B Vitamins in Thermally-Treated Pinto Beans

Virginia Anne West

A thesis submitted to the faculty of  
Brigham Young University  
in partial fulfillment of the requirements for the degree of  
Master of Science

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## ABSTRACT

### Stability of Selected B Vitamins in Thermally-Treated Pinto Beans

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Master of Science

Beans are a commonly consumed food and a staple in many regions worldwide. Pinto beans (*Phaseolus vulgaris*), categorized as legumes, are dried seeds from plants and are high in protein, carbohydrate and fiber, and low in fat. They are also a good source of various minerals and well as thiamin, riboflavin, niacin, vitamin B6, and folate. Beans are typically soaked and thermally processed before consumption. Different processing methods can impact the composition of beans. The purpose of this study was to examine the effect of thermal treatments on vitamin concentration in pinto beans. Beans were simmered, canned, dried-flaked, or dried-extruded, and measured for thiamin, riboflavin, folate, and vitamin B6. Beans were then reheated and measured again for vitamin concentration.

Vitamin loss was comparable between the most commonly consumed stages of processing: Simmered, canned reheated, dried-flaked reheated and dried-extruded reheated. The only statistically significant differences were that simmering caused the least amount of degradation of thiamin and dried-flaked product had the least amount of vitamin B6 degradation. Though dried-flaked and dried-extruded beans generally decreased in vitamin concentration, these two products were comparable to the simmered and canned reheated products. This suggests that drying is a nutritionally acceptable means of processing pinto beans, resulting in products that are more economical to transport and more convenient to prepare.

Keywords: pinto bean, thermal processing, thiamin, riboflavin, folate, vitamin B6, simmered, canned, flaked, extruded, dried

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## **Introduction**

Beans are consumed regularly in many countries and cultures. Latin America, Africa, and Asia account for over 75% of the yearly worldwide production of 18.8 metric tons [1]. The leading producers of beans worldwide are India, Brazil, Myanmar, United States of America, and Mexico and, together, account for 60% of the total world production. Bean classes produced differ based on region. In the United States, pinto beans are the main bean class produced [2].

Pinto beans (*Phaseolus vulgaris*), categorized as legumes, are dried seeds from plants. They are a food high in protein, carbohydrate, and fiber, and low in fat. They are also a good source of thiamin, riboflavin, niacin, vitamin B6, and folate, as well as various minerals [3].

Beans must be processed before consumption and different treatment methods can impact nutritional quality of beans [4]. Generally, there is a decrease in protein quality via loss of some amino acids [5]. Protein digestibility increases, however [5]. Starch crystalline structures, formed after cooking then cooling, slow digestion and the rate of glucose release into the blood [5]. Also during thermal processing, there is typically a loss in total dietary fiber [5]. Additionally, water-soluble vitamins and minerals are lost during processing.

In addition to canning and simmering, beans can be treated in different ways to create a refried bean product. One method involves cooking the beans, roller pressing the mixture, and then drying to form a product that has a flake-like appearance. This product will be referred to herein as “dried-flaked”. Another method involves cooking the beans under pressure and then releasing pressure to

form a dry product. This product will be referred to as “extruded dried”. Flaking and extrusion of beans are less common methods of processing but are desirable from the standpoint of consumer convenience because of the ease and speed of product preparation for consumption. Beans processed in this manner are also a lighter weight, decreasing transportation costs. These dried products may also have a longer shelf life. Because they are less common thermal treatment methods, however, there is little research regarding their effect on vitamin retention.

Wide variation exists in reported data of vitamin concentration in raw, simmered, and canned pinto beans. Reported thiamin in raw pinto beans ranges from 0.390 to 0.713 mg/100g [6-9]. Thiamin in simmered pinto beans ranges from 0.193 to 0.720 mg/100g [6,2,9]. Thiamin values reported for canned pinto beans range from 0.052 to 0.370 mg/100g [8,9].

Reported riboflavin values in raw pinto beans range from 0.140 to 0.280 mg/100g [6,7,2]. Simmered pinto bean values range from 0.062 to 0.122 mg/100g [6,2,9]. The USDA [9] reports 0.019 mg/100 g riboflavin in canned pinto beans.

Folate values reported for pinto beans vary widely as well [10]. Values for folate in raw pinto beans range from 190 to 525 ug/100g [8,9]. Folate values in simmered beans range from 110 to 172 µg/100g [8,2,9]. Folate in canned pinto beans range from 24 to 170 µg/100g [8,9].

Vitamin B6 values reported for raw pinto beans range from 0.474 to 0.690 mg/100g [8,9]. Vitamin B6 in simmered beans range from 0.299 to 0.400 mg/100g [8,9]. There is one report of 0.390 mg/100g [8] for vitamin B6 in canned pinto beans.

In general, thiamin, riboflavin, folate, and vitamin B6 are unstable to heat exposure. In contrast, niacin is very stable to thermal treatments [11]. Simmering and retort canning are common methods for processing beans and their effect on vitamin concentration has been studied, as discussed above. Leaching is the most common means of vitamin and mineral loss during simmering and canning [3,12,13]. A 48% thiamin and 29% riboflavin loss in beans has been reported during simmering [13]. Canning of beans led to a 67% loss of thiamin and a 48% loss of riboflavin [13]. Reported folate losses due to canning have ranged from no loss [14] to a 10% loss [8].

The first objective of this study was to measure the thiamin, riboflavin, folate, and vitamin B6 stability of thermally-treated (simmered, canned, dried-flaked, and dried-extruded) and reheated pinto beans in comparison to unprocessed dry beans. The second objective was to determine the vitamin stability of processed pinto beans prepared for consumption by reheating. It was hypothesized that vitamin concentration would decrease when beans were thermally processed and that vitamin concentration would further decrease with reheating of the processed beans.

## **Materials and Methods**

### **Bean Samples**

Raw and dried-flaked pinto beans, from the 2012 production year, were obtained from the LDS Church Welfare System (Salt Lake City, UT). Extruded dried pinto beans, and the raw beans from which they were produced, were obtained from a commercial source. One 11.3 kg bag of raw pinto beans was thoroughly

mixed then divided into ten 1.1 kg plastic, reclosable bags. Two 1.1 kg bags were randomly assigned to raw treatment (control), four 1.1 kg bags were assigned to simmering treatment, and four 1.1 kg bags were assigned to canning treatment. Likewise, an 11.3 kg bags of dried-flaked beans was mixed then divided into ten 1.1 kg plastic, reclosable bags. Two 1.1 kg bags were randomly selected for vitamin analysis. Samples of dry product were stored in the dark at room temperature until analysis.

### Simmered Samples

Beans from two of the randomly selected 1.1 kg bags were soaked for 14 hrs, following the method of [15], and 250 g were placed into a 3 L stainless steel pot with 600 mL of distilled water. The covered pot was placed on a range and heated until a boil was reached. Heat was adjusted throughout cooking process to maintain a simmer. Beans were cooked for 66 min, until they were soft to touch when pressed between the thumb and finger [16]. Solid and liquid portions were separated by draining on a #35 sieve until no liquid portion for one minute, then the solid portion was sampled immediately. Samples were kept at -80°C until analysis. This entire process was repeated on a different day using the remaining two, randomly selected 1.1 kg bags.

### Reheated Simmered Samples

After beans were simmered and held at 4°C for 24 hrs, they were reheated in a 1100 W microwave (General Electric, Louisville, KY). The beans were placed in a glass bowl, covered, and heated on high for 3.5 min. The beans were stirred and the temperature was taken. If the temperature was below 74°C, it was heated for 30 sec

intervals until this temperature was reached. Solid and liquid portions were separated using a #35 sieve and sampled immediately. Samples were kept at -80°C until analysis.

#### Canned Samples

Canned beans were prepared according to the LDS Church Welfare Services Processing protocol. Beans from two of the randomly selected 1.1 kg bags were soaked and approximately 200 g of soaked beans were placed into each of 8 cans (number 300 size). One teaspoon of salt was placed in each can. Distilled water was heated to about 63°C and poured into each can leaving a headspace of 0.8 cm. Cans were held until they reached 60°C. Cans that were below 60°C were placed in an exhaust box until 60°C was reached. Cans were then sealed and placed in the retort. The steam on the retort was turned on for 4 min to vent the pipes and air pockets. Once the retort reached 101°C, the steam vent was closed. When the retort reached 116°C and 621 kPa, the timer was started and the cans were left in the retort for 55 min. The cans were removed and placed directly into an ice water bath. The cans were shaken under the water to speed up the cooling process. Cans were stored at room temperature until sampling. Solid and liquid portions were separated using a #35 sieve and the solid portion was sampled immediately. Samples were kept at -80°C until analysis. The entire process was repeated on a different day with the two remaining 1.1 kg bags.

### Reheated Canned Samples

The contents of the can were heated in a stainless steel pot on a range and stirred until the temperature reached 74°C. Solid and liquid portions were separated using a #35 sieve and the solid portion was sampled immediately. Samples were kept at -80°C until analysis.

### Dried-Flaked Samples

Dried-flaked beans were processed commercially. Beans were cleaned, soaked, cooked for 70-95 min (depending on bean hardness), and then roller pressed at 103 kPa. After pressing, beans were dried to a moisture content of about 5% (Figure 1.). Samples were taken and stored in the dark at room temperature until analysis.



Figure 1: Photograph of dried-flaked pinto beans.

### Dried-flaked Reheated Samples

Dried-flaked beans were prepared according to package instructions by adding 114 g to 237 mL of boiling water in a steam table pan. The pan was covered for ten min

and the contents were stirred. Samples were taken immediately and kept at  $-80^{\circ}\text{C}$  until analysis.

#### Dried-extruded Samples

Dried-extruded beans were processed commercially. The raw beans were prepared by cleaning, soaking, and then cooking for 60 min at 151 kPa. After being extruded, they were dried to a moisture content of about 6% (Figure 2).



Figure 2: Photograph of dried-extruded pinto beans.

#### Dried-extruded Reheated Samples

The reheating of dried-extruded beans followed package instructions. A steam table was heated to  $82^{\circ}\text{C}$ . Water was boiled and 473 mL was poured into the pan followed by 191 g of dried-extruded bean. The beans were briefly stirred then covered for 25 min. The steam table was maintained at  $82^{\circ}\text{C}$  throughout the 25 min. The beans were thoroughly stirred and samples were taken immediately and kept at  $-80^{\circ}\text{C}$  until analysis.

## Vitamin Analysis

Thiamin, riboflavin, folate, and vitamin B6 were analyzed in each of the bean samples at each stage of processing. Thiamin and riboflavin were measured using the method of Arella et al. [17] with modifications from AOAC method 953.17 (AOAC 2006) and El-Arab et al. [18]. Low moisture samples (raw, dried-flaked, and dried-extruded) were ground for approximately 5 sec with a ZM 200 centrifugal grinder (Retsch, Newtown, PA). Higher moisture samples (simmered, canned, and reheated samples) were ground for 15 sec with a coffee grinder (Hamilton Beach, Glen Allen, VA). Five g of ground bean was measured into a 250 mL Erlenmeyer flask and 50 mL of 0.1 N HCl was added. One mL of octanol was added and then the sample was autoclaved at 121°C for 30 min. After cooling in an ice bath for 4 min, the pH was adjusted to 4.5 with 2.5 M sodium acetate. Next, 500 mg Taka-diazyme from *Aspergillus oryzae* (100 U/mg) was added and the flask was swirled. The flask was then covered and incubated at 37°C for 18 hrs. The bean suspension was filtered using Whatman #541 filter paper and a Buchner funnel and then diluted to 200 mL in a volumetric flask. Approximately 1 mL of this solution was filtered through a 0.2 µm membrane for riboflavin analysis on HPLC. Ten mL of the filtered solution was added to a 50 mL centrifuge tube and then 3 mL oxidizing reagent (1 mL of 1% potassium ferricyanide in 25mL volumetric flask and filled to volume with 15% NaOH) and 15 mL isobutanol were added to the tube. After shaking for 20 sec and then an additional 2 min, the tube was centrifuged (1200g, 4 min). One mL of supernatant was filtered through a 0.2 µm membrane and used for thiamin analysis on HPLC.



Separation and quantification of thiamin and riboflavin was performed isocratically using an Agilent 1100 Series HPLC (Agilent Technologies Inc., Santa Clara, CA), equipped with an octadecylsilyl column (150 mm x 4.60 mm, 5 µm particle size, Phenomenex Inc., Torrance, CA). The sample injection volume was 10 µL, and the mobile phase was methanol-0.05 M sodium acetate (30:70 v/v) with a flow rate of 1 mL/min. The analytes were detected using a fluorometric detector with excitation and emission wavelengths at 422 nm and 522 nm, respectively, for riboflavin, and 366 nm and 435 nm, respectively, for thiochrome.

Folate was measured using the AOAC trienzyme extraction method 2004.05 (AOAC 2006) with slight alterations from Chapman et al. [19]. Rat plasma (0.1mL) (male, non-sterile with lithium and heparin; Pel-Freez Biologicals, Catalog #36161-2, Rogers AR) with anticoagulant factors was used as the folate conjugase enzyme. The folic acid working standard was brought to a concentration of 2 ng/mL, instead of the 10ng/mL concentration used in the original method. Following filtration through Whatman 2V filter paper, samples were diluted with distilled water by an appropriate amount to have the samples fall within the standard curve.

The microbiological assay (*L. casei subsp. Rhamnosus*, ATCC #7469) of Tamura [20] was used with 96-well microtiter plates and minor modifications. Inoculum was maintained by transfers into fresh lactobacilli broth weekly. Prior to plating the samples, cultures were transferred to depletion media, prepared from lactobacilli broth and folic acid casei media following the method of Chen and Eitenmiller [21]

Vitamin B6 was measured by NP Analytical Laboratories (St. Louis, MO) using AOAC Method 961.5. The microbiological method used *Saccharomyces uvarum* ATCC 9080.

### Study Design

As shown in Table 1, the study was designed with treatment variables being process type (simmered, canned, dried-flaked, and dried-extruded) and process stage (raw, heated, and reheated) and process day, with sample and analytical replicates. Response variables measured were the amounts of thiamin, riboflavin, folate, and vitamin B6.

Table 1: Study design.

<u>Treatment Variables</u>	<u>Level</u>	
Process Type	4	Simmered, Canned, Dried-flaked, and Dried-extruded
Process Stage	3	Raw, Heated, Reheated
Day	2	
Sample Repetition	2	
Analytical Repetition	2	
Total number of samples analyzed	80*	

\*Raw samples did not have a sample replication

### Data Analysis

Data were analyzed for significance ( $p < 0.05$ ) using Statistical Analysis System software (Version 9.3 SAS Institute, Cary, NC). Vitamin data were analyzed for significant differences using multi-way analysis of variance. Significant differences among means were determined using post hoc Tukey pair-wise comparisons. The analysis used thiamin, riboflavin, folate, and vitamin B6 as

dependent variables and process type, stage, and day as independent variables. Day was not significant ( $p < 0.05$ ).

