ENHANCING SEED AND END-USE QUALITY OF ORGANIC DRY BEANS THROUGH GENETIC IMPROVEMENT AND INNOVATIVE PROCESSING METHODS

By

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ABSTRACT

The consumer demand for organically produced dry beans (*Phaseolus vulgaris* L.) continues to increase in the U.S. Organic farmers are faced with production and harvest challenges that impact the processing and end-use quality of dry beans. One major challenge is seed coat mechanical damage induced during harvest, which is a concern for both organic and conventional growers, but the latter has more tools to manage harvest to reduce the impact of the damage. The goal of this research is to improve organic dry bean end-use quality by exploring the genetic variability for mechanically induced seed coat damage, developing enhanced end-use product processing methods, and investigating the variations in seed quality related characteristics. Kidney and black bean market classes are the focus of this work due to their importance in organic production and challenges with end-use quality, especially seed coat splitting and canning quality. Genetic variability for seed coat splitting was evaluated in a collection of 61 kidney beans from three market classes including dark red kidney (DRK), light red kidney (LRK), and white kidney (WK) beans. The beans were harvested under two conditions to induce mild and severe seed coat damage as well as the downstream processing quality of the beans. LRK was found to be more susceptible to seed coat damage than the other two market classes with the highest average seed coat check (SCC) severity score. Genotypes less susceptible to seed coat damage than others were identified within each market class. Seed coat thickness was found not to be a good indicator for SCC severity. SCC severity scores and canning quality appearance scores were found to be negatively correlated. SCC severity score of combine threshed seeds can be used as a selection factor in variety improvement for resilience to mechanical damage. Canning and pouch processing methods were further explored as means to add value to organically produced beans. Pilot-scale pouch and canning processing protocols were developed and updated for use in dry bean breeding programs. The protocols included innovative virtual quality evaluation methods and methods to improve the processing quality of organic dry beans. Both methods were utilized with dry beans from different market classes to test the methods and identify the expected variations in processing quality. A harvest survey was conducted with participation from Michigan bean growers. Nine out of 22 black bean samples had a higher than 20% SCC percentage which is considered unacceptable in the industry standard. In general, variations in mechanical seed coat damage is a result of a series of factors including the genetics of the seeds, the seed moisture content, the environmental condition, and

the operation of harvest method. The end-use quality of organic dry beans can be enhanced using the identified genetic variability and improved processing methods.

This dissertation is dedicated to my husband, my two daughters, and my mother. Without them, I cannot be where I am.

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CHAPTER 1: GENETIC VARIABILITY FOR SEED AND END-USE QUALITY IN KIDNEY BEANS

[Preparing for publication in *Crop Sciences*]

Genetic Variability for Seed and End-Use Quality in Kidney Beans

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Abstract

Kidney beans are a type of common bean (*Phaseolus vulgaris* L.) in the Andean gene pool with strict market specifications for seed characteristics and end-use quality. Kidney beans are usually sold to US consumers as dry seeds or canned products. One major quality concern is the appearance of cracks/checks in the seed coat that result from mechanical damage that happens at harvest and post-harvest. Genetic variability and control of seed coat checks (SCC) in response to mechanical damage has not been well characterized in kidney beans. The objective of this study was to explore the genetic variability for mechanically induced seed coat damage in kidney bean and characterize through seed coat check and seed coat thickness measurements and its relationship to end-use quality including canning quality and cooking time, and to identify potential germplasm resources for genetic improvement. A collection of dark red kidney (DRK), light red kidney (LRK) and white kidney (WK) beans were grown over two years and harvested with two threshing methods to mimic mild and severe treatments of mechanical damage. Belt threshing produced mild damage (SCC severity score 1.1±0.1) and combine threshing produced severe damage (SCC severity score 1.7±0.4). Seed coat thickness was not correlated with SCC of the combine threshed beans but was negatively correlated with the belt threshed beans at r = -0.33. Seed moisture content played an important role in SCC abundance and was challenging to keep constant during the study. Higher SCC severity scores resulted in lower canning quality appearance scores (r = -0.51). Compared to DRKs and LRKs, WKs are thinner in seed coat, shorter in cooking time and are more vulnerable to mechanical seed coat damage. Genotypes with low SCC severity score while having good canning quality and short cooking time were selected from each market class and can be used for future breeding programs. The SCC severity score of combine threshed seeds can be used as a selection factor in variety improvement for resilience to mechanical damage.

Introduction

Kidney beans are a type of the common bean (*Phaseolus vulgaris* L.) which is a nutritious food legume grown and consumed worldwide. Kidney beans come in a diverse range of colors and patterns and the most common market classes of kidney beans in the US are dark red kidney (DRK), light red kidney (LRK), and white kidney (WK). Kidney beans belong to the Andean gene pool of common bean which has narrower genetic diversity and less breeding efforts being made for, as compared to the Middle American gene pool (Beebe et al., 2001). To address the narrow genetic diversity in commercially grown cultivars the Andean genepool, a panel of germplasm was collected globally, but mostly from North America and Africa to be used to catalyze breeding progress in this gene pool (Cichy et al. 2015a). The Andean diversity panel (ADP) of 396 genotypes contains 87 kidney bean genotypes including most of the major commercially grown varieties from the last 100 years (Cichy et al., 2015a). This collection serves as a potentially useful source of genetic variability for most traits of interest.

Seed and end-use quality traits are especially important in kidney beans since they are sold as dry or canned products. In these forms, seed damage is easily detected by consumers. Seed coat checks refer to physical cracks or openings in the protective layer surrounding a seed. The seed coat, also known as testa, serves to protect the developing embryo inside the seed from various stressors such as dehydration and pathogens (Souza and Marcos-Filho, 2001; Smýkal et al., 2014). Seed coat checks can be induced by various factors, including mechanical damage such as scraping or cutting of the seed coat during harvesting and handling. In the dry bean industry, seed coat checks have been considered as a major quality concern because they affect the downstream bean product quality as whole seeds or canned products. Genetic variability for dry beans to withstand mechanical damage has been documented in previous research for navy beans (Gillard and Park, 2002). In previous studies, significant differences were identified in seed coat cracking among navy bean varieties with different seed weight and shape (Dorrell and Adams, 1969) and the resistance to mechanical damage was found to be quantitatively inherited in navy bean (Park & Rupert, 2003).

Seed coat crack that may appear minor on dry seed become a greater problem once beans are thermally processed. The canning quality of dry beans is affected by many factors and seed splits is a major consideration in the appearance evaluation. Seed splitting happens during heat processing when small cracks on the seed coat become major cracks and major cracks can cause

the seed to separate. Seed size was another factor found to be related to seed splits and canning quality that beans with smaller seed size was found to be correlated with less splits after canning (Forney et al., 1990) and more damaged beans in can were found when the ratio of seed coat weight to bean volume decreased (Heil et al., 1992).

Canned beans have a long history in the US and are an important form of shelf-stable fully cooked food (Petrick, 2010). Canned beans provide consumers with nutritious, convenient, and affordable meals while protecting our planet with less energy use than refrigerated and frozen food (Cannedbeans.org, 2023). Thus, canning quality is an important end-use quality of beans, which is defined as how well beans withstand the canning process. It is evaluated by the canned products' appearance, color, and texture (Wang et al., 2022). Cooking time is another important end-use seed quality trait that affects meal selection decisions since nowadays people tend to spend less time preparing food (Plessz and Étilé, 2019). It has also been found that using fast-cooking beans to reduce retort processing time during canning can improve canning quality of the beans (Bassett et al., 2020).

The study in this paper aimed to 1) develop a screening method for mechanically induced seed coat mechanical damage, 2) explore the genetic variability of North American kidney bean germplasm in resilience to seed coat mechanical damage and 3) explore the relationship with seed coat mechanical damage and downstream end use quality factors, including canning quality and cooking time; 4) identify potential germplasm resources for future variety improvement.

Materials and Methods

Germplasm, Field trials and Mechanical Damage Induction

A group of 61 North American kidney bean germplasm consisting of three market classes originally compiled in the Andean Diversity Panel (Cichy et al., 2015a) was evaluated in this study. The germplasm included 31 light red kidney (LRK) beans, 21 dark red kidney (DRK) beans, and 9 white kidney (WK) beans. The beans were planted at the Michigan State University (MSU) Montcalm Research Farm (Entrican, MI) in the 2020 and 2021 field seasons with two replicates of each genotype and a complete randomized design. Each field replicate consisted of a two-row plot with two border rows. The 2020 field trial was planted on Jun.12 and harvested on multiple days (Sept.11, Sept.18, Sept.25 and Oct.2) according to the maturity of the plants. The 2021 field trial was planted on Jun.10 and harvested on Spet.29.

Agronomic data including days to flower (DF), days to maturity (DM) were collected through the season, and common bacterial blight (CBB) disease scores and lodging scores were given to each field plot before harvest. CBB scores range from 1-5 (where 1 = no symptom and 5 = very severe CBB). Lodging scores range from 1-5 (where 1 = 100% of plants standing erect and 5 = 100% of plants flat on the ground).

All field plots were hand-harvested in both years, where entire plants were pulled and windrowed. For each field plot in 2020, all plants were transported to MSU campus, and a few plants were threshed with a belt thresher (Almaco BT-14) while the rest of the plants were threshed with a plot combine (Hege 140). For each field plot in 2021, one field row of plants was transported to campus and threshed with the belt thresher (Almaco BT-14), and the other row was threshed with the plot combine in the field (Hege 140). Belt threshing is a gentle threshing process in which dry pods were rubbed between two rubber belts to pop open, thus minimizing mechanical damage to the seed coat. Combine threshing requires the feeding of dry plants into the metal threshing cylinder to separate pods and seeds, which usually causes more severe mechanical damage to the seed coat.

Seed coat check (SCC) measurement

The seed coat check/crack of all harvested genotypes from the two years field trials was visualized through a staining test adapted from a previous study (Gillard & Park, 2002). In the test, one hundred seeds were randomly taken from each field replicate each year and were soaked in Iodine solution (Grams Iodine Solution, LabChem, Cat#:LC149004) for 5min. Seeds were then placed into five groups (Figure 1.1 a) according to their levels of seed coat check (Group1 = no visible seed coat damage, Group 2 = one or two minor cracks in the seed coat, Group 3 = several minor cracks or one major crack in the seed coat, Group 4 = more than 1 major crack, Group 5 = split seed). Number of seeds in each group was used to calculate a weighted seed coat check severity score as: {(Number of seeds in Group1 x 1) + (Group2 x 2) + (Group3 x 3) + (Group4 x 4) + (Group5 x 5)}/ 100. The possible SCC severity score for each sample ranges from 1 (no seed coat damage in all seeds) to 5 (all seeds split). Seed coat check percentage (%) was also calculated as: Number of seeds with cracks/Total seed number, to measure the number of seeds with checked seed coat in every 100 seeds.

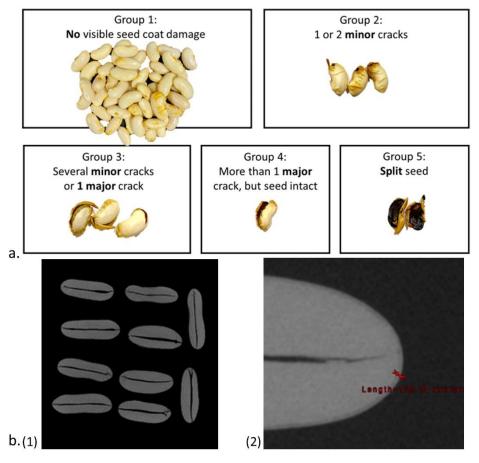


Figure 1.1 Staining test (a) to sort seeds into five groups according to their level of seed coat checks, and CT scan images (b) of bean seeds cross-section (1) and enlarged image (2) to measure the seed coat thickness.

Computerized tomography (CT) scan for seed coat thickness

All genotypes in the ADP-kidney collection from the two years of field trials were scanned with the NSI X3000 industrial CT X-ray scanner (North Star Imaging, Rogers) in Chitwood lab at MSU. From each field trial entry of the belt threshed seeds, five seeds were randomly selected for the CT-scan. Images were obtained with a continuous scan, consisting of 720 projections and 3 frame averages per projection. The projections were then combined into a 3D CT image using efX-CT software by NSI (Rogers, Minnesota). A cross-section image was obtained from each 3D image and then enlarged to show each individual seed to identify the seed coat as shown in Figure 1.1 b. The seed coat thickness of each seed was then measured using the built-in length measurement tool in efX-CT software. Two thickness measurements were

performed for each scanned seed and the average of the 10 measurements of the five seeds was used as the seed coat thickness of each field replicate of each genotype.

Canning quality evaluation

The combine threshed seeds from 2020 and both belt and combine threshed seeds from 2021 were processed through canning in December 2021 following our pilot-scale canning protocol (Wang et al., 2022) at the MSU Food Processing and Innovative Center (FPIC). The belt-threshed seeds from 2020 were not canned due to limited seed. The canning process included cleaning of the seeds, soaking, blanching, filling cans, sealing, and retorting. The DRKs and LRKs were soaked in room temperature for 12h and WKs were hot soaked at 51.7°C (125°F) for 30min. The soaking water, blanching water and brine all had 100ppm of added calcium. Cans of samples were opened for evaluation of their canning quality characteristics. A picture of each sample was taken with a machine vision system (camera box). The machine vision system consists of an illumination source, a color digital camera, and an image processing software to adjust the camera settings (Mendoza et al., 2017).

Appearance scores were given to each sample by a group of trained panelists of 6 people via the pictures of samples. The rating scale was 1-5 (1 = unacceptable and 5 = excellent) and seed coat splitting was a major consideration in the rating scale of appearance (Wang et al., 2022). Texture of each sample was measured by the kilogram force needed to cut through 100g of sample using TA.XTPlus texture analyzer (Texture Technologies Corp., USA). Color of each sample was measured using Color Hunter Labscan XE colorimeter (Hunter Associates Laboratory Inc., USA). The color measurements (a*, b* and L* scores) were interpreted and presented as Hue angle (Arc tan (b*/a*)) and Chroma ($[(a*)2 + (b*)2]^{1/2}$) and L*. Hydration coefficient (HC) and washed drained coefficient (WDC) were calculated to indicate the water uptake of the beans during the soaking and canning process, respectively.