## **Results and Discussion**

### **Thiamin**

As seen in Figure 3, compared to its control, there was no significant thiamin decrease in simmered or simmered reheated beans. In comparison to its control, there was a significant decrease in canned beans but no significant decrease in canned reheated beans. There was a significant decrease of thiamin in dried-flaked and dried-flaked reheated compared to its control. Dried-extruded beans showed a significant decrease compared to its control while dried-extruded reheated showed no significant decrease compared to its control. There is significant decrease in thiamin content in dried-flaked beans followed by significant increase once the beans are reheated. Though not statistically significant, a similar pattern is observed in canned and dried-extruded process types. It may be that the thiamin is incompletely extracted in the processed product prior to reheating.

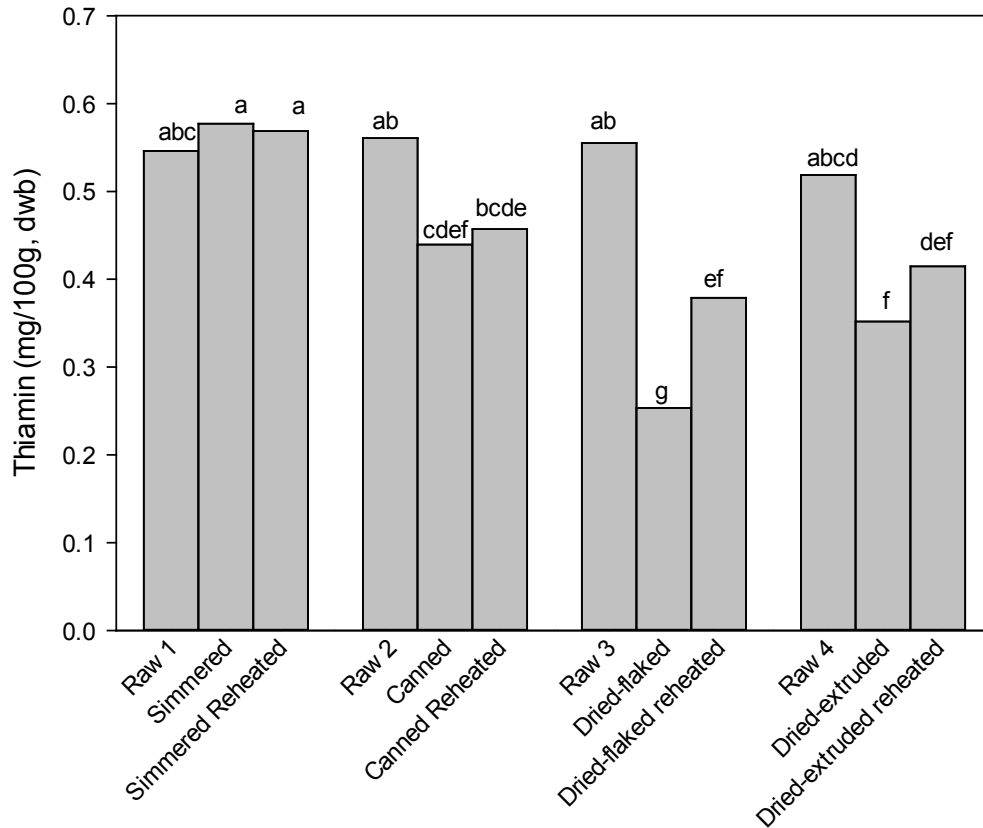


Figure 3: Thiamin concentration in raw, heated (simmered, canned, dried-flaked, and dried-extruded), and reheated pinto beans.

The most commonly consumed process stages are simmered, canned reheated, dried-flaked reheated, and dried-extruded reheated. There was no significant difference between canned reheated, dried-flaked reheated, and dried-extruded reheated. All three of these, however, were significantly lower than simmered.

The USDA Nutrient Database for Standard Reference reports nutritional data on a wet-weight basis for raw, boiled and canned pinto beans (no data is reported for dried pinto bean products). Utilizing the percent moisture reported, it is possible to calculate values on a dry-weight basis for comparative purposes.

Literature values for thiamin in pinto beans range from 0.39 to 0.713 mg/100g [6-9] and our values (0.519 to 0.561 mg/100g) fall within this range . For simmered beans, there is a reported range of 0.193 to 0.72 mg/100g [6,2,9] and our simmered bean value, 0.577 mg/100g, falls within this range. Literature values for thiamin in canned beans range from 0.052 to 0.370 mg/100g [8,9] while our value, 0.439 mg/100g, falls slightly above that range. Processing parameters may differ between the data sets and may account for at least some of the discrepancy.

No literature values exist for dried-flaked, dried-flaked reheated, dried-extruded, or dried-extruded reheated. Our thiamin values of, 0.253, 0.379, 0.352, 0.414 mg/100g respectively, are, evidently, the first to be reported for these dried bean products.

#### Riboflavin

Riboflavin showed no significant changes across all process stages for all process types (Figure 4). In the literature, riboflavin values for raw beans range from 0.14 to 0.28 mg/100g [6,7,2] and our values (0.207 to 0.228 mg/100g) fall within this range. Literature values for simmered beans range from 0.062-0.220 mg/100g [6,2,9] while our value of 0.224 mg/100g falls just above this range. The USDA value for riboflavin in canned beans is 0.019 mg/100g while ours is much higher at 0.276 mg/100g.

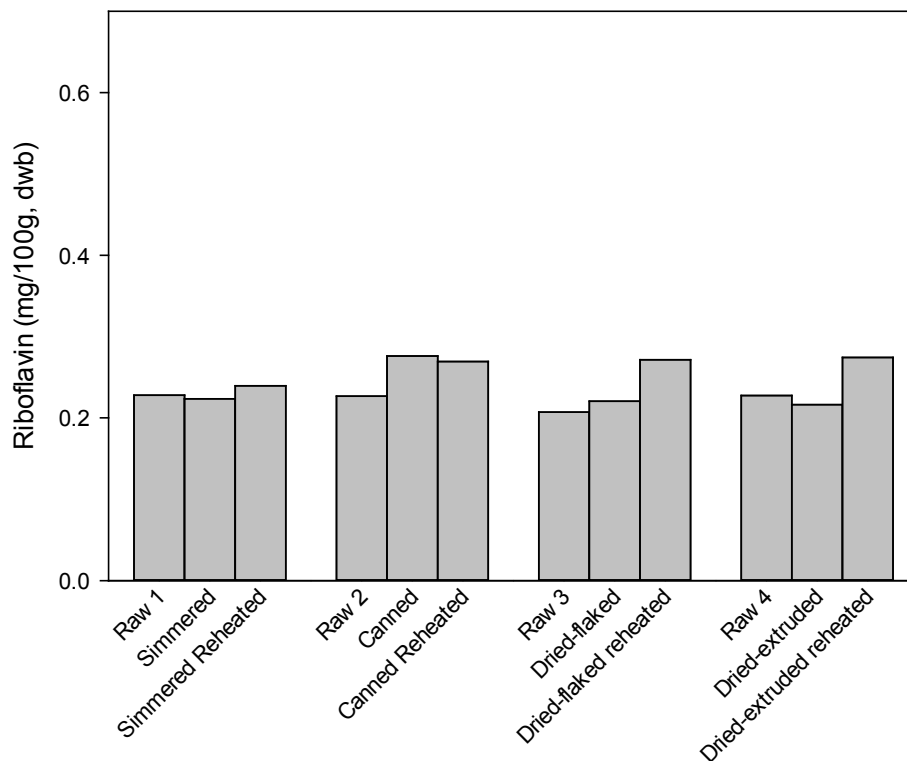


Figure 4: Riboflavin concentration in raw, heated (simmered, canned, dried-flaked, and dried-extruded), and reheated pinto beans. There are no significant differences between means.

No literature values exist for dried-flaked, dried-flaked reheated, dried-extruded, or dried-extruded reheated. Our riboflavin values of 0.221, 0.272, 0.216, 0.274 mg/100g respectively are, evidently, the first to be reported for these dried products.

### Folate

As shown in Figure 5, the folate values in the raw controls range from 183-336 ug/100g, with the dried-extruded raw control being significantly higher than the raw control for simmered and canned processes. The significant differences between the folate content of the raw controls suggests that raw pinto beans vary widely in folate content. This is reflected in the range of values reported in the

literature: 190-525 ug/100g [8,9]. The variation in the data is expected because of the wide variation in folate measurements. Even using an approved method, which is a well-known challenge in microbiological analyses of vitamins [22].

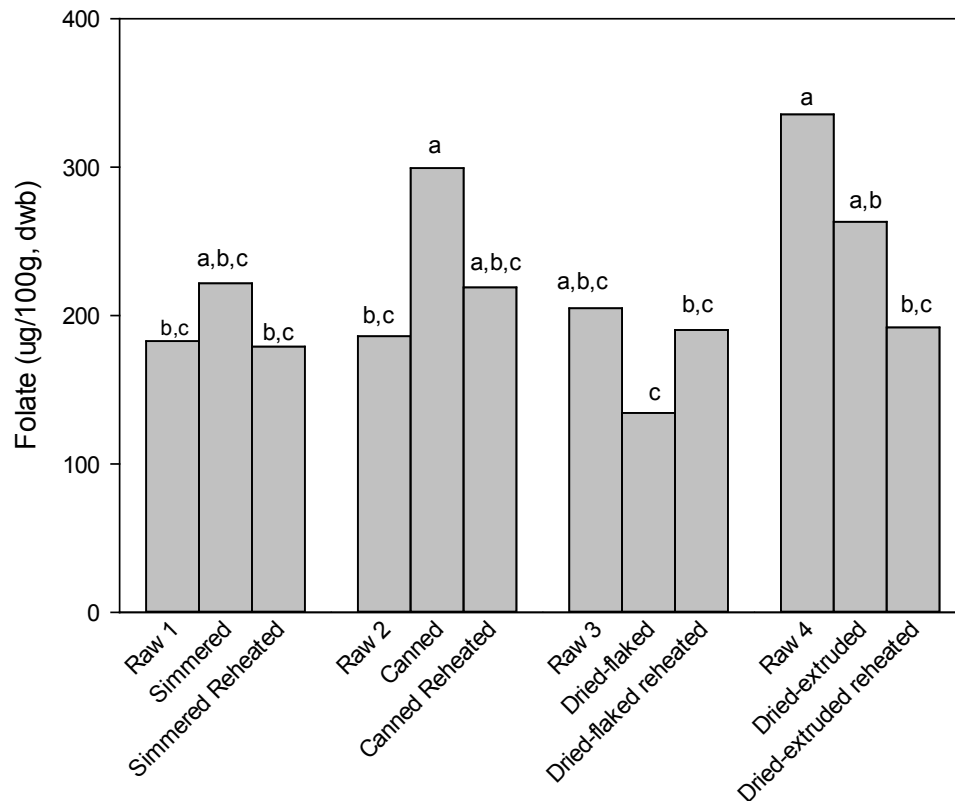


Figure 5: Folate concentration in raw, heated (simmered, canned, dried-flaked, and dried-extruded), and reheated beans.

Compared to its control, there was no significant decrease in simmered or simmered reheated beans. Canned beans showed a significant increase in folate, but canned reheated showed no significant decrease, compared to their control. Additionally, there was no significant decrease in dried-flaked or dried-flaked reheated compared to their raw. Though there was no significant decrease in dried-extruded compared to its control, there was a significant decrease in dried-extruded reheated.

Importantly there was no significant difference between simmered, canned reheated, dried-flaked reheated, and dried-extruded reheated samples, the commonly consumed products.

Literature values for simmered beans range from 110 to 190 ug/100g [8,2,9] and our value was higher, at 222 ug/100g. Values in canned pinto beans range from 24 to 170 ug/100g [8,9] while our value was also higher at 299 ug/100g.

No literature values exist for dried-flaked, dried-flaked reheated, dried-extruded, or dried-extruded reheated. Our folate values of 134, 190, 263, and 192 ug/100g respectively are, evidently, the first to be reported for these dried bean products.

#### Vitamin B6

As shown in Figure 6, compared to their raw controls, there was a significant decrease of vitamin B6 in all four process types.



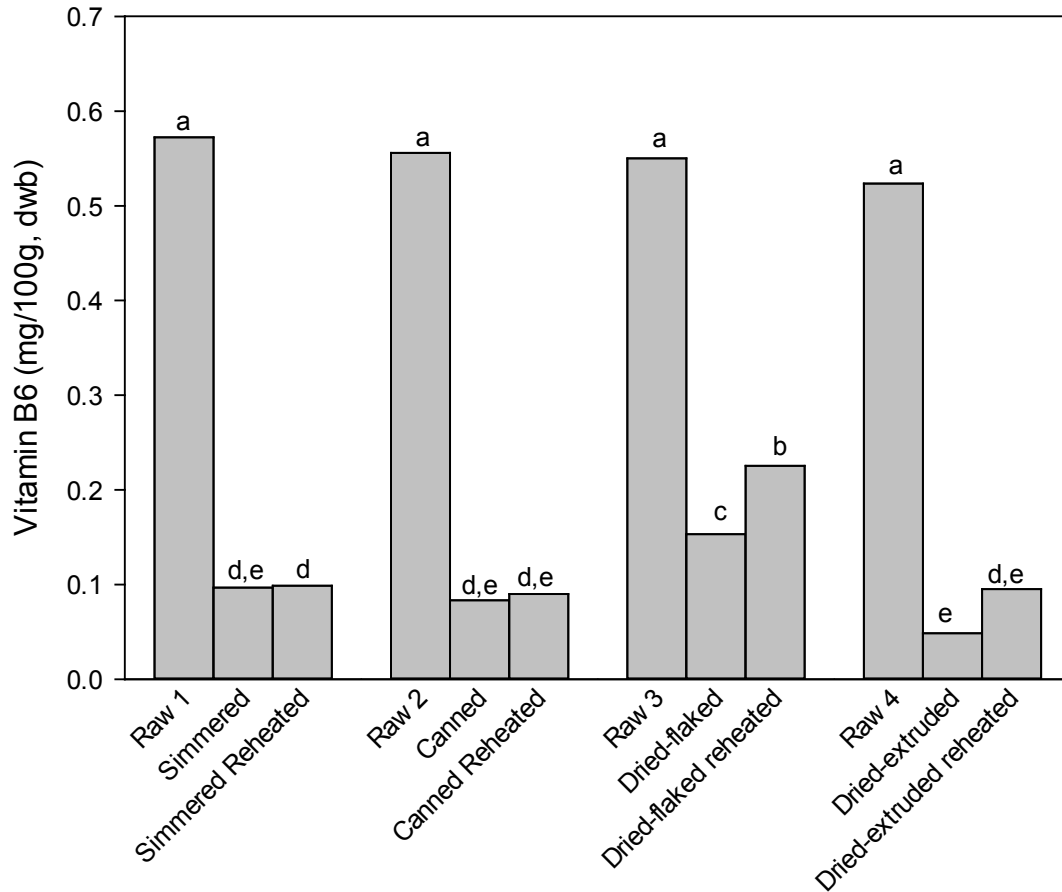


Figure 6: Vitamin B6 concentration in raw, heated (simmered, canned, dried-flaked, and dried-extruded), and reheated pinto beans.