Cooking time

The belt threshed seeds of all genotypes in the ADP-kidney collection were used for cooking time measurement. The seeds were placed in a cold room (4°C, 75% relative humidity) to equilibrate for seed moisture to be in the range of 10-14% before soaking and cooking. A total of 30 seeds were soaked for each replicate of each genotype and 25 seeds were used to cook on an automated Mattson cookers apparatus (Wang & Daun, 2005) in boiling distilled water to

measure the time needed to fully cook the sample. Two replicates of each genotype were cooked and the average of the two replicates was used as the cooking time of the genotype in each year. *Data analysis*

Two data sets were generated for statistical analysis according to whether the threshing methods were involved in the phenotypic traits (responsible variables). The agronomic data including DF, DM, CBB scores, lodging scores, and seed coat thickness and cooking time data that do not involve different threshing methods were included in data set one. The SCC severity score, SCC percentage, and canning quality data (HC, WDC, texture, hue angle, chroma, and appearance scores) were included in data set two with threshing methods as a variable in the statistical model. ANOVA was used to test the statistical significance of variations caused by genotype, year, market class, and threshing methods. Tukey's Honestly Significant Difference (Tukey's HSD) test was used to conduct pairwise comparisons among market classes for all traits. Pearson's correlation analysis was used to detect the correlation between two phenotypic traits. All statistical analysis was performed in R with "Ime4" package for linear mixed-effects models (R Core Team, 2020).

Broad sense heritability ($H^2 = \frac{V_G}{V_P}$, where V_G stands for genetic variance and V_P stands for phenotypic variance) explains the proportion of phenotypic variance that is due to genetic variance. It was calculated based on the two-year data of all genotypes with two replicates in each year for seed coat thickness, cooking time, combine threshed seed coat check severity score, belt threshed seed coat check severity score, and canning quality appearance score. The calculation was conducted in R using the "variability" package (R Core Team, 2020).

Results and Discussion

Agronomic Performance

Agronomic data of the ADP-kidney collection in this study including days to flower, days to maturity, lodging, agronomic desirability score, and common bacterial blight score are shown in Figure 1.2. ANOVA results indicated that genotype had a significant effect on the variance of days to flower, days to maturity, and lodging, while year had a significant effect on the variance of CBB disease score (Table 1.5). The percentage of phenotypic variation explained by market class, year or genotype for each agronomic trait was also calculated (Table 1.6). The DF of the genotypes ranged from 35 to 55 days with an average of 43 days. Market class explained 81% of variations and genotype also explained 89% of variations in DF. The DM ranged from 83 to 107

days with an average of 94 days. Genotype explained 73% of phenotypic variations in DM. CBB is a disease that commonly occurs on bean plants in the later growing season. It was identified on all genotypes in this ADP-kidney collection with variations in severity. The CBB scores ranged from 1.75 to 4 and one genotype had a score of 5 (where 1= no symptom and 5 = very severe CBB). The lodging scores in this study skewed to the left with most of the genotypes having a lodging score of 2.5 or less. That is because most kidney beans are determinate, and lodging is usually not a problem.

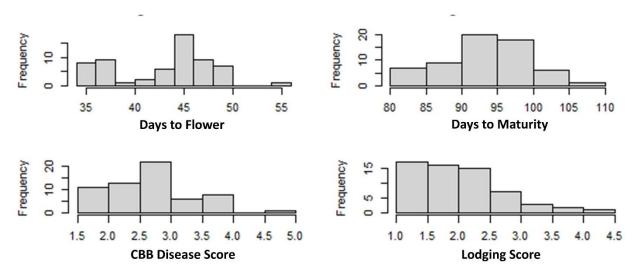


Figure 1.2 Histogram of the agronomic data of the ADP-Kidney panel, including days to flower, days to maturity, common bacterial blight (CBB) disease score, and lodging score.

Seed Coat Quality

Seed coat quality measurements included assessments of seed coat checks and seed coat thickness. Seed coat checks was evaluated under two threshing methods, belt threshing (gentle) and combine threshing (severe). The ANOVA results showed that the market class, genotype, year, and threshing methods all significantly impacted the SCC severity score and percentage. Genotype and year also significantly impacted SCC severity score and percentage when the combine threshing and belt threshing results were tested separately (Table 1.7). Among all factors, threshing methods explained the highest percentage of phenotypic variations (77%) in seed coat check severity scores (Table 1.8).

Comparison of threshing methods showed that combine threshed had 32% more seed coat check than belt threshed beans on average. The average seed coat check severity score of combine threshed beans was 1.7 ± 0.4 while belt threshed beans was 1.1 ± 0.1 (Figure 1.3a),

which proved that combine threshing method can induce more seed coat mechanical damage than the belt threshing method.

Comparison of the two years' SCC severity scores of combine threshed samples showed that 2020 harvested seeds had on average 19% more seed coat check than 2021 seeds with an average seed coat check severity score being 1.9 and 1.5, respectively (Figure 1.3b). This was mainly due to the average moisture content of the seeds in 2020 being only 9.9% and the average moisture content of the 2021 seeds being 13.8%. The importance of seed moisture content at harvest for preventing mechanical seed coat damage has been well documented in previous research (Barriga, 1961; Dexter, 1966; Forney et al., 1990). Higher moisture content can reduce mechanical seed coat damage at harvest (Uebersax et al., 2022). Research on mechanical damage to navy beans also indicated that 15% is the optimum moisture to minimize the damage (Shahbazi et al., 2011). In this study, negative correlation between seed moisture content and seed coat damage was also identified that seeds with higher moisture content at harvest tended to have lower SCC scores for both belt threshed and combine threshed samples (r = -0.53, p <0.001) (Figure 1.3 c). It would be ideal to keep a constant moisture content for the measured seeds. But this was challenging because the moisture content was affected by many factors, including the changing environment from year to year, temperature and humidity condition at harvest, and the different pace of maturity of the genotypes in research experiments. Despite the importance of seed moisture content in the prevention of seed coat checks, the control of moisture content has also been a challenge for commercial bean growers. In the conventional system, desiccants are usually used to uniformly dry down the seeds to optimize the seed moisture (Goffnett et al., 2016), however, organic growers do not have the option which makes seed coat check more of a problem in the organic system.

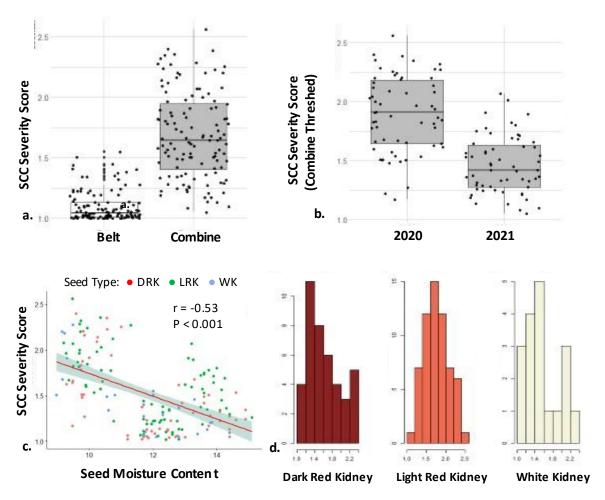


Figure 1.3 Seed coat check (SCC) severity score boxplot of belt-threshed versus combine-threshed samples in two years (a) and combine-threshed samples only from 2020 versus 2021 (b), the correlation between seed moisture content and SCC severity score of all samples from two years (c), and the histogram of SCC severity score of the combine-threshed samples for the three market classes of kidney beans (d).