Regarding the commonly consumed products, there was so no significant difference between simmered, canned reheated, and dried-extruded reheated. But all three of these products were significantly lower than dried-flaked reheated. As discussed previously, the reheating step may increase the extraction of vitamin B6 from its matrix.

Literature values for vitamin B6 in raw pinto beans range from 0.474 to 0.690 mg/100g [8,9] and our values (0.523 to 0.572 mg/100g) fall within this range. In simmered pinto beans, there is a range of 0.400 to 0.299 mg/100g [8,9]. Miller et

al. [8] reports a 0.39 mg/100g value in canned pinto beans compared to the much lower 0.083 mg/100g obtained in our study.

No literature values are reported for dried-flaked, dried-flaked reheated, dried-extruded, or dried-extruded reheated. Our vitamin B6 values of 0.153, 0.225, 0.048, and 0.095 mg/100g respectively are, evidently, the first to be reported for these dried bean products.

## **Conclusion**

Vitamin loss was comparable between the most commonly consumed stages of processing: Simmered, canned reheated, dried-flaked reheated and dried-extruded reheated. The only statistically significant differences were that simmering caused the least amount of degradation of thiamin and dried-flaked product had the least amount of vitamin B6 degradation.

Though dried-flaked and dried-extruded beans showed some loss in vitamin concentration, these two products were comparable to the simmered and canned reheated products. This suggests that drying is a nutritionally acceptable means of processing pinto beans, resulting in products that are more economical to transport and more convenient to prepare.

Further work should investigate the possibility that reheating results in an increase of vitamin bioavailability for canned and dried pinto bean products. Because of the wide variations in reported vitamin concentrations, these results should be compared with future studies of vitamin stability in thermally-treated pinto beans.

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## APPENDIX A: EXPANDED LITERATURE REVIEW

### Bean Varieties and Consumption

Pinto beans (*Phaseolus vulgaris*), categorized as legumes, are dried seeds from plants. They are also known as pulses. It is thought by historians that dry beans were first domesticated in Peru and Mexico more than 7,000 years ago [23] [24] [25] and then slowly introduced to other parts of the world. Of the approximate 13,000 species of legumes, only about 20 are commonly consumed by humans [26]. The most common bean classes include Black Beans, Cranberry Beans, Great Northern Beans, Red Kidney Beans, Navy Beans, and Pinto Beans. The leading producers of dry beans, accounting for 60% of the world production, are India, Brazil, Myanmar, United States, and Mexico. The dry bean yield increased from 0.54 million metric tons (MT) per hectare in 1980 to 0.73 MT/hectare in 2008. Within the United States, North Dakota, Michigan, Nebraska, Minnesota, and Idaho are the top bean producing states. Pinto and Navy beans are the leading dry bean classes produced in the United States. Changing demographics in the United States are thought to be the reason for changes in bean class production. For example, there has been a rise in Garbanzo bean production due to the increase in populations of Hispanic and Asian origins. About a quarter of the beans produced in the United States are exported to other countries [12].

Bean consumption in the United States reached a high of 9.6 pounds per person a year between 1941-1943. A low of 5.5 pounds per person was reached during 1978-90. Since that time there has been a rise in consumption, with an

average of 7.7 pounds per person in 1992-94 and 6.5 pounds per person during 2006-08. It is important to note that dry beans have not been included in Federal price support programs since the late 1960s. The Food Conservation and Energy Act of 2008 (2008 Farm Act) included large chickpeas in programs such marketing assistance loans, loan deficiency payments, and counter-cyclical payments. Also, the USDA now buys dry packed and canned beans for school lunch, child nutrition, and other feeding programs [27].

### Bean Processing Methods

Canning is the most common processing procedure used in bean product manufacturing. This includes beans canned in brine, in combination with meat stews, chilies, refried beans, pork and beans, as well as baked beans [12]. Other processing methods for dry beans have been reported, including bean flour [28], extruded products [29], minimally processed sugar-coated beans [30], spaghetti [31], pasta [32], tortilla [33], snack bars [34], use in beef sausages [35], and intermediate-moisture foods [36]. Bean-based products use bean flour processed by different methods. Bean flours can be used to improve nutritional quality of a variety of processed foods [37]. Potential uses of dry bean products are continuously being explored and expanded [12]. Furthermore, there was a 516% increase in production of gluten-free foods in the United States from 2003 to 2008 and a greater rise expected in the future. Bean flour is common in these product and use is expected to rise with the increasing demand for gluten free products [38].

### Bean Nutrient Content

Dry beans of different varieties are compositionally and nutritionally fairly similar [39]. The variations in vitamin contents of beans in different classes and within the same class are likely due to differences in analytical methods, sampling differences, and different sample preparation techniques. Beans cannot be eaten raw; they must be processed before consumption. Different processing methods can cause different effects on the composition beans. [2] Much of the nutrition research relevant to beans focuses on protein, carbohydrate, and mineral content and changes that occur during processing. For example, Eyaru et al. [40] studied the effect of common domestic processing methods, splitting, soaking, boiling, and pressure-cooking on starch content in Red kidney beans and Yellow and Green beans. Soaking led to a reduced amount of total starch, while boiling and pressure-cooking increased the total starch.

Dry beans have an average composition of 21-25% crude protein [3]. Protein content in beans is of interest because of the adverse effects of high fat animal protein sources [41]. However, protein digestibility of cooked dry beans is of lower quality than animal proteins. Bean protein is rich in the essential amino acid lysine but deficient in essential amino acids methionine and tryptophan [42,43]. Rich in dietary fiber and low in fat [41], regular intake of beans can contribute to the lowering of cholesterol [44]. Consistent consumption of legumes in the diet can lower the risk for common metabolic diseases, such as diabetes mellitus, coronary heart disease (CHD) and cancer [45]. In regards to minerals, dry beans are considered good sources of calcium, iron, copper, zinc, phosphorus, potassium, and magnesium. One serving of dry beans (1 cup cooked) can provide 29% of the RDA

for iron for females and 55% for males, 10-15% of phosphorus, magnesium and manganese, about 20% of potassium and copper, and 10% of calcium and zinc [2].

Regarding vitamins, dry beans are an excellent source of thiamin, riboflavin, niacin, vitamin B6, and folic acid [3]. Though cooking decreases the amounts of these vitamins, cooked beans are still a significant source of some of these vitamins. Thermal processing in cans dramatically decreases the quantity of these vitamins in pinto beans (See Table 1).

Table 1: USDA vitamin values in raw, cooked, and canned Pinto Beans [9].

		Thiamin (mg/100g)	Riboflavin (mg/100g)	Folate (DFE, µ)	Vitamin B-6 (mg/100g)
Pinto Beans	Raw	0.713	0.212	525	0.474
	Cooked	0.193	0.062	172	0.229
	Canned	0.052	0.019	24	--

### General Vitamin Stability

The general stability of vitamins under various conditions is shown in Table 2. This table is a generalization, however, and does not represent the vitamin stability under all circumstances. Stability can vary in different forms of each vitamin. For example, tetrahydrofolic acid and folic acid are both folates with almost identical nutritional properties. Tetrahydrofolic acid, however, is prone to oxidation whereas folic acid is very stable.

Nutrient	Neutral	Acid	Alkaline	Air or Oxygen	Light	Heat	Maximum Cooking Loss (%)
Vitamin A	S	U	S	U	U	U	40
Ascorbic Acid	U	S	U	U	U	U	100
Biotin	S	S	S	S	S	U	60
Carotenes	S	U	S	U	U	U	30



Choline	S	S	S	U	S	S	5
Vitamin B12	S	S	S	U	U	S	10
Vitamin D	S	S	U	U	U	U	4
Folate	U	U	U	U	U	U	100
Vitamin K	S	U	U	S	U	S	5
Niacin	S	S	S	S	S	S	75
Pantothenic acid	S	U	U	S	S	U	50
Vitamin B6	S	S	S	S	U	U	40
Riboflavin	S	S	U	S	U	U	75
Thiamin	U	S	U	U	S	U	80
Tocopherols	S	S	S	U	U	U	55

U indicates unstable, S indicates stable

Table 2: Vitamin Stability [46]

Thiamin is widely distributed in plant and animal tissues and exists naturally in foods in the form of thiamin pyrophosphate. It is relatively stable to oxidation and light but is among the least stable of the vitamins when in a neutral or alkaline pH. Next to vitamin C, thiamin is the least stable of the vitamins to heat processing [11]. Thiamin loss can also occur from leaching into surrounding aqueous media, exposure to sulfiting agents, and fully hydrated foods in storage. Thiamin shows excellent stability in low water activities at ambient temperatures. The form of thiamin also influences its stability. Free thiamin and thiamin pyrophosphate exhibit the same rates of thermal degradation at pH 4.5 but with an increase in pH, thiamin pyrophosphate exhibits a degradation rate three times faster than free thiamin [46].

Riboflavin exhibits greatest stability in an acidic environment, is less stable at a neutral pH, and degrades quickly in an alkaline medium. During conventional thermal processing, handling, and preparation, riboflavin stability is moderate to very good [46].

The naturally occurring forms of folate are polyglutamyl species of 5,6,7,8-tetrahydrofolate and are assumed to make up approximately 50-80% of naturally occurring dietary folate. Folate stability in food can be difficult to predict; it is dependent on leaching, forms of folate present, and the environment in which it is present [46].

#### Vitamin Stability During Thermal Processing

The amount of any vitamin loss during cooking depends on the type of food, the variety of food, the cutting technique, preparation, and duration and method of cooking [46]. Improper cooking technique can lead to large losses of vitamins as a result of leaching. In general, a major source of vitamin and mineral loss is the final preparation step before consumption. Vitamin losses can be minimized during cooking by using proper cooking procedures. This includes refraining from over chopping or excessive trimming, cooking in covered pans, not using excessive water, preventing food from being overcooked, and using cooking water for soups [47]. Cooking in conditions that minimize exposure to oxygen and excess liquid minimizes oxidation and leaching of vitamins and minerals [46].

Processing food makes it possible to consume safe products year round that are organoleptically acceptable. Processing can cause changes in the color, consistency, flavor, and nutritive value of a food [13]. For instance, heat processing can cause desirable changes in food products, including increased digestibility, palatability, storage capacity, and/or functionality as ingredients [48].

Beans are usually purchased dry (raw), frozen, canned, flaked, or extruded. Drum drying is used to create texture in foods and is widely used in the production

of instant flours. It is an economical operation for dehydrating food purees. Drum dried products have high nutritional and textural qualities [49]. Little is known regarding the nutritional quality of extruded bean products.

Though this study focuses on pinto beans, other bean varieties and foods will be discussed in the remainder of this review due to limited literature on vitamin stability in pinto beans products.

#### Thiamin and Riboflavin Stability During Thermal Processing

Canning appears to be much more destructive to thiamin and riboflavin than freezing because of the thermal processing and the aqueous environment in canning. The effect of freezing and canning on thiamin and riboflavin stability in immature seeds of five cultivars of common beans was evaluated. Blanching of the seeds before freezing reduced the content of thiamin and riboflavin by 18-42% and 18-26% respectively. Cooking to consumption consistency before freezing resulted in reductions of 43-54% and 22-32%. Vitamin retention was lower in the canned samples. Thiamin content was 23-36% and 31-48% lower in canning than in the two freezing methods, while riboflavin content was 11-30% and 14-36% lower. After one year of storage, the two freezing methods showed less thiamin and riboflavin loss (70-82% and 48-68% loss and 57-78% and 39-69% loss) than the canned methods (78-87% and 54-77% loss). The loss of thiamin and riboflavin during canning was credited to leaching into the liquid portion of the canned beans [13].

Thiamin and riboflavin levels were studied in immature grass pea seeds during freezing and canning. Blanching and freezing led to a 6.5% and 22% loss in

thiamin and riboflavin respectively. Cooking of the frozen peas led to an additional 25% and 22% loss. Canning caused more of a loss, with a 58% and 50% reduction of thiamin and riboflavin from the raw seeds. Values for cooked, canned peas were not reported [50].

Soaking of beans is a common practice to reduce cooking times. There is, however, a concern of vitamin loss during this stage. Thiamin content of faba beans did not change when soaked in an acid solution. There was a 15% decrease, however, when soaking in a water or basic solution. Riboflavin decreased 11% in the beans after soaking in a basic solution but did not change in water or an acidic solution. After cooking the beans soaked in an acidic solution, water, and a basic solution, thiamin decreased 19, 24, at 35% respectively while the riboflavin content did not change. Soaking in the basic solution increased thiamin and riboflavin loss during soaking and cooking. Soaking in an acidic solution was the best for thiamin and riboflavin retention in faba beans [51].

Vitamin loss is also reported in products enriched with thiamin and riboflavin. Watanabe, Ciacco [52] studied thiamin and riboflavin stability during the cooking of spaghetti. The spaghetti dough was enriched with thiamin at four levels (0.943, 1.283, 0.513, and 1.743mg/100g) and with riboflavin at four levels (0.447, 0.647, 0.767, and 0.887mg/100g). During cooking, thiamin levels were reduced to 0.325mg/100g (59% loss), 0.434mg/100g (59% loss), 0.521mg/100g (58% loss), and 0.636mg/100g (57% loss) respectively. Riboflavin levels were reduced to 0.170mg/100g (52% loss), 0.236mg/100g (51% loss), 0.272mg/100g (55% loss),

and 0.298mg/100g (55% loss) respectively. The level of added thiamin or riboflavin did not affect the percent of thiamin lost during cooking.

Leaching into cook water is a concern in all food products that involve immersion in water during cooking. Vandrasek, Warthesen [53] studied the effects of cooking on thiamin loss in rice (brown and enriched white) and pasta. The original thiamin content was 6.6 µg/g for the white rice and 3.2 µg/g for the brown rice. Samples of cooking water were taken throughout the cooking process. During cooking, thiamin seemed fairly stable to heat but significant amounts were lost due to leaching. After five min of cooking the white rice, 65% of the thiamin had leached into the water. Upon completion of the cooking, however, the rice had reabsorbed some of the water containing the thiamin, leaving only 50% of the original thiamin content in the cooking water. In the unenriched brown rice, the leaching of thiamin into cooking water was much slower. By the end of the cooking period, 44% of the native thiamin had leached into the cooking water.

The loss of thiamin in enriched products can be different from the loss of native thiamin. Thiamin stability was tested in enriched egg noodles, elbow pasta, spinach ribbon, and their whole-wheat counterparts. Initial contents of the whole-wheat pasta were less than 50% of the enriched products. The sum of the thiamin recovered from the cooked pasta and their corresponding cooking waters ranged from 96 to 104%, relative to the thiamin amount in the raw counterparts. This indicates that very little thiamin degradation due to heat exposure occurred. Thiamin retention of all the cooked pastas ranged from 47 to 66% with whole-wheat pastas retaining slightly more. There were significant variations in retention

among the types of pasta, indicating that pasta type or composition is an important factor in thiamin leaching [53].