Genetic variability of the SCC severity scores of combine threshed samples ranged from 1.1 to 2.4 for DRKs, 1.2 to 2.6 for LRKs and 1.1 to 2.3 for WKs (Figure 1.3 d) while the SCC percentage of combine threshed samples ranged from 4.5% to 64.5% for DRKs, 12% to 74.5% for LRKs, and 5% to 63.5% for WKs. A significant difference was found in SCC severity score and SCC percentage under the two threshing methods for each of the three market classes (Table 1.1). Among the three market classes, LRKs had the highest average SCC severity score (1.77) and SCC percentage (40.7%) in the combine threshed seeds (Table 1.1). But WKs were more vulnerable to mechanical damage than the other two types under the gentle belt threshing. There were in total 30 genotypes had a combine threshed SCC percentage of less than 20% which is an

industry standard of acceptable seed coat check percentage. These genotypes were spread through all three market classes as 15 DRKs, nine LRKs, and six WKs.

Table 1.1 Comparing the two-year average of seed coat check measurement results (SCC severity score and SCC percentage) under the two threshing methods of the three kidney market classes.

Market Class	Threshing Method	SCC Severity Score	SCC Percentage
Dark Red Kidney	Belt	1.06a*	2.7% a
	Combine	1.61b	33.0%b
Light	Belt	1.10a	4.2%a
Red Kidney	Combine	1.77b	40.7%b
White Kidney	Belt	1.19a	10.6%a
	Combine	1.55b	30.0%b

^{*}The different letters indicate the significant statistic differences of the data between the threshing methods in each market class.

The ANOVA of seed coat thickness showed that both market class and genotype had a significant impact on seed coat thickness (Supp. Table 1.3). The seed coat thickness ranged from 108.2 to 128.8 um for DRKs, 106.6 to 127.8 um for LRKs and 97 to 118.7 um for WKs. DRKs and LRKs had thicker seed coats on average than the WKs (Figure 1.4a). A negative correlation was detected between seed coat thickness and SCC severity score of belt threshed seeds (r = -0.33, p < 0.001), which indicated that seeds with thicker seed coat were more resistant to seed coat damage during the gentle belt threshing (Figure 1.4 b). However, that negative correlation was mainly caused by the WKs which has generally thinner seed coat than DRKs and LRKs. Most DRKs and LRKs had very limited seed coat damage during the belt threshing process, while the WKs were more sensitive to mechanical seed coat damage that the genotypes with thinner seed coat had more seed coat cracking. No correlation was identified between seed coat thickness and SCC severity score of combine threshed seeds. Therefore, during the combine threshing process, the moisture content is more important than seed coat thickness in terms of the beans' resilience to mechanical seed coat damage.

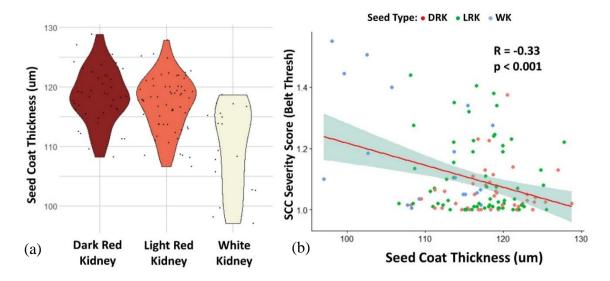


Figure 1.4 Violin plot of the seed coat thickness of the three kidney bean market classes (a) and correlation between seed coat thickness and seed coat thickness severity score cooking time (b) of dark red kidney (DRK), light red kidney (LRK), and white kidney (WK) beans in this study.

End-use Quality: Canning quality and cooking time

End-use quality measurements included canning quality evaluation and cooking time assessment. ANOVA results indicated that market class, genotype had significant impact in the variance of the canning quality traits. Threshing method did not have significant impact in water uptake within market class. But water uptake during soaking (HC) of DRKs were higher than LRKs and WKs, and the water uptake in cans (WDC) of DRK were lower than LRKs and WKs on average (Table 1.2). Most of the genotypes absorbed 1.3 to 2 times of their self-weight of water during soaking and absorbed another 1.3 to 1.7 times of their self-weight of water during canning process. The seeds that take up more water in soaking process tend to take up less water in canning process and vice versa. The average appearance scores of belt threshed DRKs and LRKs were higher than their combine threshed seeds, but there was no significant difference in WKs. The average texture (kg force to cut through 100g sample) of belt threshed LRKs was higher than the combine threshed seeds, but no significant difference was found in DRKs or WKs. The correlation analysis showed that, in combine threshed samples, SCC severity scores negatively correlated with appearance scores (r = -0.51, p < 0.001), which indicated that seeds with more seed coat damage at harvest resulted in more splits (lower appearance scores) in canned product. However, no such correlation was identified in belt threshed seeds since most of belt threshed seeds had mild or no seed coat damage. In both belt and combine threshed samples, a positive correlation was identified between texture and appearance of all canned samples (r = 0.46, p < 0.01) which indicated the samples that were firmer in texture tended to have better appearance scores (Figure 1.5). Similar discovery was found in previous research that identified higher drained weight of canned samples was associated with less acceptability of the canning quality (Forney et al., 1990). The higher drained weight could be a result of too much water uptake through cracks on seed coat therefore resulted in musher sample texture and reduced quality acceptability.

The color measurements of DRKs and LRKs were affected by the threshing method (Table 1.2). Difference was identified between belt and combine threshed DRK samples in their chroma value and between belt and combine threshed LRK samples in their L value (the higher means the color is lighter). This could be the result of more cracks in the combine threshed seeds and the exposure of the cotyledon through the cracks contributed to the detection of lighter color. The difference was not found in WKs due to the color of seed coat and cotyledon were similar.

Table 1.2 Comparing the two-year average of canning quality results including HC (hydration coefficient), WDC (washed drained coefficient), Texture (Kg Peak Force), Appearance score, Hue angle, Chroma, and L under the two threshing methods of the three kidney market classes.

Market Class	Threshing Method	нс	WDC	Appearance	Texture	Hue angle	Chroma	L
Dark Red	Belt	1.9a	1.4a	3.2a	62.3a	26.6a	21.2a	22.0a
Kidney	Combine	1.9a	1.5a	2.5b	59.3a	23.7a	19.5b	22.7a
Light	Belt	1.8a	1.6a	3.1a	58.7a	36.9a	22.6a	30.3a
Red Kidney	Combine	1.8a	1.6a	2.3b	49.7b	37.7a	22.2a	31.6b
White	Belt	1.8a	1.6a	2.6a	53.4a	75.1a	22.1a	58.8a
Kidney	Combine	1.8a	1.5a	2.2a	50.2a	74.2a	21.8a	57.9a

^{*}The different letters indicate the significant statistic differences of the data between the threshing methods in each market class.