Augustin et al. [54] compared riboflavin retention in potatoes boiled with and without skins, baked with skins, and microwave cooked. The raw potato contained 0.143 mg/150g riboflavin. Microwave cooking was the best method, conserving 0.051mg/150g (64% loss) of riboflavin. Boiling the potatoes without skins proved to be the worst method, with only 0.034mg/150 g of riboflavin retained (76% loss).

It appears that while heat degradation is a concern, the majority of thiamin and riboflavin, both water-soluble vitamins, loss in cooking is due to leaching into cooking water during boiling or in the liquid portion of the canned food.

#### Folate Stability During Thermal Processing

Changes in folic acid contents during thermal processing of peas, lentils, white and colored beans, and soybeans were monitored. During pretreatment of legumes, there was a partial loss of folic acid due to leaching. Cooking the legumes in the soaking water seemed to minimize folate losses. High losses of folate were seen during microwave cooking [55]. Previous studies have indicated that a large portion of the folate in beans is lost during the canning process [56] [57] [58].

Like thiamin and riboflavin, leaching is also a concern with folate. Xue et al. [59] studied the effect of soaking, ordinary cooking, and pressure cooking on folate concentrations in navy beans. Raw navy beans contained 0.82µg/g folate. With soaking times of 3, 6, 9, and 12 hrs and a seed to water ratio of 1:3, the folate concentrations were 13.86 (1.1% reduction), 13.16 (6.1% reduction), 10.81 (22.8%

reduction), and 9.74 µg/g (30.5% reduction) respectively. With the same soaking times but a high ratio of seed to water, 1:7, the folate concentrations were 13.28 (5.2% reduction), 12.03 (14.1% reduction), 8.71 (37.8% reduction), and 7.79µg/g (44.7% reduction). The results suggested that the seed to water ratio has more pronounced effects than soaking time when the beans are soaked for under 6 hours. Past 6 hours, increasing the amount of water and the soaking time caused a higher degradation of folate. A short soaking time combined with a limited amount of soaking water favored folate retention in the beans.

Delchier et al. [60] reported the content of folate in spinach before and after cooking. After boiling fresh spinach (which had 1.73 mg/kg), the folate concentration was 0.89mg/kg (49% loss). There were high concentrations of folate in the cooking water (0.26mg/L), indicating it had leached into the water during cooking. There was not a significant difference (6% loss) in folate during steaming of spinach. Frozen spinach was boiled and maintained only 42% of the folate concentrations of the fresh spinach.

McKillop et al. [61]reported a 51% loss of folate in spinach and a 56% loss of folate in broccoli during boiling the raw vegetable. Steaming showed no significant folate loss in spinach or broccoli. Length of cooking time also affected folate retention. There was a significant decrease in folate retention the longer the cooking time (up to 16 min) for boiling spinach and broccoli. There was no significant decrease, however, during steaming of the vegetables for longer periods of time (up to 16 min).

Augustin et al. [54] compared folate retention in potatoes boiled with and without skins, baked with skins, and microwaved cooked. The raw potato initially contained 19.2 mg/150 g folic acid. Microwave cooking conserved 17.6mg/150 g (8% loss) folic acid. Boiling the potatoes without skins proved to be the worst method. This method retained 14.7mg/150g (23% loss) of folic acid.

It can be concluded that folate stability is relevant to a variety of conditions and can vary with only slight differences in food. Generally, cooking seems to contribute to some loss in food. Soaking time and amount of water seem to be the biggest factors in folate loss.



## APPENDIX B: EXPANDED MATERIALS AND METHODS

### Simmering

The standardized method of Romero Del Castillo et al. [15] was followed to prepare beans for simmering. Two hundred fifty g samples of dry beans were placed in a glass, wide mouth jar. Seven hundred fifty mL of distilled water was added and the beans were soaked for 14 hrs in the dark. After, beans were drained and ready for processing.

### Canning

The standardized method of Romero Del Castillo et al. [15] was followed to prepare beans for canning. Two hundred fifty g samples of dry beans were placed in a glass, wide mouth jar. Seven hundred fifty mL of distilled water was added and beans were soaked for 14 hrs in the dark. After, beans were drained and ready for canning.

The swell factor of the beans was calculated to determine the fill weight, lower control limit, and upper control limit of the cans. One hundred twenty five g of beans were soaked in 375 mL of distilled water in a glass jar for 14 hrs. One hundred dry beans were counted out and weighed. After soaking, 100 beans were counted out and weighed. The swell factor was determined by the weight of soaked beans divided by the weight of the dry beans. The swell factor was determined to be 2.

### Vitamin Analysis

Vitamin B6 was measured and reported by NP Analytical Laboratories (St. Louis, MO). The microbiological method used *Saccharomyces uvarum* ATCC 9080, which requires vitamin B6 for growth. A nutritionally complete (except vitamin B6)

basal medium was used as the diluent for the final dilutions of the standards and samples. After incubation, percent transmittance was used to measure growth response of the bacterial cultures. A dose-response line was created and the sample concentrations were calculated.

APPENDIX C: ADDITIONAL TABLES

		Thiamin mg/100g	Riboflavin mg/100g	Folate ug/100g	Vitamin B6 mg/100g
Simmered	Raw	0.5461	0.228	182.83	0.5722
	Processed	0.5771	0.2236	221.81	0.09669
	Reheated	0.5687	0.2395	179.16	0.09876
Canned	Raw	0.5607	0.2267	186.06	0.5559
	Processed	0.4394	0.2761	299.39	0.08332
	Reheated	0.4572	0.2693	219.06	0.08997
Flaked	Raw	0.5551	0.2071	204.98	0.5502
	Processed	0.2534	0.2206	134.37	0.1531
	Reheated	0.3789	0.2716	190.31	0.2254
Extruded	Raw	0.5187	0.2275	335.59	0.5234
	Processed	0.352	0.2164	263.31	0.04842
	Reheated	0.4146	0.2744	192.06	0.09508

Table 1: Thiamin, riboflavin, folate, and vitamin B6 values for three stages (raw, processed, and reheated) of four bean processing methods (simmered, canned, flaked, and extruded).

## APPENDIX D: STATISTICAL ANALYSIS

The SAS System 10:07 Wednesday, November 26, 2014 134

The Mixed Procedure

Model Information

Data Set	WORK.IN
Dependent Variable	Thiamin
Covariance Structure	Variance Components
Estimation Method	REML
Residual Variance Method	Profile
Fixed Effects SE Method	Model-Based
Degrees of Freedom Method	Containment

Class Level Information

Class	Levels	Values
large_bag	5	1 2 3 5 6
small_bag	16	1 2 3 4 5 7 8 9 10 12 13 14 15 17 18 19
Process	3	Canned Flaked Simmered
Stage	3	Processed Raw Reheated

Dimensions

Covariance Parameters	3
Columns in X	16

Columns in Z	44
Subjects	1
Max Obs Per Subject	56

Number of Observations

Number of Observations Read	56
Number of Observations Used	56
Number of Observations Not Used	0

Iteration History

Iteration	Evaluations	-2 Res Log Like	Criterion
0	1	-129.35887073	
1	3	-129.98409683	0.00103580
2	2	-130.07308935	0.00006372
3	1	-130.08022455	0.00000019
4	1	-130.08024488	0.00000000

The Mixed Procedure

Convergence criteria met.

Covariance Parameter Estimates

Cov Parm	Estimate
small_bag*Process	0
small_bag*Stage	0.000489
Residual	0.002304

Fit Statistics

-2 Res Log Likelihood	-130.1
AIC (smaller is better)	-126.1
AICC (smaller is better)	-125.8
BIC (smaller is better)	-124.5

Type 3 Tests of Fixed Effects

Effect	Num DF	Den DF	F Value	Pr > F
Process	2	10	45.32	<.0001
Stage	2	9	11.47	0.0034
Process*Stage	4	28	5.66	0.0018

Least Squares Means

Effect	Process	Stage	Estimate	Standard Error	DF	t Value	Pr >  t
Process	Canned		0.4865	0.01654	10	29.41	<.0001
Process	Flaked		0.3689	0.01654	10	22.31	<.0001
Process	Simmered		0.5721	0.01350	10	42.37	<.0001
Stage		Processed	0.4233	0.01169	9	36.19	<.0001
Stage		Raw	0.5360	0.02135	9	25.10	<.0001
Stage		Reheated	0.4683	0.01169	9	40.04	<.0001
Process*Stage	Canned	Processed	0.4394	0.02026	28	21.69	<.0001
Process*Stage	Canned	Raw	0.5629	0.04051	28	13.89	<.0001
Process*Stage	Canned	Reheated	0.4572	0.02026	28	22.57	<.0001
Process*Stage	Flaked	Processed	0.2534	0.02026	28	12.51	<.0001
Process*Stage	Flaked	Raw	0.4745	0.04051	28	11.71	<.0001
Process*Stage	Flaked	Reheated	0.3789	0.02026	28	18.70	<.0001
Process*Stage	Simmered	Processed	0.5771	0.02026	28	28.49	<.0001
Process*Stage	Simmered	Raw	0.5706	0.02865	28	19.92	<.0001
Process*Stage	Simmered	Reheated	0.5687	0.02026	28	28.08	<.0001

The Mixed Procedure

Differences of Least Squares Means

Effect	Process	Stage	Process	Stage	Estimate	Standard Error	DF	t Value
Process	Canned		Flaked		0.1176	0.02339	10	5.03
Process	Canned		Simmered		-0.08565	0.02135	10	-4.01
Process	Flaked		Simmered		-0.2032	0.02135	10	-9.52
Stage		Processed		Raw	-0.1127	0.02434	9	-4.63
Stage		Processed		Reheated	-0.04499	0.01654	9	-2.72
Stage		Raw		Reheated	0.06774	0.02434	9	2.78
Process*Stage	Canned	Processed	Canned	Raw	-0.1235	0.04529	28	-2.73
Process*Stage	Canned	Processed	Canned	Reheated	-0.01784	0.02865	28	-0.62
Process*Stage	Canned	Processed	Flaked	Processed	0.1860	0.02865	28	6.49
Process*Stage	Canned	Processed	Flaked	Raw	-0.03513	0.04529	28	-0.78
Process*Stage	Canned	Processed	Flaked	Reheated	0.06049	0.02865	28	2.11
Process*Stage	Canned	Processed	Simmered	Processed	-0.1377	0.02865	28	-4.81
Process*Stage	Canned	Processed	Simmered	Raw	-0.1313	0.03508	28	-3.74
Process*Stage	Canned	Processed	Simmered	Reheated	-0.1293	0.02865	28	-4.52
Process*Stage	Canned	Raw	Canned	Reheated	0.1057	0.04529	28	2.33
Process*Stage	Canned	Raw	Flaked	Processed	0.3095	0.04529	28	6.83
Process*Stage	Canned	Raw	Flaked	Raw	0.08838	0.05729	28	1.54
Process*Stage	Canned	Raw	Flaked	Reheated	0.1840	0.04529	28	4.06
Process*Stage	Canned	Raw	Simmered	Processed	-0.01418	0.04529	28	-0.31
Process*Stage	Canned	Raw	Simmered	Raw	-0.00776	0.04962	28	-0.16
Process*Stage	Canned	Raw	Simmered	Reheated	-0.00584	0.04529	28	-0.13
Process*Stage	Canned	Reheated	Flaked	Processed	0.2038	0.02865	28	7.11
Process*Stage	Canned	Reheated	Flaked	Raw	-0.01728	0.04529	28	-0.38
Process*Stage	Canned	Reheated	Flaked	Reheated	0.07833	0.02865	28	2.73
Process*Stage	Canned	Reheated	Simmered	Processed	-0.1198	0.02865	28	-4.18
Process*Stage	Canned	Reheated	Simmered	Raw	-0.1134	0.03508	28	-3.23
Process*Stage	Canned	Reheated	Simmered	Reheated	-0.1115	0.02865	28	-3.89



Process*Stage	Flaked	Processed	Flaked	Raw	-0.2211	0.04529	28	-4.88
Process*Stage	Flaked	Processed	Flaked	Reheated	-0.1255	0.02865	28	-4.38
Process*Stage	Flaked	Processed	Simmered	Processed	-0.3237	0.02865	28	-11.30
Process*Stage	Flaked	Processed	Simmered	Raw	-0.3172	0.03508	28	-9.04
Process*Stage	Flaked	Processed	Simmered	Reheated	-0.3153	0.02865	28	-11.01
Process*Stage	Flaked	Raw	Flaked	Reheated	0.09562	0.04529	28	2.11
Process*Stage	Flaked	Raw	Simmered	Processed	-0.1026	0.04529	28	-2.26
Process*Stage	Flaked	Raw	Simmered	Raw	-0.09614	0.04962	28	-1.94
Process*Stage	Flaked	Raw	Simmered	Reheated	-0.09422	0.04529	28	-2.08
Process*Stage	Flaked	Reheated	Simmered	Processed	-0.1982	0.02865	28	-6.92
Process*Stage	Flaked	Reheated	Simmered	Raw	-0.1918	0.03508	28	-5.47
Process*Stage	Flaked	Reheated	Simmered	Reheated	-0.1898	0.02865	28	-6.63
Process*Stage	Simmered	Processed	Simmered	Raw	0.006420	0.03508	28	0.18
Process*Stage	Simmered	Processed	Simmered	Reheated	0.008342	0.02865	28	0.29
Process*Stage	Simmered	Raw	Simmered	Reheated	0.001923	0.03508	28	0.05

The Mixed Procedure

Differences of Least Squares Means

Effect	Process	Stage	Process	Stage	Pr >  t	Adjustment	Adj P
Process	Canned		Flaked		0.0005	Tukey-Kramer	0.0014
Process	Canned		Simmered		0.0025	Tukey-Kramer	0.0063
Process	Flaked		Simmered		<.0001	Tukey-Kramer	<.0001
Stage		Processed		Raw	0.0012	Tukey-Kramer	0.0032
Stage		Processed		Reheated	0.0236	Tukey-Kramer	0.0559
Stage		Raw		Reheated	0.0213	Tukey-Kramer	0.0508
Process*Stage	Canned	Processed	Canned	Raw	0.0109	Tukey-Kramer	0.1837
Process*Stage	Canned	Processed	Canned	Reheated	0.5384	Tukey-Kramer	0.9993
Process*Stage	Canned	Processed	Flaked	Processed	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Processed	Flaked	Raw	0.4445	Tukey-Kramer	0.9967
Process*Stage	Canned	Processed	Flaked	Reheated	0.0438	Tukey-Kramer	0.4858
Process*Stage	Canned	Processed	Simmered	Processed	<.0001	Tukey-Kramer	0.0013
Process*Stage	Canned	Processed	Simmered	Raw	0.0008	Tukey-Kramer	0.0202
Process*Stage	Canned	Processed	Simmered	Reheated	0.0001	Tukey-Kramer	0.0029
Process*Stage	Canned	Raw	Canned	Reheated	0.0271	Tukey-Kramer	0.3573
Process*Stage	Canned	Raw	Flaked	Processed	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Raw	Flaked	Raw	0.1341	Tukey-Kramer	0.8260
Process*Stage	Canned	Raw	Flaked	Reheated	0.0004	Tukey-Kramer	0.0092
Process*Stage	Canned	Raw	Simmered	Processed	0.7565	Tukey-Kramer	1.0000
Process*Stage	Canned	Raw	Simmered	Raw	0.8768	Tukey-Kramer	1.0000
Process*Stage	Canned	Raw	Simmered	Reheated	0.8983	Tukey-Kramer	1.0000
Process*Stage	Canned	Reheated	Flaked	Processed	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Reheated	Flaked	Raw	0.7056	Tukey-Kramer	1.0000
Process*Stage	Canned	Reheated	Flaked	Reheated	0.0107	Tukey-Kramer	0.1811
Process*Stage	Canned	Reheated	Simmered	Processed	0.0003	Tukey-Kramer	0.0068
Process*Stage	Canned	Reheated	Simmered	Raw	0.0031	Tukey-Kramer	0.0656
Process*Stage	Canned	Reheated	Simmered	Reheated	0.0006	Tukey-Kramer	0.0140

Process*Stage	Flaked	Processed	Flaked	Raw	<.0001	Tukey-Kramer	0.0011
Process*Stage	Flaked	Processed	Flaked	Reheated	0.0002	Tukey-Kramer	0.0041
Process*Stage	Flaked	Processed	Simmered	Processed	<.0001	Tukey-Kramer	<.0001
Process*Stage	Flaked	Processed	Simmered	Raw	<.0001	Tukey-Kramer	<.0001
Process*Stage	Flaked	Processed	Simmered	Reheated	<.0001	Tukey-Kramer	<.0001
Process*Stage	Flaked	Raw	Flaked	Reheated	0.0438	Tukey-Kramer	0.4862
Process*Stage	Flaked	Raw	Simmered	Processed	0.0315	Tukey-Kramer	0.3952
Process*Stage	Flaked	Raw	Simmered	Raw	0.0628	Tukey-Kramer	0.5953
Process*Stage	Flaked	Raw	Simmered	Reheated	0.0468	Tukey-Kramer	0.5053
Process*Stage	Flaked	Reheated	Simmered	Processed	<.0001	Tukey-Kramer	<.0001
Process*Stage	Flaked	Reheated	Simmered	Raw	<.0001	Tukey-Kramer	0.0002
Process*Stage	Flaked	Reheated	Simmered	Reheated	<.0001	Tukey-Kramer	<.0001
Process*Stage	Simmered	Processed	Simmered	Raw	0.8561	Tukey-Kramer	1.0000
Process*Stage	Simmered	Processed	Simmered	Reheated	0.7730	Tukey-Kramer	1.0000
Process*Stage	Simmered	Raw	Simmered	Reheated	0.9567	Tukey-Kramer	1.0000

The Mixed Procedure

Model Information

Data Set	WORK.IN
Dependent Variable	Thiamin
Covariance Structure	Variance Components
Estimation Method	REML
Residual Variance Method	Profile
Fixed Effects SE Method	Model-Based
Degrees of Freedom Method	Containment

Class Level Information

Class	Levels	Values
large_bag	4	3 4 6 7
small_bag	6	5 6 11 15 16 20
Stage	3	Processed Raw Reheated

Dimensions

Covariance Parameters	2
Columns in X	4
Columns in Z	10
Subjects	1
Max Obs Per Subject	20

Number of Observations

Number of Observations Read	20
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Number of Observations Used	20
Number of Observations Not Used	0

Iteration History

Iteration	Evaluations	-2 Res Log Like	Criterion
0	1	-39.20810516	
1	1	-45.90136983	0.00000000

Convergence criteria met.

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The Mixed Procedure

Covariance Parameter  
Estimates

Cov Parm	Estimate
small_bag(Stage)	0.003486
Residual	0.001339

Fit Statistics

-2 Res Log Likelihood	-45.9
AIC (smaller is better)	-41.9
AICC (smaller is better)	-41.0
BIC (smaller is better)	-41.3

Type 3 Tests of Fixed Effects

Effect	Num DF	Den DF	F Value	Pr > F
Stage	2	7	4.47	0.0560

Least Squares Means

Effect	Stage	Estimate	Standard Error	DF	t Value	Pr >  t
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Stage	Processed	0.3520	0.03223	7	10.92	<.0001
Stage	Raw	0.5187	0.04558	7	11.38	<.0001
Stage	Reheated	0.4146	0.03223	7	12.86	<.0001

Differences of Least Squares Means

Effect	Stage	Stage	Estimate	Standard Error	DF	t Value	Pr >  t	Adjustment	Adj P
Stage	Processed	Raw	-0.1667	0.05582	7	-2.99	0.0203	Tukey-Kramer	0.0473
Stage	Processed	Reheated	-0.06265	0.04558	7	-1.37	0.2117	Tukey-Kramer	0.4031
Stage	Raw	Reheated	0.1041	0.05582	7	1.86	0.1046	Tukey-Kramer	0.2186

Obs	Process	Stage	_Process	_Stage	Estimate	StdErr	AdjP
1	Canned		Flaked		0.1176	0.02339	0.0014
2	Canned		Simmered		-0.08565	0.02135	0.0063
3	Flaked		Simmered		-0.2032	0.02135	<.0001
4		Processed		Raw	-0.1127	0.02434	0.0032
5		Processed		Reheated	-0.04499	0.01654	0.0559
6		Raw		Reheated	0.06774	0.02434	0.0508
7	Canned	Processed	Canned	Raw	-0.1235	0.04529	0.1837
8	Canned	Processed	Canned	Reheated	-0.01784	0.02865	0.9993
9	Canned	Processed	Flaked	Processed	0.1860	0.02865	<.0001
10	Canned	Processed	Flaked	Raw	-0.03513	0.04529	0.9967
11	Canned	Processed	Flaked	Reheated	0.06049	0.02865	0.4858
12	Canned	Processed	Simmered	Processed	-0.1377	0.02865	0.0013
13	Canned	Processed	Simmered	Raw	-0.1313	0.03508	0.0202
14	Canned	Processed	Simmered	Reheated	-0.1293	0.02865	0.0029
15	Canned	Raw	Canned	Reheated	0.1057	0.04529	0.3573
16	Canned	Raw	Flaked	Processed	0.3095	0.04529	<.0001
17	Canned	Raw	Flaked	Raw	0.08838	0.05729	0.8260
18	Canned	Raw	Flaked	Reheated	0.1840	0.04529	0.0092
19	Canned	Raw	Simmered	Processed	-0.01418	0.04529	1.0000
20	Canned	Raw	Simmered	Raw	-0.00776	0.04962	1.0000
21	Canned	Raw	Simmered	Reheated	-0.00584	0.04529	1.0000
22	Canned	Reheated	Flaked	Processed	0.2038	0.02865	<.0001
23	Canned	Reheated	Flaked	Raw	-0.01728	0.04529	1.0000
24	Canned	Reheated	Flaked	Reheated	0.07833	0.02865	0.1811
25	Canned	Reheated	Simmered	Processed	-0.1198	0.02865	0.0068
26	Canned	Reheated	Simmered	Raw	-0.1134	0.03508	0.0656
27	Canned	Reheated	Simmered	Reheated	-0.1115	0.02865	0.0140
28	Flaked	Processed	Flaked	Raw	-0.2211	0.04529	0.0011
29	Flaked	Processed	Flaked	Reheated	-0.1255	0.02865	0.0041
30	Flaked	Processed	Simmered	Processed	-0.3237	0.02865	<.0001
31	Flaked	Processed	Simmered	Raw	-0.3172	0.03508	<.0001
32	Flaked	Processed	Simmered	Reheated	-0.3153	0.02865	<.0001



33	Flaked	Raw	Flaked	Reheated	0.09562	0.04529	0.4862
34	Flaked	Raw	Simmered	Processed	-0.1026	0.04529	0.3952
35	Flaked	Raw	Simmered	Raw	-0.09614	0.04962	0.5953
36	Flaked	Raw	Simmered	Reheated	-0.09422	0.04529	0.5053
37	Flaked	Reheated	Simmered	Processed	-0.1982	0.02865	<.0001
38	Flaked	Reheated	Simmered	Raw	-0.1918	0.03508	0.0002
39	Flaked	Reheated	Simmered	Reheated	-0.1898	0.02865	<.0001
40	Simmered	Processed	Simmered	Raw	0.006420	0.03508	1.0000
41	Simmered	Processed	Simmered	Reheated	0.008342	0.02865	1.0000
42	Simmered	Raw	Simmered	Reheated	0.001923	0.03508	1.0000

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The Mixed Procedure

Model Information

Data Set	WORK.IN
Dependent Variable	Riboflavin

Covariance Structure	Variance Components
Estimation Method	REML
Residual Variance Method	Profile
Fixed Effects SE Method	Model-Based
Degrees of Freedom Method	Containment

#### Class Level Information

Class	Levels	Values
large_bag	5	1 2 3 5 6
small_bag	16	1 2 3 4 5 7 8 9 10 12 13 14 15 17 18 19
Process	3	Canned Flaked Simmered
Stage	3	Processed Raw Reheated

#### Dimensions

Covariance Parameters	3
Columns in X	16
Columns in Z	44
Subjects	1
Max Obs Per Subject	56

#### Number of Observations

Number of Observations Read	56
Number of Observations Used	56
Number of Observations Not Used	0

#### Iteration History

Iteration	Evaluations	-2 Res Log Like	Criterion
0	1	-221.36842692	
1	1	-221.36842692	0.00000000

Convergence criteria met.

The Mixed Procedure

Covariance Parameter Estimates

Cov Parm	Estimate
small_bag*Process	0
small_bag*Stage	0
Residual	0.000381

Fit Statistics

-2 Res Log Likelihood	-221.4
AIC (smaller is better)	-219.4
AICC (smaller is better)	-219.3
BIC (smaller is better)	-218.6

Type 3 Tests of Fixed Effects

Effect	Num DF	Den DF	F Value	Pr > F
Process	2	10	9.14	0.0055
Stage	2	9	39.57	<.0001
Process*Stage	4	28	32.59	<.0001

Least Squares Means

Effect	Process	Stage	Estimate	Standard Error	DF	t Value	Pr >  t
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Process	Canned		0.2616	0.005636	10	46.42	<.0001
Process	Flaked		0.2617	0.005636	10	46.44	<.0001
Process	Simmered		0.2356	0.004602	10	51.21	<.0001
Stage		Processed	0.2416	0.003985	9	60.62	<.0001
Stage		Raw	0.2314	0.007276	9	31.81	<.0001
Stage		Reheated	0.2860	0.003985	9	71.75	<.0001
Process*Stage	Canned	Processed	0.2761	0.006903	28	39.99	<.0001
Process*Stage	Canned	Raw	0.2395	0.01381	28	17.35	<.0001
Process*Stage	Canned	Reheated	0.2693	0.006903	28	39.02	<.0001
Process*Stage	Flaked	Processed	0.2206	0.006903	28	31.96	<.0001
Process*Stage	Flaked	Raw	0.2155	0.01381	28	15.61	<.0001
Process*Stage	Flaked	Reheated	0.3491	0.006903	28	50.57	<.0001
Process*Stage	Simmered	Processed	0.2281	0.006903	28	33.05	<.0001
Process*Stage	Simmered	Raw	0.2394	0.009762	28	24.52	<.0001
Process*Stage	Simmered	Reheated	0.2395	0.006903	28	34.70	<.0001

The Mixed Procedure

Differences of Least Squares Means

Effect	Process	Stage	Process	Stage	Estimate	Standard Error	DF	t Value
Process	Canned		Flaked		-0.00011	0.007970	10	-0.01
Process	Canned		Simmered		0.02597	0.007276	10	3.57
Process	Flaked		Simmered		0.02608	0.007276	10	3.58
Stage		Processed		Raw	0.01014	0.008296	9	1.22
Stage		Processed		Reheated	-0.04437	0.005636	9	-7.87
Stage		Raw		Reheated	-0.05451	0.008296	9	-6.57
Process*Stage	Canned	Processed	Canned	Raw	0.03658	0.01543	28	2.37
Process*Stage	Canned	Processed	Canned	Reheated	0.006750	0.009762	28	0.69
Process*Stage	Canned	Processed	Flaked	Processed	0.05546	0.009762	28	5.68
Process*Stage	Canned	Processed	Flaked	Raw	0.06055	0.01543	28	3.92
Process*Stage	Canned	Processed	Flaked	Reheated	-0.07301	0.009762	28	-7.48
Process*Stage	Canned	Processed	Simmered	Processed	0.04796	0.009762	28	4.91
Process*Stage	Canned	Processed	Simmered	Raw	0.03671	0.01196	28	3.07
Process*Stage	Canned	Processed	Simmered	Reheated	0.03657	0.009762	28	3.75
Process*Stage	Canned	Raw	Canned	Reheated	-0.02983	0.01543	28	-1.93
Process*Stage	Canned	Raw	Flaked	Processed	0.01888	0.01543	28	1.22
Process*Stage	Canned	Raw	Flaked	Raw	0.02397	0.01952	28	1.23
Process*Stage	Canned	Raw	Flaked	Reheated	-0.1096	0.01543	28	-7.10
Process*Stage	Canned	Raw	Simmered	Processed	0.01138	0.01543	28	0.74
Process*Stage	Canned	Raw	Simmered	Raw	0.000128	0.01691	28	0.01
Process*Stage	Canned	Raw	Simmered	Reheated	-6.63E-6	0.01543	28	-0.00
Process*Stage	Canned	Reheated	Flaked	Processed	0.04870	0.009762	28	4.99
Process*Stage	Canned	Reheated	Flaked	Raw	0.05380	0.01543	28	3.49
Process*Stage	Canned	Reheated	Flaked	Reheated	-0.07976	0.009762	28	-8.17
Process*Stage	Canned	Reheated	Simmered	Processed	0.04121	0.009762	28	4.22
Process*Stage	Canned	Reheated	Simmered	Raw	0.02996	0.01196	28	2.51
Process*Stage	Canned	Reheated	Simmered	Reheated	0.02982	0.009762	28	3.06

Process*Stage	Flaked	Processed	Flaked	Raw	0.005093	0.01543	28	0.33
Process*Stage	Flaked	Processed	Flaked	Reheated	-0.1285	0.009762	28	-13.16
Process*Stage	Flaked	Processed	Simmered	Processed	-0.00749	0.009762	28	-0.77
Process*Stage	Flaked	Processed	Simmered	Raw	-0.01875	0.01196	28	-1.57
Process*Stage	Flaked	Processed	Simmered	Reheated	-0.01888	0.009762	28	-1.93
Process*Stage	Flaked	Raw	Flaked	Reheated	-0.1336	0.01543	28	-8.65
Process*Stage	Flaked	Raw	Simmered	Processed	-0.01259	0.01543	28	-0.82
Process*Stage	Flaked	Raw	Simmered	Raw	-0.02384	0.01691	28	-1.41
Process*Stage	Flaked	Raw	Simmered	Reheated	-0.02398	0.01543	28	-1.55
Process*Stage	Flaked	Reheated	Simmered	Processed	0.1210	0.009762	28	12.39
Process*Stage	Flaked	Reheated	Simmered	Raw	0.1097	0.01196	28	9.18
Process*Stage	Flaked	Reheated	Simmered	Reheated	0.1096	0.009762	28	11.23
Process*Stage	Simmered	Processed	Simmered	Raw	-0.01125	0.01196	28	-0.94
Process*Stage	Simmered	Processed	Simmered	Reheated	-0.01139	0.009762	28	-1.17
Process*Stage	Simmered	Raw	Simmered	Reheated	-0.00013	0.01196	28	-0.01