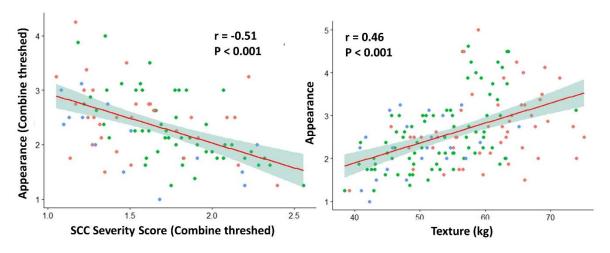


Figure 1.5 The correlation between seed coat check (SCC) severity score and appearance score of combine threshed samples, and the correlation between texture and appearance of all samples (SCC severity score: 1-5, where 1= no seed coat damage, 5= all seeds split; Appearance score:1-5, where 1= worst canning quality, and 5= best canning quality).

The ANOVA results for cooking time indicated that the variation was affected by both market classes and genotypes. The DRKs and LRKs cooked longer than WKs on average, but variations were identified within each market class (Figure 1.6). This result matches with previous research finding about cooking time of ADP beans (Cichy et al., 2015b). A previous study revealed a positive correlation between thicker seed coat and longer cooking time in unsoaked beans (Bassett et al., 2021). In this study, positive correlation was identified between seed coat thickness and cooking time (r = 0.23, p-value = 0.01) with soaked kidney beans (Figure 1.7).

Heritability of seed quality characteristics

The broad sense heritability of the seed coat check severity score (combine thresh and belt thresh), seed coat thickness, cooking time, and canning quality characteristics (hydration coefficient, washed drained coefficient, texture, hue angle, chroma and appearance) were calculated and listed in Table 1.3. The heritability of SCC severity score of combine threshed samples (H²= 0.45) was higher than that of belt threshed samples (H²= 0.36). Thus, the seed coat check severity score of combine threshed samples will be used as a selection factor instead of SCC score of belt threshed samples in the selection against mechanical seed coat damage. The color related traits (hue angle and chroma) and water uptake related traits (HC and WDC)

showed high heritability. However, they cannot be directly used as predictors of seed coat check severity level.

Table 1.3 Broad sense heritability of the end-use seed quality characteristics.

Seed Quality Characteristics	Heritability (H ²)
Seed Coat Check Severity Score (Combine thresh)	0.45
Seed Coat Check Severity Score (Belt thresh)	0.36
Seed Coat Thickness	0.41
Cooking Time	0.61
Hydration Coefficient (HC)	0.77
Washed Drained Coefficient (WDC)	0.76
Texture (Peak force)	0.57
Hue Angle	0.96
Chroma	0.73
Appearance	0.47

Selected varieties for future use

Varieties were selected according to the lowest seed coat check severity score (combine threshed samples in 2021) in each market class. All of them except one LRK genotype had less than 20% of checked seed coat, which is the industry standard for rejecting a load of beans. The selected varieties were also among the top in each market class for canning quality (appearance score), except the DRK variety 'Dynasty' which had below to average appearance score even though its seed coat checks were mild. Most of the selected varieties had cooking time lower than average in each market class, which is ideal since a shorter cooking time is preferred for variety selection.

Table 1.4 Selected varieties according to the lowest seed coat check severity score of combine threshed samples in each market class. ADP ID, variety name, seed coat check severity score, seed coat check percentage, appearance score (canning quality), seed coat thickness and cooking time of each variety are presented.

Market class	ADP ID	Variety Name	SCC Severity Score (Combine	SCC Percentag e	Appearanc e Score	Seed Coat Thicknes s (um)	Cookin g Time (Min)
Dark Red Kidney	ADP0546	Red Canadian Wonder	1.05	4.5%	3.3	112.5	21.1
Darl Kie	ADP0776 ADP0656	Dynasty Royal Red	1.14 1.17	9.5% 10.0%	1.8 4.3	125.4 124.1	30.4 26.8

Table 1.4 (cont'd)

	ADP0598	Charlevoix	1.22	13.5%	3.0	109.5	35.8
	ADP0569	MDRK	1.24	14.5%	3.4	124.0	31.6
pe /	ADP0634	UC Red Kidney	1.26	15.5%	2.9	117.9	32.8
Light Red Kidney	ADP0647	Red Kanner	1.30	15.5%	2.6	113.7	25.1
	ADP0633	TARS- HT2	1.34	21.0%	2.4	112.8	23.8
e e	ADP0674	UCD0704	1.08	6.0%	3.0	114.9	28.4
White Kidney	ADP0640	Beluga	1.10	5.0%	2.4	117.1	29.2
> \(\frac{1}{2} \)	ADP0666	USWK-6	1.13	9.0%	2.5	105.7	23.3

Conclusion

The end-use seed quality characteristics studied in this paper include the seed coat tolerance to mechanical damage, canning quality, seed coat thickness, and cooking time. The seed coat checks due to mechanical damage has been a major quality concern in the bean industry but has not been well studied in recent years. This paper described a staining test method to measure the seed coat checks in beans and expressed the results in seed coat check severity score and seed coat check percentage. This method was able to identify the variations of seed coat cracking in all three types of kidney beans from the ADP collection, especially among the combine threshed seeds. The seed coat check severity score of combine threshed seeds will be used as a selection factor for its higher heritability and significant variations compared to that of belt threshed seeds.

The seed coat thickness of kidney beans in this study was measured with the CT-scan technology, which provides a non-disruptive way to run physical measurements of the bean seeds and seed coat. Seed coat check as a factor that affects canning quality, however, does not have direct correlation with seed coat thickness. Seed moisture content plays a more important role than seed coat thickness in terms of seed coat mechanical damage especially under combine threshing method.

The white kidney beans are thinner in seed coat than the dark red kidneys and light red kidneys. White kidneys are more susceptible to mechanical seed coat damage than the other two types, and seed coat cracking happens in white kidney beans even under the gentle belt threshing method. The white kidney beans are shorter in cooking times than the dark red kidneys and light red kidneys which agreed with previous studies of dry bean cooking time.

In general, genetic variation was found in all the seed and end-use quality related characteristics. The information will provide us with resources to use for future breeding and improvement of kidney bean varieties for better seed and end-use quality.

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APPENDIX

Table 1.5 Range and mean of days to flower, days to maturity, common bacterial blight (CBB) disease scores, lodging, and desirability of ADP-Kidney collection and the ANOVA results to show statistical significance of market class, genotype and year on these traits.

		Days to Flower	Days to Maturity	CBB Disease Scores	Lodging	Desirability
Dark Red	Range	37-49.5	85-107	1-5	1-4.5	1.5-4.5
Kidney	Mean	44.7	93.7	2.7	2.2	3.3
Light Red	Range	34.5-54.5	79-105.5	1-5	1-4.5	2-5
Kidney	Mean	42.3	92.7	2.8	2.1	3.9
White	Range	35.5-50	83.5-105	1-5	1-4.5	2-4.5
Kidney	Mean	42.7	94.6	2.97	2.2	3.4
Statistical	Market	*				**
Significance	class	·	-	-	-	
	Genotype	***	***	-	***	***
	Year	-	*	***	***	NA

Significant level: p-value <0.5 *, <0.1 **, <0.001 ***, - not significant

Table 1.6 The percentage of phenotypic variation explained and the statistical significance of the impact of market class, genotype, year, and their interactions on each of the agronomic trait.