The Mixed Procedure

Differences of Least Squares Means

Effect	Process	Stage	Process	Stage	Pr >  t	Adjustment	Adj P
Process	Canned		Flaked		0.9891	Tukey-Kramer	0.9999
Process	Canned		Simmered		0.0051	Tukey-Kramer	0.0129
Process	Flaked		Simmered		0.0050	Tukey-Kramer	0.0126
Stage		Processed		Raw	0.2526	Tukey-Kramer	0.4704
Stage		Processed		Reheated	<.0001	Tukey-Kramer	<.0001
Stage		Raw		Reheated	0.0001	Tukey-Kramer	0.0003
Process*Stage	Canned	Processed	Canned	Raw	0.0249	Tukey-Kramer	0.3377
Process*Stage	Canned	Processed	Canned	Reheated	0.4949	Tukey-Kramer	0.9985
Process*Stage	Canned	Processed	Flaked	Processed	<.0001	Tukey-Kramer	0.0001
Process*Stage	Canned	Processed	Flaked	Raw	0.0005	Tukey-Kramer	0.0130
Process*Stage	Canned	Processed	Flaked	Reheated	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Processed	Simmered	Processed	<.0001	Tukey-Kramer	0.0010
Process*Stage	Canned	Processed	Simmered	Raw	0.0047	Tukey-Kramer	0.0930
Process*Stage	Canned	Processed	Simmered	Reheated	0.0008	Tukey-Kramer	0.0200
Process*Stage	Canned	Raw	Canned	Reheated	0.0635	Tukey-Kramer	0.5986
Process*Stage	Canned	Raw	Flaked	Processed	0.2315	Tukey-Kramer	0.9443
Process*Stage	Canned	Raw	Flaked	Raw	0.2298	Tukey-Kramer	0.9432
Process*Stage	Canned	Raw	Flaked	Reheated	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Raw	Simmered	Processed	0.4670	Tukey-Kramer	0.9977
Process*Stage	Canned	Raw	Simmered	Raw	0.9940	Tukey-Kramer	1.0000
Process*Stage	Canned	Raw	Simmered	Reheated	0.9997	Tukey-Kramer	1.0000
Process*Stage	Canned	Reheated	Flaked	Processed	<.0001	Tukey-Kramer	0.0008
Process*Stage	Canned	Reheated	Flaked	Raw	0.0016	Tukey-Kramer	0.0371
Process*Stage	Canned	Reheated	Flaked	Reheated	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Reheated	Simmered	Processed	0.0002	Tukey-Kramer	0.0061
Process*Stage	Canned	Reheated	Simmered	Raw	0.0183	Tukey-Kramer	0.2716
Process*Stage	Canned	Reheated	Simmered	Reheated	0.0049	Tukey-Kramer	0.0960



Process*Stage	Flaked	Processed	Flaked	Raw	0.7439	Tukey-Kramer	1.0000
Process*Stage	Flaked	Processed	Flaked	Reheated	<.0001	Tukey-Kramer	<.0001
Process*Stage	Flaked	Processed	Simmered	Processed	0.4491	Tukey-Kramer	0.9969
Process*Stage	Flaked	Processed	Simmered	Raw	0.1281	Tukey-Kramer	0.8133
Process*Stage	Flaked	Processed	Simmered	Reheated	0.0632	Tukey-Kramer	0.5975
Process*Stage	Flaked	Raw	Flaked	Reheated	<.0001	Tukey-Kramer	<.0001
Process*Stage	Flaked	Raw	Simmered	Processed	0.4216	Tukey-Kramer	0.9954
Process*Stage	Flaked	Raw	Simmered	Raw	0.1695	Tukey-Kramer	0.8846
Process*Stage	Flaked	Raw	Simmered	Reheated	0.1316	Tukey-Kramer	0.8207
Process*Stage	Flaked	Reheated	Simmered	Processed	<.0001	Tukey-Kramer	<.0001
Process*Stage	Flaked	Reheated	Simmered	Raw	<.0001	Tukey-Kramer	<.0001
Process*Stage	Flaked	Reheated	Simmered	Reheated	<.0001	Tukey-Kramer	<.0001
Process*Stage	Simmered	Processed	Simmered	Raw	0.3546	Tukey-Kramer	0.9882
Process*Stage	Simmered	Processed	Simmered	Reheated	0.2532	Tukey-Kramer	0.9571
Process*Stage	Simmered	Raw	Simmered	Reheated	0.9911	Tukey-Kramer	1.0000

The Mixed Procedure

Model Information

Data Set	WORK.IN
Dependent Variable	Riboflavin
Covariance Structure	Variance Components
Estimation Method	REML
Residual Variance Method	Profile
Fixed Effects SE Method	Model-Based
Degrees of Freedom Method	Containment

Class Level Information

Class	Levels	Values
large_bag	4	3 4 6 7
small_bag	6	5 6 11 15 16 20
Stage	3	Processed Raw Reheated

Dimensions

Covariance Parameters	2
Columns in X	4
Columns in Z	10
Subjects	1
Max Obs Per Subject	20

Number of Observations

Number of Observations Read	20
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Number of Observations Used	20
Number of Observations Not Used	0

Iteration History

Iteration	Evaluations	-2 Res Log Like	Criterion
0	1	-61.99850989	
1	1	-67.30613103	0.00000000

Convergence criteria met.

The Mixed Procedure

Covariance Parameter Estimates

Cov Parm	Estimate
small_bag(Stage)	0.000834
Residual	0.000415

Fit Statistics

-2 Res Log Likelihood	-67.3
AIC (smaller is better)	-63.3
AICC (smaller is better)	-62.4
BIC (smaller is better)	-62.7

Type 3 Tests of Fixed Effects

Effect	Num DF	Den DF	F Value	Pr > F
Stage	2	7	3.48	0.0892

Least Squares Means

Effect	Stage	Estimate	Standard Error	DF	t Value	Pr >  t
Stage	Processed	0.2164	0.01613	7	13.41	<.0001
Stage	Raw	0.2275	0.02282	7	9.97	<.0001

Stage	Reheated	0.2744	0.01613	7	17.01	<.0001
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Differences of Least Squares Means

Effect	Stage	Stage	Estimate	Standard Error	DF	t Value	Pr >  t	Adjustment	Adj P
Stage	Processed	Raw	-0.01113	0.02795	7	-0.40	0.7023	Tukey-Kramer	0.9173
Stage	Processed	Reheated	-0.05804	0.02282	7	-2.54	0.0384	Tukey-Kramer	0.0867
Stage	Raw	Reheated	-0.04692	0.02795	7	-1.68	0.1371	Tukey-Kramer	0.2779

Obs	Process	Stage	_Process	_Stage	Estimate	StdErr	AdjP
1	Canned		Flaked		-0.00011	0.007970	0.9999
2	Canned		Simmered		0.02597	0.007276	0.0129
3	Flaked		Simmered		0.02608	0.007276	0.0126
4		Processed		Raw	0.01014	0.008296	0.4704
5		Processed		Reheated	-0.04437	0.005636	<.0001
6		Raw		Reheated	-0.05451	0.008296	0.0003
7	Canned	Processed	Canned	Raw	0.03658	0.01543	0.3377
8	Canned	Processed	Canned	Reheated	0.006750	0.009762	0.9985
9	Canned	Processed	Flaked	Processed	0.05546	0.009762	0.0001
10	Canned	Processed	Flaked	Raw	0.06055	0.01543	0.0130
11	Canned	Processed	Flaked	Reheated	-0.07301	0.009762	<.0001
12	Canned	Processed	Simmered	Processed	0.04796	0.009762	0.0010
13	Canned	Processed	Simmered	Raw	0.03671	0.01196	0.0930
14	Canned	Processed	Simmered	Reheated	0.03657	0.009762	0.0200
15	Canned	Raw	Canned	Reheated	-0.02983	0.01543	0.5986
16	Canned	Raw	Flaked	Processed	0.01888	0.01543	0.9443
17	Canned	Raw	Flaked	Raw	0.02397	0.01952	0.9432
18	Canned	Raw	Flaked	Reheated	-0.1096	0.01543	<.0001
19	Canned	Raw	Simmered	Processed	0.01138	0.01543	0.9977
20	Canned	Raw	Simmered	Raw	0.000128	0.01691	1.0000
21	Canned	Raw	Simmered	Reheated	-6.63E-6	0.01543	1.0000
22	Canned	Reheated	Flaked	Processed	0.04870	0.009762	0.0008
23	Canned	Reheated	Flaked	Raw	0.05380	0.01543	0.0371
24	Canned	Reheated	Flaked	Reheated	-0.07976	0.009762	<.0001
25	Canned	Reheated	Simmered	Processed	0.04121	0.009762	0.0061
26	Canned	Reheated	Simmered	Raw	0.02996	0.01196	0.2716
27	Canned	Reheated	Simmered	Reheated	0.02982	0.009762	0.0960
28	Flaked	Processed	Flaked	Raw	0.005093	0.01543	1.0000
29	Flaked	Processed	Flaked	Reheated	-0.1285	0.009762	<.0001
30	Flaked	Processed	Simmered	Processed	-0.00749	0.009762	0.9969
31	Flaked	Processed	Simmered	Raw	-0.01875	0.01196	0.8133
32	Flaked	Processed	Simmered	Reheated	-0.01888	0.009762	0.5975

33	Flaked	Raw	Flaked	Reheated	-0.1336	0.01543	<.0001
34	Flaked	Raw	Simmered	Processed	-0.01259	0.01543	0.9954
35	Flaked	Raw	Simmered	Raw	-0.02384	0.01691	0.8846
36	Flaked	Raw	Simmered	Reheated	-0.02398	0.01543	0.8207
37	Flaked	Reheated	Simmered	Processed	0.1210	0.009762	<.0001
38	Flaked	Reheated	Simmered	Raw	0.1097	0.01196	<.0001
39	Flaked	Reheated	Simmered	Reheated	0.1096	0.009762	<.0001
40	Simmered	Processed	Simmered	Raw	-0.01125	0.01196	0.9882
41	Simmered	Processed	Simmered	Reheated	-0.01139	0.009762	0.9571
42	Simmered	Raw	Simmered	Reheated	-0.00013	0.01196	1.0000

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The Mixed Procedure

Model Information

Data Set

WORK.IN

Dependent Variable	Folate
Covariance Structure	Variance Components
Estimation Method	REML
Residual Variance Method	Profile
Fixed Effects SE Method	Model-Based
Degrees of Freedom Method	Containment

#### Class Level Information

Class	Levels	Values
large_bag	5	1 2 3 5 6
small_bag	16	1 2 3 4 5 7 8 9 10 12 13 14 15 17 18 19
Process	3	Canned Flaked Simmered
Stage	3	Processed Raw Reheated

#### Dimensions

Covariance Parameters	3
Columns in X	16
Columns in Z	44
Subjects	1
Max Obs Per Subject	56

#### Number of Observations

Number of Observations Read	56
Number of Observations Used	56
Number of Observations Not Used	0

#### Iteration History



Iteration	Evaluations	-2 Res Log Like	Criterion
0	1	85.27063495	
1	3	30.09081808	0.00835292
2	1	29.82031472	0.00063600
3	1	29.80136286	0.00000472
4	1	29.80122861	0.00000000

The Mixed Procedure

Convergence criteria met.

Covariance Parameter Estimates

Cov Parm	Estimate
small_bag*Process	0
small_bag*Stage	0.2965
Residual	0.02000

Fit Statistics

-2 Res Log Likelihood	29.8
AIC (smaller is better)	33.8
AICC (smaller is better)	34.1
BIC (smaller is better)	35.3

Type 3 Tests of Fixed Effects

Effect	Num DF	Den DF	F Value	Pr > F
Process	2	10	2.99	0.0957
Stage	2	9	4.39	0.0468
Process*Stage	4	28	3.29	0.0248

Least Squares Means

Effect	Process	Stage	Estimate	Standard Error	DF	t Value	Pr >  t
Process	Canned		2.7589	0.2260	10	12.21	<.0001
Process	Flaked		2.2888	0.2260	10	10.13	<.0001
Process	Simmered		2.0464	0.1845	10	11.09	<.0001
Stage		Processed	2.1852	0.1598	9	13.67	<.0001
Stage		Raw	2.9471	0.2918	9	10.10	<.0001
Stage		Reheated	1.9618	0.1598	9	12.27	<.0001
Process*Stage	Canned	Processed	2.9939	0.2768	28	10.82	<.0001
Process*Stage	Canned	Raw	3.0922	0.5536	28	5.59	<.0001
Process*Stage	Canned	Reheated	2.1906	0.2768	28	7.91	<.0001
Process*Stage	Flaked	Processed	1.3437	0.2768	28	4.85	<.0001
Process*Stage	Flaked	Raw	3.6196	0.5536	28	6.54	<.0001
Process*Stage	Flaked	Reheated	1.9031	0.2768	28	6.87	<.0001
Process*Stage	Simmered	Processed	2.2181	0.2768	28	8.01	<.0001
Process*Stage	Simmered	Raw	2.1295	0.3915	28	5.44	<.0001
Process*Stage	Simmered	Reheated	1.7916	0.2768	28	6.47	<.0001

The Mixed Procedure

Differences of Least Squares Means

Effect	Process	Stage	Process	Stage	Estimate	Standard Error	DF	t Value
Process	Canned		Flaked		0.4702	0.3196	10	1.47
Process	Canned		Simmered		0.7125	0.2918	10	2.44
Process	Flaked		Simmered		0.2424	0.2918	10	0.83
Stage		Processed		Raw	-0.7619	0.3327	9	-2.29
Stage		Processed		Reheated	0.2235	0.2260	9	0.99
Stage		Raw		Reheated	0.9853	0.3327	9	2.96
Process*Stage	Canned	Processed	Canned	Raw	-0.09829	0.6190	28	-0.16
Process*Stage	Canned	Processed	Canned	Reheated	0.8034	0.3915	28	2.05
Process*Stage	Canned	Processed	Flaked	Processed	1.6503	0.3915	28	4.22
Process*Stage	Canned	Processed	Flaked	Raw	-0.6256	0.6190	28	-1.01
Process*Stage	Canned	Processed	Flaked	Reheated	1.0909	0.3915	28	2.79
Process*Stage	Canned	Processed	Simmered	Processed	0.7759	0.3915	28	1.98
Process*Stage	Canned	Processed	Simmered	Raw	0.8644	0.4795	28	1.80
Process*Stage	Canned	Processed	Simmered	Reheated	1.2023	0.3915	28	3.07
Process*Stage	Canned	Raw	Canned	Reheated	0.9016	0.6190	28	1.46
Process*Stage	Canned	Raw	Flaked	Processed	1.7486	0.6190	28	2.82
Process*Stage	Canned	Raw	Flaked	Raw	-0.5273	0.7830	28	-0.67
Process*Stage	Canned	Raw	Flaked	Reheated	1.1892	0.6190	28	1.92
Process*Stage	Canned	Raw	Simmered	Processed	0.8741	0.6190	28	1.41
Process*Stage	Canned	Raw	Simmered	Raw	0.9627	0.6781	28	1.42
Process*Stage	Canned	Raw	Simmered	Reheated	1.3006	0.6190	28	2.10
Process*Stage	Canned	Reheated	Flaked	Processed	0.8469	0.3915	28	2.16
Process*Stage	Canned	Reheated	Flaked	Raw	-1.4290	0.6190	28	-2.31
Process*Stage	Canned	Reheated	Flaked	Reheated	0.2875	0.3915	28	0.73
Process*Stage	Canned	Reheated	Simmered	Processed	-0.02750	0.3915	28	-0.07
Process*Stage	Canned	Reheated	Simmered	Raw	0.06109	0.4795	28	0.13
Process*Stage	Canned	Reheated	Simmered	Reheated	0.3989	0.3915	28	1.02

Process*Stage	Flaked	Processed	Flaked	Raw	-2.2759	0.6190	28	-3.68
Process*Stage	Flaked	Processed	Flaked	Reheated	-0.5594	0.3915	28	-1.43
Process*Stage	Flaked	Processed	Simmered	Processed	-0.8744	0.3915	28	-2.23
Process*Stage	Flaked	Processed	Simmered	Raw	-0.7859	0.4795	28	-1.64
Process*Stage	Flaked	Processed	Simmered	Reheated	-0.4480	0.3915	28	-1.14
Process*Stage	Flaked	Raw	Flaked	Reheated	1.7165	0.6190	28	2.77
Process*Stage	Flaked	Raw	Simmered	Processed	1.4015	0.6190	28	2.26
Process*Stage	Flaked	Raw	Simmered	Raw	1.4901	0.6781	28	2.20
Process*Stage	Flaked	Raw	Simmered	Reheated	1.8279	0.6190	28	2.95
Process*Stage	Flaked	Reheated	Simmered	Processed	-0.3150	0.3915	28	-0.80
Process*Stage	Flaked	Reheated	Simmered	Raw	-0.2264	0.4795	28	-0.47
Process*Stage	Flaked	Reheated	Simmered	Reheated	0.1114	0.3915	28	0.28
Process*Stage	Simmered	Processed	Simmered	Raw	0.08859	0.4795	28	0.18
Process*Stage	Simmered	Processed	Simmered	Reheated	0.4264	0.3915	28	1.09
Process*Stage	Simmered	Raw	Simmered	Reheated	0.3379	0.4795	28	0.70

The Mixed Procedure

Differences of Least Squares Means

Effect	Process	Stage	Process	Stage	Pr >  t	Adjustment	Adj P
Process	Canned		Flaked		0.1721	Tukey-Kramer	0.3445
Process	Canned		Simmered		0.0347	Tukey-Kramer	0.0813
Process	Flaked		Simmered		0.4256	Tukey-Kramer	0.6935
Stage		Processed		Raw	0.0478	Tukey-Kramer	0.1087
Stage		Processed		Reheated	0.3487	Tukey-Kramer	0.6017
Stage		Raw		Reheated	0.0159	Tukey-Kramer	0.0384
Process*Stage	Canned	Processed	Canned	Raw	0.8750	Tukey-Kramer	1.0000
Process*Stage	Canned	Processed	Canned	Reheated	0.0496	Tukey-Kramer	0.5229
Process*Stage	Canned	Processed	Flaked	Processed	0.0002	Tukey-Kramer	0.0062
Process*Stage	Canned	Processed	Flaked	Raw	0.3208	Tukey-Kramer	0.9816
Process*Stage	Canned	Processed	Flaked	Reheated	0.0095	Tukey-Kramer	0.1642
Process*Stage	Canned	Processed	Simmered	Processed	0.0574	Tukey-Kramer	0.5673
Process*Stage	Canned	Processed	Simmered	Raw	0.0822	Tukey-Kramer	0.6800
Process*Stage	Canned	Processed	Simmered	Reheated	0.0047	Tukey-Kramer	0.0928
Process*Stage	Canned	Raw	Canned	Reheated	0.1563	Tukey-Kramer	0.8655
Process*Stage	Canned	Raw	Flaked	Processed	0.0086	Tukey-Kramer	0.1526
Process*Stage	Canned	Raw	Flaked	Raw	0.5061	Tukey-Kramer	0.9988
Process*Stage	Canned	Raw	Flaked	Reheated	0.0649	Tukey-Kramer	0.6059
Process*Stage	Canned	Raw	Simmered	Processed	0.1689	Tukey-Kramer	0.8837
Process*Stage	Canned	Raw	Simmered	Raw	0.1667	Tukey-Kramer	0.8807
Process*Stage	Canned	Raw	Simmered	Reheated	0.0448	Tukey-Kramer	0.4923
Process*Stage	Canned	Reheated	Flaked	Processed	0.0392	Tukey-Kramer	0.4543
Process*Stage	Canned	Reheated	Flaked	Raw	0.0286	Tukey-Kramer	0.3706
Process*Stage	Canned	Reheated	Flaked	Reheated	0.4688	Tukey-Kramer	0.9977
Process*Stage	Canned	Reheated	Simmered	Processed	0.9445	Tukey-Kramer	1.0000
Process*Stage	Canned	Reheated	Simmered	Raw	0.8995	Tukey-Kramer	1.0000
Process*Stage	Canned	Reheated	Simmered	Reheated	0.3169	Tukey-Kramer	0.9806

Process*Stage	Flaked	Processed	Flaked	Raw	0.0010	Tukey-Kramer	0.0237
Process*Stage	Flaked	Processed	Flaked	Reheated	0.1641	Tukey-Kramer	0.8770
Process*Stage	Flaked	Processed	Simmered	Processed	0.0337	Tukey-Kramer	0.4128
Process*Stage	Flaked	Processed	Simmered	Raw	0.1124	Tukey-Kramer	0.7759
Process*Stage	Flaked	Processed	Simmered	Reheated	0.2622	Tukey-Kramer	0.9616
Process*Stage	Flaked	Raw	Flaked	Reheated	0.0098	Tukey-Kramer	0.1685
Process*Stage	Flaked	Raw	Simmered	Processed	0.0315	Tukey-Kramer	0.3953
Process*Stage	Flaked	Raw	Simmered	Raw	0.0364	Tukey-Kramer	0.4339
Process*Stage	Flaked	Raw	Simmered	Reheated	0.0063	Tukey-Kramer	0.1184
Process*Stage	Flaked	Reheated	Simmered	Processed	0.4278	Tukey-Kramer	0.9958
Process*Stage	Flaked	Reheated	Simmered	Raw	0.6404	Tukey-Kramer	0.9999
Process*Stage	Flaked	Reheated	Simmered	Reheated	0.7780	Tukey-Kramer	1.0000
Process*Stage	Simmered	Processed	Simmered	Raw	0.8547	Tukey-Kramer	1.0000
Process*Stage	Simmered	Processed	Simmered	Reheated	0.2853	Tukey-Kramer	0.9711
Process*Stage	Simmered	Raw	Simmered	Reheated	0.4868	Tukey-Kramer	0.9983

The Mixed Procedure

Model Information

Data Set	WORK.IN
Dependent Variable	Folate
Covariance Structure	Variance Components
Estimation Method	REML
Residual Variance Method	Profile
Fixed Effects SE Method	Model-Based
Degrees of Freedom Method	Containment

Class Level Information

Class	Levels	Values
large_bag	4	3 4 6 7
small_bag	6	5 6 11 15 16 20
Stage	3	Processed Raw Reheated

Dimensions

Covariance Parameters	2
Columns in X	4
Columns in Z	10
Subjects	1
Max Obs Per Subject	20

Number of Observations

Number of Observations Read	20
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Number of Observations Used	20
Number of Observations Not Used	0

Iteration History

Iteration	Evaluations	-2 Res Log Like	Criterion
0	1	46.99447592	
1	1	19.01461889	0.00000000

Convergence criteria met.

The Mixed Procedure

Covariance Parameter Estimates

Cov Parm	Estimate
small_bag(Stage)	0.7872
Residual	0.02226

Fit Statistics

-2 Res Log Likelihood	19.0
AIC (smaller is better)	23.0
AICC (smaller is better)	23.9
BIC (smaller is better)	23.6

Type 3 Tests of Fixed Effects

Effect	Num DF	Den DF	F Value	Pr > F
Stage	2	7	1.80	0.2336

Least Squares Means

Effect	Stage	Estimate	Standard Error	DF	t Value	Pr >  t
Stage	Processed	2.6331	0.4467	7	5.89	0.0006
Stage	Raw	3.3559	0.6318	7	5.31	0.0011

Stage	Reheated	1.9206	0.4467	7	4.30	0.0036
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Differences of Least Squares Means

Effect	Stage	Stage	Estimate	Standard Error	DF	t Value	Pr >  t	Adjustment	Adj P
Stage	Processed	Raw	-0.7228	0.7738	7	-0.93	0.3814	Tukey-Kramer	0.6377
Stage	Processed	Reheated	0.7125	0.6318	7	1.13	0.2966	Tukey-Kramer	0.5286
Stage	Raw	Reheated	1.4353	0.7738	7	1.85	0.1060	Tukey-Kramer	0.2212

Obs	Process	Stage	_Process	_Stage	Estimate	StdErr	AdjP
1	Canned		Flaked		0.4702	0.3196	0.3445
2	Canned		Simmered		0.7125	0.2918	0.0813
3	Flaked		Simmered		0.2424	0.2918	0.6935
4		Processed		Raw	-0.7619	0.3327	0.1087
5		Processed		Reheated	0.2235	0.2260	0.6017
6		Raw		Reheated	0.9853	0.3327	0.0384
7	Canned	Processed	Canned	Raw	-0.09829	0.6190	1.0000
8	Canned	Processed	Canned	Reheated	0.8034	0.3915	0.5229
9	Canned	Processed	Flaked	Processed	1.6503	0.3915	0.0062
10	Canned	Processed	Flaked	Raw	-0.6256	0.6190	0.9816
11	Canned	Processed	Flaked	Reheated	1.0909	0.3915	0.1642
12	Canned	Processed	Simmered	Processed	0.7759	0.3915	0.5673
13	Canned	Processed	Simmered	Raw	0.8644	0.4795	0.6800
14	Canned	Processed	Simmered	Reheated	1.2023	0.3915	0.0928
15	Canned	Raw	Canned	Reheated	0.9016	0.6190	0.8655
16	Canned	Raw	Flaked	Processed	1.7486	0.6190	0.1526
17	Canned	Raw	Flaked	Raw	-0.5273	0.7830	0.9988
18	Canned	Raw	Flaked	Reheated	1.1892	0.6190	0.6059
19	Canned	Raw	Simmered	Processed	0.8741	0.6190	0.8837
20	Canned	Raw	Simmered	Raw	0.9627	0.6781	0.8807
21	Canned	Raw	Simmered	Reheated	1.3006	0.6190	0.4923
22	Canned	Reheated	Flaked	Processed	0.8469	0.3915	0.4543
23	Canned	Reheated	Flaked	Raw	-1.4290	0.6190	0.3706
24	Canned	Reheated	Flaked	Reheated	0.2875	0.3915	0.9977
25	Canned	Reheated	Simmered	Processed	-0.02750	0.3915	1.0000
26	Canned	Reheated	Simmered	Raw	0.06109	0.4795	1.0000
27	Canned	Reheated	Simmered	Reheated	0.3989	0.3915	0.9806
28	Flaked	Processed	Flaked	Raw	-2.2759	0.6190	0.0237
29	Flaked	Processed	Flaked	Reheated	-0.5594	0.3915	0.8770
30	Flaked	Processed	Simmered	Processed	-0.8744	0.3915	0.4128
31	Flaked	Processed	Simmered	Raw	-0.7859	0.4795	0.7759
32	Flaked	Processed	Simmered	Reheated	-0.4480	0.3915	0.9616

33	Flaked	Raw	Flaked	Reheated	1.7165	0.6190	0.1685
34	Flaked	Raw	Simmered	Processed	1.4015	0.6190	0.3953
35	Flaked	Raw	Simmered	Raw	1.4901	0.6781	0.4339
36	Flaked	Raw	Simmered	Reheated	1.8279	0.6190	0.1184
37	Flaked	Reheated	Simmered	Processed	-0.3150	0.3915	0.9958
38	Flaked	Reheated	Simmered	Raw	-0.2264	0.4795	0.9999
39	Flaked	Reheated	Simmered	Reheated	0.1114	0.3915	1.0000
40	Simmered	Processed	Simmered	Raw	0.08859	0.4795	1.0000
41	Simmered	Processed	Simmered	Reheated	0.4264	0.3915	0.9711
42	Simmered	Raw	Simmered	Reheated	0.3379	0.4795	0.9983

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The Mixed Procedure

Model Information

Data Set

WORK.IN

Dependent Variable	B6
Covariance Structure	Variance Components
Estimation Method	REML
Residual Variance Method	Profile
Fixed Effects SE Method	Model-Based
Degrees of Freedom Method	Containment

#### Class Level Information

Class	Levels	Values
large_bag	5	1 2 3 5 6
small_bag	16	1 2 3 4 5 7 8 9 10 12 13 14 15 17 18 19
Process	3	Canned Flaked Simmered
Stage	3	Processed Raw Reheated

#### Dimensions

Covariance Parameters	3
Columns in X	16
Columns in Z	44
Subjects	1
Max Obs Per Subject	56

#### Number of Observations

Number of Observations Read	56
Number of Observations Used	56
Number of Observations Not Used	0

#### Iteration History

Iteration	Evaluations	-2 Res Log Like	Criterion
0	1	251.92958580	
1	3	206.01842376	0.00267751
2	1	205.83717344	0.00014824
3	1	205.82797498	0.00000056
4	1	205.82794167	0.00000000

The Mixed Procedure

Convergence criteria met.