%Phenotypi %Phenotypi pp-Df Df c variation value c variation value < 0.00 80.8 89.4 Days to flower SeedType 2 < 0.01 Genotype 61 1 1 10.1 Year >0.05 Year 1 0.3 >0.05 2 9.2 >0.05 SeedType*Year >0.05 Geno*Year 58 10.3 < 0.00 Days to maturity SeedType 2 35.6 >0.05 Genotype 61 73.2 1 Year 1 34 >0.05 Year 1 8.2 < 0.01 2 58 >0.05 SeedType*Year 30.4 >0.05 Geno*Year 18.6 < 0.00 2 SeedType 1.9 >0.05 Genotype 61 48.1 1 CBB < 0.00 < 0.00 1 96.4 1 Year Year 31.3 1 1 SeedType*Year 1.7 Geno*Year 45 20.6 >0.05 >0.05 < 0.00 SeedType 2 0.8 >0.05 Genotype 61 51.7 Lodging 1 < 0.00 < 0.00 Year 1 97.8 Year 1 34.9 1 1 SeedType*Year 2 1.5 >0.05 Geno*Year 58 < 0.05 13.4

Table 1.7 Range and mean of seed coat quality including seed coat check severity score and percentage, and seed coat thickness of the ADP-Kidney beans in this study, the ANOVA results to show statistical significance of market class, genotype, year and threshing method on these traits.

				SCC	SCC	SCC		Seed
		SCC		Severity	Percent	Severi	SCC	Coat
		Severi	SCC	Score	age	ty	Percent	Thickn
		ty	Percent	(Combi	(Combi	Score	age	ess
		Score	age	ne)	ne)	(Belt)	(Belt)	(um)
Red	Range	1-2.4	0-64.5%	1.05-2.4	4.5- 64.5%	1-1.4	0-13.5%	108.2- 128.8
Dark Red	Mean	1.3	17.84%	1.6	32.96%	1.1	27.20%	118.5
Light Red	Range	1-2.6	0-74.5%	1.2-2.6	12- 74.5%	1-1.4	0-16.5%	106.6- 127.8
Ligh	Mean	1.4	22.51%	1.8	40.88%	1.1	4.15%	117.3
White	Range	1.005- 2.275	0.5- 63.5%	1.08-2.3	5-63.5%	1.0- 1.6	0-42%	97- 118.7
>	Mean	1.4	20.69%	1.6	30.61%	1.2	10.78%	109.6
e	Market class	**	**	*	*	**	**	***
ınc	Geno	***	***	***	***	*	*	***
fice	Year	***	***	***	***	***	***	_
gni	Threshing							
Sig	Method	***	***	NA	NA	NA	NA	NA
cal	Market							
sti	class*Year	**	**	NA	NA	NA	NA	NA
Statistical Significance	SeedType*Thre							
\sim	shing Method	***	***	NA	NA	NA	NA	NA

Significant level: p-value <0.5 *, <0.1 **, <0.001 ***, - not significant

Table 1.8 The percentage of phenotypic variation explained and statistical significance of the impact of market class, genotype, year, threshing method, and market class by threshing method interactions on seed coat check severity score.

Seed Coat Check Severity Score		% Phenotypic	
Seed Coat Check Severity Score	Df	variation	P value
SeedType	2	1.7	< 0.01
Geno	59	15.5	< 0.001
Year	1	4.0	< 0.001
Threshing Method	1	76.5	< 0.001
SeedType*Threshing Method	2	2.2	< 0.001

Table 1.9 ANOVA results of canning quality characteristics and cooking time.

	нс	WDC	PeakForce	HueAngle	Chroma	Appearance	Cooking Time
Market class	***	***	***	***	***	***	**
Geno	***	***	***	***	***	***	***
Year Threshing	-	-	***	-	***	**	*
Method	-	-	**	-	*	***	NA

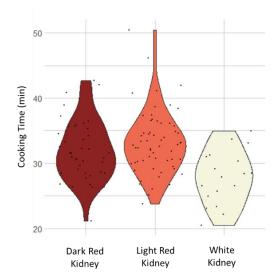


Figure 1.6 Cooking time of three kidney market classes in this study.

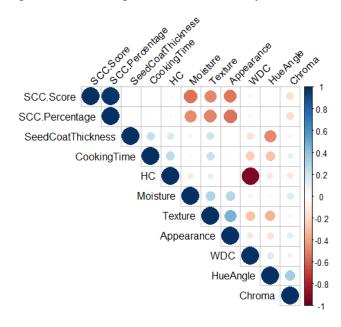


Figure 1.7 Correlation matrix of seed coat and end-use quality characteristics of all samples from two years.

CHAPTER 2: A PILOT-SCALE DRY BEAN CANNING AND EVALUATION PROTOCOL

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A Pilot-scale Dry Bean Canning and Evaluation Protocol

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Abstract

Dry beans are a nutrient-dense food that generally require long cooking times. Canned beans provide consumers safe and convenient access to this nutritious food. Canning quality is a measure of how well beans withstand the canning process and is affected by many factors including bean genotype, growing conditions, and post-harvest seed handling. The evaluation of canning quality has long been an important consideration for variety improvement in bean breeding programs. This pilot-scale dry bean canning and evaluation protocol provides a detailed step-by-step method of canning and quality evaluation for major US dry bean market classes. The post-canning process evaluation includes appearance rating by a trained sensory panel and objective measures of water uptake, color and texture. In addition, a virtual canning quality training and evaluation methods have been implemented for enhanced data quality, flexibility and engagement of the process.

Introduction

The common bean (*Phaseolus vulgaris* L.), also known as dry bean, represents a wide range of dry edible bean seed market classes that vary in size, color and shape. Dry beans are a nutrient-dense food, rich in protein, dietary fiber, minerals, and folate (Azarpazhooh and Boye, 2012). Consumption of dry beans is associated with many health benefits, including reduced risk of heart disease, managing blood sugar levels and preventing certain cancers (Hall et al., 2017). However, consumer utilization of dry beans is limited by the inconvenience of preparation and long cooking times (Doma et al., 2019; Winham et al., 2020). Canned beans are an important form of delivery of fully cooked, shelf-stable, whole beans to consumers. The canning process for beans is largely used to precook, preserve, and in some cases, add flavor to beans (Poti et al., 2015). Through this type of processing, the beans are maintained as whole seed in the same form as the raw agricultural commodity.

Canned beans: history and relevance to today's consumer

The inventions of steam retort and automated can manufacture promoted the industrialization of canning products in the late nineteenth and early twentieth century (Petrick, 2010). Canned beans were among those early canned foods in the market and became more and more common as a safe and convenient food product with the development of science and technology (Petrick, 2010). Notably, navy beans have a long history as a canned product, which gained importance and popularity with the advent of a patent for canned pork and beans referred to as baked beans by W.K. Lewis of Boston in 1877 (Bitting, 1937). Canned beans were a popular food to carry on fishing fleets in the U.S. and Great Britain as early as the 1870's (Bitting, 1937; Stilwell, 1982). The first navy bean breeding program in the U.S. was established in 1907 at the Michigan State College Agricultural Experiment Station and canning quality was a major consideration for varietal development (Andersen et al., 1960; Hedrick et al., 1931).

Cans are highly effective for the delivery of prepared foods. More processed fruit and vegetables are consumed overall than their fresh products in the United States. Canned beans consumption is nearly two times that of fresh or frozen forms (3.7 vs.1.9 pounds per capita) (Rickman et al., 2007). Research has indicated canned vegetables are as nutritious as their fresh or frozen products but are lower or comparably priced per edible amount (Miller and Knudson, 2014). Canned products require less preparation time for healthy homemade meals and reduce food waste from unconsumed and spoiled fresh produce (Can Manufacturers Institute, 2021). The canning process also destroys illness-causing microorganisms which is an important benefit as 48 million Americans get sick from foodborne illnesses each year (CDC, 2020).

Cans are highly sustainable and versatile in food packaging. It is recognized by industry leaders that up to 70 percent of cans are recycled and that more than 30 percent of steel used in cans is derived from recycled material (Williams, 2020). The steel in food cans is 100% recyclable and recycles without loss of strength or quality, while other food containers made of multiple materials can be hard to recycle and/or have significantly lower recycling rates (Can Manufacturers Institute, 2021). In general, canned foods provide convenient, safe and affordable quality food that is delivered in an environmentally friendly food package. Canned beans have the advantage of saving preparation time while providing nutritious meals.

Preparation and thermal processing of dry beans

The impact of numerous factors associated with the preparation and thermal processing of dry beans is well documented (Downing, 1996; Uebersax and Hosfield, 1985; Uebersax et al., 1991, Siddiq et al., 2010). Dry beans are hydrated with a soaking/blanching process that is prerequisite for further processing. Individual processors have established proprietary procedures that include dry bean post-harvest storage and handling (Aguilera and Rivera, 1992; Berrios et al., 1999; Paredes- López et al., 1989; Reyes-Moreno et al., 1993; Yousif et al., 2003), soaking time (Moscoso et al., 1984), soak water temperature (Hoff and Nelson, 1965; Kon, 1979; Quast DC and da Silva, 1977), blanching conditions (Drake and Kinman, 1984; Larsen et al., 1988) and water chemistry (e.g., mono and divalent cations such as calcium and sodium concentrations, or measures of total water hardness) (Wiese and Jackson, 1993; Uebersax and Bedford, 1980).

Further, dry beans are classified as a "low acid food" (pH >4.6) and require a high temperature retort process to assure commercial sterility (Anon, 2019). Factors that impact the thermal process must be understood and controlled in a standardized procedure. The numerous factors that influence heat penetration (e.g., bean moisture, bean fill weight, bean sauce/brine volume and can size) may be critical control points (Anon, 2019). Numerous retort systems are designed for uniform heating of cans and may include such factors as continuous or intermittent agitation and controlled can cooling to facilitate the adequacy of the process. The overall process conditions to assure "commercial sterility" (process time and temperature) must be established by a "process authority" to assure sufficient accumulated lethality required for food safety. A food process authority is usually a food or chemical engineer that apply their knowledge and experience to validate the process and the product. It is essential that a qualified process authority be engaged in the establishment of the process conditions.

Many innovations have been developed to improve canned beans in regard to seed handling, chemical additives such as calcium chloride, and retort temperature, time, and type. Nordstrom and Sistrunk (1979) found that beans (including pink, red kidney, navy and pinto) tend to have fewer splits after soaking when they had initially higher seed moisture content (16% compared to 10%) before processing. Junek et al. (1980) found that kidney and pinto beans soaked in solutions at 25°C had higher quality ratings than those soaked at 15°C and 35°C. Beans are usually blanched after being hydrated during the canning process. The purpose of blanching is to eliminate respiratory gases (i.e., improved uniformity of bean density), reduce the

presence of foreign material, improve color, inactivate enzymes, and achieve desirable texture of the products (Cain, 1950; Crafts, 1944; Nordstrom and Sistrunk, 1979). Research has suggested that the establishment of a blanching method should be considered separately according to bean market class (Davis, 1976). The effects of blanching methods on drained weight, texture and split rate varied between market classes. Beans not cooled after blanching had higher drained weights than those cooled after blanching (Davis et al., 1980). Calcium chloride is commonly added at various stages of the preparation (soak water, blanch water, or cover brine to improve firmness of the beans. The pectin-calcium complexes formed during soaking in bean seeds create the firmness (Uebersax and Bedford, 1980), and the firmness increases with the increase of calcium level (Balasubramanian et al., 2000). Uebersax and Bedford (1980) also found that calcium in soak water had a greater effect on the processed bean firmness than calcium in brine. However, consumers today are less interested in additives and prefer a clean label, which is also a reason people choose organic products (Kaptan and Kayısoglu, 2015; Naspetti and Zanoli, 2009). Commercial thermal process delivery systems (e.g., continuous and discontinuous retorts, with and without mechanical agitation, or hydrostatic cookers etc.) provide differential process conditions (temperature and time) that may be optimized to obtain best product quality of distinctive attributes while meeting the industrial sterilization standards to endure food safety. Uebersax and Hosfield (1985) suggested a retort at 115.6°C for 45min while Wang et al. (1988) used 121°C for 14min (navy bean) and 16min (pinto bean) to achieve the equivalent results.

Selection of a test procedure for screening the processing potential of diverse beans (i.e., cultivars and breeding lines) must consider these preparation and preservation conditions. They must also be designed to transcend the numerous specific proprietary procedures used commercially. Thus, the process must be standardized and sufficiently universal to provide appropriate rigor to be useful for the screening of beans that will withstand the typical canning process. It is frequently noted that dry bean processes exceed requirements for "commercial sterility" and, thus, beans are "cooked in the can" to achieve sufficient tenderization of the seed to yield desired palatable texture and appearance.

Canned bean quality attributes

Pilot-scale canning and evaluation protocols are essential for the evaluation of bean characteristics that are not readily apparent in the dry seeds. The protocol also allows bean breeders to assess canning quality in their breeding lines and to characterize genetic and

environmental variability for canning quality. Canning quality regarding appearance, color, and texture can be defined as how well beans withstand the canning process. After harvest and prior to canning, seeds are cleaned to remove defects that are visible on the dry seeds. The canning process ideally begins with dry seeds that fit market class expectations for seed size, shape and color. There are ten major market classes of dry bean produced in the U.S., each with a specific dry seed size, shape and color. They are dark red kidney, light red kidney, white kidney, cranberry, pinto, small red, pink, great northern, black and navy. The evaluation of canning quality includes a sensory evaluation for the appearance and objective quality traits measurements. The sensory evaluation requires a group of trained panelists to score the samples with specific standards. The quality traits measurements provide data on hydration of the seeds, and texture and color of the canned samples.

Canning quality remains a key trait in the development of new navy bean varieties and it is also important for other market classes. The black bean cultivar 'Zenith' released by Michigan State University was developed specifically for improved canning quality and anthracnose resistance while retaining the other grower preferred agronomic performance traits (Kelly et al., 2015). The recently released light red kidney bean cultivar 'Coho', navy bean cultivar 'AAC Shock' and slow-darkening (SD) pinto beans have all been tested for canning quality as an important quality trait (Kelly et al., 2020; Balasubramanian et al., 2017; Miklas et al., 2020).