Covariance Parameter Estimates

Cov Parm	Estimate
small_bag*Process	0
small_bag*Stage	9.9235
Residual	0.9826

Fit Statistics

-2 Res Log Likelihood	205.8
AIC (smaller is better)	209.8
AICC (smaller is better)	210.1
BIC (smaller is better)	211.4

Type 3 Tests of Fixed Effects

Effect	Num DF	Den DF	F Value	Pr > F
Process	2	10	8.07	0.0082
Stage	2	9	275.83	<.0001
Process*Stage	4	28	5.32	0.0026

Least Squares Means



Effect	Process	Stage	Estimate	Standard Error	DF	t Value	Pr >  t
Process	Canned		22.9006	1.3175	10	17.38	<.0001
Process	Flaked		30.3861	1.3175	10	23.06	<.0001
Process	Simmered		26.7512	1.0757	10	24.87	<.0001
Stage		Processed	11.1026	0.9316	9	11.92	<.0001
Stage		Raw	55.1308	1.7009	9	32.41	<.0001
Stage		Reheated	13.8045	0.9316	9	14.82	<.0001
Process*Stage	Canned	Processed	8.3315	1.6136	28	5.16	<.0001
Process*Stage	Canned	Raw	51.3732	3.2272	28	15.92	<.0001
Process*Stage	Canned	Reheated	8.9972	1.6136	28	5.58	<.0001
Process*Stage	Flaked	Processed	15.3078	1.6136	28	9.49	<.0001
Process*Stage	Flaked	Raw	53.3102	3.2272	28	16.52	<.0001
Process*Stage	Flaked	Reheated	22.5402	1.6136	28	13.97	<.0001
Process*Stage	Simmered	Processed	9.6686	1.6136	28	5.99	<.0001
Process*Stage	Simmered	Raw	60.7091	2.2820	28	26.60	<.0001
Process*Stage	Simmered	Reheated	9.8760	1.6136	28	6.12	<.0001

The Mixed Procedure

Differences of Least Squares Means

Effect	Process	Stage	Process	Stage	Estimate	Standard Error	DF	t Value
Process	Canned		Flaked		-7.4854	1.8632	10	-4.02
Process	Canned		Simmered		-3.8506	1.7009	10	-2.26
Process	Flaked		Simmered		3.6349	1.7009	10	2.14
Stage		Processed		Raw	-44.0282	1.9393	9	-22.70
Stage		Processed		Reheated	-2.7019	1.3175	9	-2.05
Stage		Raw		Reheated	41.3263	1.9393	9	21.31
Process*Stage	Canned	Processed	Canned	Raw	-43.0417	3.6081	28	-11.93
Process*Stage	Canned	Processed	Canned	Reheated	-0.6657	2.2820	28	-0.29
Process*Stage	Canned	Processed	Flaked	Processed	-6.9763	2.2820	28	-3.06
Process*Stage	Canned	Processed	Flaked	Raw	-44.9787	3.6081	28	-12.47
Process*Stage	Canned	Processed	Flaked	Reheated	-14.2087	2.2820	28	-6.23
Process*Stage	Canned	Processed	Simmered	Processed	-1.3371	2.2820	28	-0.59
Process*Stage	Canned	Processed	Simmered	Raw	-52.3776	2.7948	28	-18.74
Process*Stage	Canned	Processed	Simmered	Reheated	-1.5445	2.2820	28	-0.68
Process*Stage	Canned	Raw	Canned	Reheated	42.3760	3.6081	28	11.74
Process*Stage	Canned	Raw	Flaked	Processed	36.0654	3.6081	28	10.00
Process*Stage	Canned	Raw	Flaked	Raw	-1.9370	4.5639	28	-0.42
Process*Stage	Canned	Raw	Flaked	Reheated	28.8329	3.6081	28	7.99
Process*Stage	Canned	Raw	Simmered	Processed	41.7046	3.6081	28	11.56
Process*Stage	Canned	Raw	Simmered	Raw	-9.3359	3.9525	28	-2.36
Process*Stage	Canned	Raw	Simmered	Reheated	41.4972	3.6081	28	11.50
Process*Stage	Canned	Reheated	Flaked	Processed	-6.3106	2.2820	28	-2.77
Process*Stage	Canned	Reheated	Flaked	Raw	-44.3130	3.6081	28	-12.28
Process*Stage	Canned	Reheated	Flaked	Reheated	-13.5430	2.2820	28	-5.93
Process*Stage	Canned	Reheated	Simmered	Processed	-0.6714	2.2820	28	-0.29
Process*Stage	Canned	Reheated	Simmered	Raw	-51.7119	2.7948	28	-18.50
Process*Stage	Canned	Reheated	Simmered	Reheated	-0.8788	2.2820	28	-0.39

Process*Stage	Flaked	Processed	Flaked	Raw	-38.0024	3.6081	28	-10.53
Process*Stage	Flaked	Processed	Flaked	Reheated	-7.2324	2.2820	28	-3.17
Process*Stage	Flaked	Processed	Simmered	Processed	5.6392	2.2820	28	2.47
Process*Stage	Flaked	Processed	Simmered	Raw	-45.4013	2.7948	28	-16.24
Process*Stage	Flaked	Processed	Simmered	Reheated	5.4318	2.2820	28	2.38
Process*Stage	Flaked	Raw	Flaked	Reheated	30.7699	3.6081	28	8.53
Process*Stage	Flaked	Raw	Simmered	Processed	43.6416	3.6081	28	12.10
Process*Stage	Flaked	Raw	Simmered	Raw	-7.3989	3.9525	28	-1.87
Process*Stage	Flaked	Raw	Simmered	Reheated	43.4342	3.6081	28	12.04
Process*Stage	Flaked	Reheated	Simmered	Processed	12.8717	2.2820	28	5.64
Process*Stage	Flaked	Reheated	Simmered	Raw	-38.1688	2.7948	28	-13.66
Process*Stage	Flaked	Reheated	Simmered	Reheated	12.6643	2.2820	28	5.55
Process*Stage	Simmered	Processed	Simmered	Raw	-51.0405	2.7948	28	-18.26
Process*Stage	Simmered	Processed	Simmered	Reheated	-0.2074	2.2820	28	-0.09
Process*Stage	Simmered	Raw	Simmered	Reheated	50.8331	2.7948	28	18.19

The Mixed Procedure

Differences of Least Squares Means

Effect	Process	Stage	Process	Stage	Pr >  t	Adjustment	Adj P
Process	Canned		Flaked		0.0024	Tukey-Kramer	0.0063
Process	Canned		Simmered		0.0471	Tukey-Kramer	0.1080
Process	Flaked		Simmered		0.0583	Tukey-Kramer	0.1317
Stage		Processed		Raw	<.0001	Tukey-Kramer	<.0001
Stage		Processed		Reheated	0.0705	Tukey-Kramer	0.1557
Stage		Raw		Reheated	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Processed	Canned	Raw	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Processed	Canned	Reheated	0.7726	Tukey-Kramer	1.0000
Process*Stage	Canned	Processed	Flaked	Processed	0.0049	Tukey-Kramer	0.0956
Process*Stage	Canned	Processed	Flaked	Raw	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Processed	Flaked	Reheated	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Processed	Simmered	Processed	0.5626	Tukey-Kramer	0.9996
Process*Stage	Canned	Processed	Simmered	Raw	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Processed	Simmered	Reheated	0.5041	Tukey-Kramer	0.9987
Process*Stage	Canned	Raw	Canned	Reheated	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Raw	Flaked	Processed	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Raw	Flaked	Raw	0.6745	Tukey-Kramer	1.0000
Process*Stage	Canned	Raw	Flaked	Reheated	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Raw	Simmered	Processed	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Raw	Simmered	Raw	0.0254	Tukey-Kramer	0.3419
Process*Stage	Canned	Raw	Simmered	Reheated	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Reheated	Flaked	Processed	0.0099	Tukey-Kramer	0.1709
Process*Stage	Canned	Reheated	Flaked	Raw	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Reheated	Flaked	Reheated	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Reheated	Simmered	Processed	0.7708	Tukey-Kramer	1.0000
Process*Stage	Canned	Reheated	Simmered	Raw	<.0001	Tukey-Kramer	<.0001
Process*Stage	Canned	Reheated	Simmered	Reheated	0.7031	Tukey-Kramer	1.0000

Process*Stage	Flaked	Processed	Flaked	Raw	<.0001	Tukey-Kramer	<.0001
Process*Stage	Flaked	Processed	Flaked	Reheated	0.0037	Tukey-Kramer	0.0753
Process*Stage	Flaked	Processed	Simmered	Processed	0.0198	Tukey-Kramer	0.2875
Process*Stage	Flaked	Processed	Simmered	Raw	<.0001	Tukey-Kramer	<.0001
Process*Stage	Flaked	Processed	Simmered	Reheated	0.0243	Tukey-Kramer	0.3324
Process*Stage	Flaked	Raw	Flaked	Reheated	<.0001	Tukey-Kramer	<.0001
Process*Stage	Flaked	Raw	Simmered	Processed	<.0001	Tukey-Kramer	<.0001
Process*Stage	Flaked	Raw	Simmered	Raw	0.0717	Tukey-Kramer	0.6369
Process*Stage	Flaked	Raw	Simmered	Reheated	<.0001	Tukey-Kramer	<.0001
Process*Stage	Flaked	Reheated	Simmered	Processed	<.0001	Tukey-Kramer	0.0001
Process*Stage	Flaked	Reheated	Simmered	Raw	<.0001	Tukey-Kramer	<.0001
Process*Stage	Flaked	Reheated	Simmered	Reheated	<.0001	Tukey-Kramer	0.0002
Process*Stage	Simmered	Processed	Simmered	Raw	<.0001	Tukey-Kramer	<.0001
Process*Stage	Simmered	Processed	Simmered	Reheated	0.9282	Tukey-Kramer	1.0000
Process*Stage	Simmered	Raw	Simmered	Reheated	<.0001	Tukey-Kramer	<.0001

The Mixed Procedure

Model Information

Data Set	WORK.IN
Dependent Variable	B6
Covariance Structure	Variance Components
Estimation Method	REML
Residual Variance Method	Profile
Fixed Effects SE Method	Model-Based
Degrees of Freedom Method	Containment

Class Level Information

Class	Levels	Values
large_bag	4	3 4 6 7
small_bag	6	5 6 11 15 16 20
Stage	3	Processed Raw Reheated

Dimensions

Covariance Parameters	2
Columns in X	4
Columns in Z	10
Subjects	1
Max Obs Per Subject	20

Number of Observations

Number of Observations Read	20
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Number of Observations Used	20
Number of Observations Not Used	0

Iteration History

Iteration	Evaluations	-2 Res Log Like	Criterion
0	1	52.59171386	
1	1	44.57130737	0.00000000

Convergence criteria met.

The Mixed Procedure

Covariance Parameter Estimates

Cov Parm	Estimate
small_bag(Stage)	0.8236
Residual	0.2537

Fit Statistics

-2 Res Log Likelihood	44.6
AIC (smaller is better)	48.6
AICC (smaller is better)	49.4
BIC (smaller is better)	49.2

Type 3 Tests of Fixed Effects

Effect	Num DF	Den DF	F Value	Pr > F
Stage	2	7	1740.00	<.0001

Least Squares Means

Effect	Stage	Estimate	Standard Error	DF	t Value	Pr >  t
Stage	Processed	4.8423	0.4875	7	9.93	<.0001
Stage	Raw	52.3417	0.6894	7	75.93	<.0001



Stage	Reheated	9.5078	0.4875	7	19.51	<.0001
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Differences of Least Squares Means

Effect	Stage	Stage	Estimate	Standard Error	DF	t Value	Pr >  t	Adjustment	Adj P
Stage	Processed	Raw	-47.4994	0.8443	7	-56.26	<.0001	Tukey-Kramer	<.0001
Stage	Processed	Reheated	-4.6655	0.6894	7	-6.77	0.0003	Tukey-Kramer	0.0007
Stage	Raw	Reheated	42.8338	0.8443	7	50.73	<.0001	Tukey-Kramer	<.0001

Obs	Process	Stage	_Process	_Stage	Estimate	StdErr	AdjP
1	Canned		Flaked		-7.4854	1.8632	0.0063
2	Canned		Simmered		-3.8506	1.7009	0.1080
3	Flaked		Simmered		3.6349	1.7009	0.1317
4		Processed		Raw	-44.0282	1.9393	<.0001
5		Processed		Reheated	-2.7019	1.3175	0.1557
6		Raw		Reheated	41.3263	1.9393	<.0001
7	Canned	Processed	Canned	Raw	-43.0417	3.6081	<.0001
8	Canned	Processed	Canned	Reheated	-0.6657	2.2820	1.0000
9	Canned	Processed	Flaked	Processed	-6.9763	2.2820	0.0956
10	Canned	Processed	Flaked	Raw	-44.9787	3.6081	<.0001
11	Canned	Processed	Flaked	Reheated	-14.2087	2.2820	<.0001
12	Canned	Processed	Simmered	Processed	-1.3371	2.2820	0.9996
13	Canned	Processed	Simmered	Raw	-52.3776	2.7948	<.0001
14	Canned	Processed	Simmered	Reheated	-1.5445	2.2820	0.9987
15	Canned	Raw	Canned	Reheated	42.3760	3.6081	<.0001
16	Canned	Raw	Flaked	Processed	36.0654	3.6081	<.0001
17	Canned	Raw	Flaked	Raw	-1.9370	4.5639	1.0000
18	Canned	Raw	Flaked	Reheated	28.8329	3.6081	<.0001
19	Canned	Raw	Simmered	Processed	41.7046	3.6081	<.0001
20	Canned	Raw	Simmered	Raw	-9.3359	3.9525	0.3419
21	Canned	Raw	Simmered	Reheated	41.4972	3.6081	<.0001
22	Canned	Reheated	Flaked	Processed	-6.3106	2.2820	0.1709
23	Canned	Reheated	Flaked	Raw	-44.3130	3.6081	<.0001
24	Canned	Reheated	Flaked	Reheated	-13.5430	2.2820	<.0001
25	Canned	Reheated	Simmered	Processed	-0.6714	2.2820	1.0000
26	Canned	Reheated	Simmered	Raw	-51.7119	2.7948	<.0001
27	Canned	Reheated	Simmered	Reheated	-0.8788	2.2820	1.0000
28	Flaked	Processed	Flaked	Raw	-38.0024	3.6081	<.0001
29	Flaked	Processed	Flaked	Reheated	-7.2324	2.2820	0.0753
30	Flaked	Processed	Simmered	Processed	5.6392	2.2820	0.2875
31	Flaked	Processed	Simmered	Raw	-45.4013	2.7948	<.0001
32	Flaked	Processed	Simmered	Reheated	5.4318	2.2820	0.3324

33	Flaked	Raw	Flaked	Reheated	30.7699	3.6081	<.0001
34	Flaked	Raw	Simmered	Processed	43.6416	3.6081	<.0001
35	Flaked	Raw	Simmered	Raw	-7.3989	3.9525	0.6369
36	Flaked	Raw	Simmered	Reheated	43.4342	3.6081	<.0001
37	Flaked	Reheated	Simmered	Processed	12.8717	2.2820	0.0001
38	Flaked	Reheated	Simmered	Raw	-38.1688	2.7948	<.0001
39	Flaked	Reheated	Simmered	Reheated	12.6643	2.2820	0.0002
40	Simmered	Processed	Simmered	Raw	-51.0405	2.7948	<.0001
41	Simmered	Processed	Simmered	Reheated	-0.2074	2.2820	1.0000
42	Simmered	Raw	Simmered	Reheated	50.8331	2.7948	<.0001

## APPENDIX E: EXPANDED REFERENCES

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