The goal of this paper is to provide a detailed pilot-scale bean canning protocol for the major U.S. dry bean market classes (but can be adapted to other classes as well) and a post-canning evaluation and analysis protocol that is suitable for research purposes. This protocol adopted the major steps from the protocol developed by Hosfield and Uebersax (1980) which simulated common industrial canning processes with a small sample size. Modifications have been made according to current research results and experience in the past years and to fit a wider range of bean types. This revised protocol provides greater analytical sensitivity of key quality measures and encompasses a broader range of preparation procedures (soak/blanch time and temperature conditions) that are optimized for different commercial classes of beans based primarily on seed size than that reported by Hosfield and Uebersax (1980). The sensory evaluation has been adapted to video format and is suitable for broad review by interested individuals. This aspect of a visional "can cutting" style assessment adds considerable flexibility, engagement and utility to the protocol.

Materials and Methods

Equipment and Materials

Equipment and materials needed for canning preparation, processing and post-canning analysis are listed in Table 2.1.

Table 2.1 List of equipment and materials used in this pilot-scale canning protocol.

Name	Model, size	Manufacturers
Pre-canning preparation		
Moisture analyzer Scale Laptop computer and barcode scanner	DICKEY-John GAC 2500GMA Ohaus Scout Pro SP-401	DICKEY-john Ohaus
Mesh bags and strip tags	MIDCO Global mesh harvest bags (8"x8", 20.3cm x 20.3cm); MIDCO Global heavy-duty side to side strip tag (1/2"x7",1.3cm x 17.8cm)	MIDCO
Canning process		
Steam kettles	80 gallons and 40 gallons; Model: DL Accu- Temp	Crown Food Equipment
Strainers	10x10 inches, with 3/16" perforations	
Seamer	Model: 2006 RCM4 Closing Machine	Silgan Containers
Retort	Model: Versatort	Allpax
Cans	Size: 307x407	Silgan Containers
Water/Heat-resistant labels for cans	Laser Cryo-Babies(R), white, LxW:1.0" x 1.0"	Diversified Biotech
Post-canning analysis		
Scale		
Camera box	A Cannon digital camera remotely controlled by computer software and a stable illumination source in the box	Cannon
Texture analyzer	TA.XTPlus	Texture Technologies Corp., USA
Colorimeter	Hunter Labscan XE	Hunter Associates Laboratory Inc., USA
Paper food tray	Size: 5lb	0011
Others		

<u>Others</u>

Plastic buckets (20gal) for soaking beans; Insulated gloves; Small pitchers to pour brine into cans; Rain boots to prevent hot soaking water drips on feet.

Procedure

a.

The canning preparation and processing steps and post-canning evaluation procedure are presented in flow chart format (Figure 2.1) with details explained below. The procedure is also presented as photos in Figure 2.4.

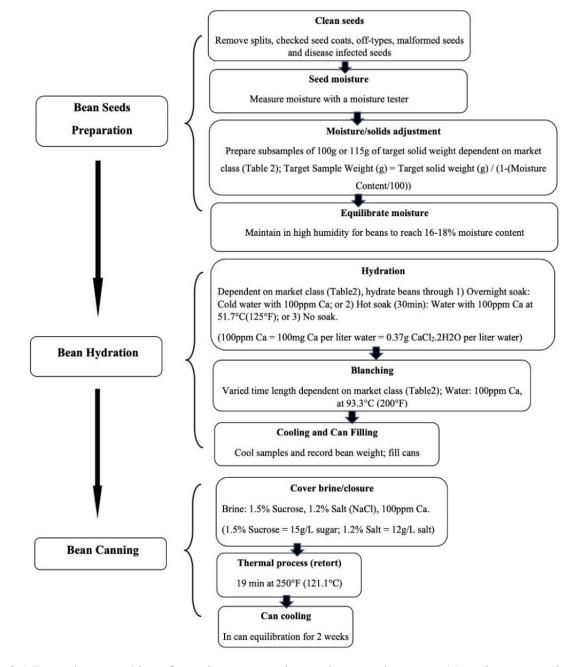
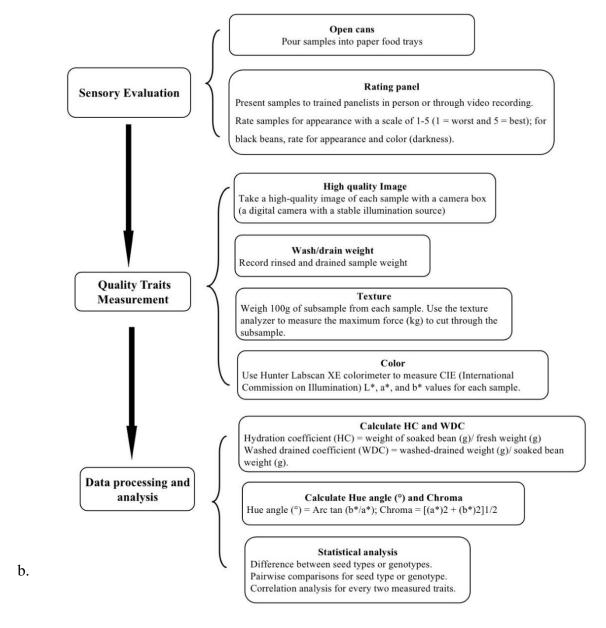


Figure 2.1 Procedure graphics of canning preparation and processing steps (a) and post-canning evaluations (b) of the pilot-scale dry bean canning and evaluation protocol.

Figure 2.1 (cont'd)



Bean seeds preparation

- i. Start with a 250g sample of dry beans.
- ii. Clean the dry beans manually to eliminate splits, checked seed coats, off-types, malformed beans and disease infected beans (Figure 2.4).
- iii. Measure moisture content of seeds with a moisture tester (Dicky-John GAC2500AGRI).

- iv. Prepare subsamples of 100g or 115g of target solid weight (moisture free basis) depending on market class (Table 2.2) based on the measured moisture content. The formula to calculate the target sample weight is: Target Sample Weight (g) = Target solid weight (g) / (1-(Moisture Content/100)). Record actual sample weight (within +/- 0.5g of target sample weight) of each subsample.
- v. Place the subsamples in mesh bags. Close bag with drawstring and attach a yellow tag with assigned bag number and corresponding barcode.
- vi. Place labeled bags in a moist environment such as cooler at high humidity to equilibrate moisture so that the moisture content of the seeds reaches to about 16-18% at canning.

Table 2.2 Soaking and blanching conditions, and the solid fill weight per can for each market class of beans.

Market class	Blanch ^{\$}	Fill Weight(g)
Overnight Soak*		
Dark Red Kidney	5 min	100
Light Red Kidney	5 min	100
Cranberry	5 min	100
Pinto	5 min	100
Hot Soak#		
Navy	5 min	115
Great Northern	15 min	115
Small Red	15 min	115
Pink	15 min	115
White Kidney	15 min	100
Yellow	15 min	100
No Soak		
Black	90 sec	115

^{*}Overnight soak: 12 hrs in cold tap water with 100ppm calcium added.

^{\$} Hot soak: 30 min at 51.7°C (125°F) in tap water with 100ppm calcium added.

[&]amp;Blanch is at 93.3°C (200°F) in tap water with 100ppm calcium added.

Soaking and canning process

The soaking and canning process was conducted at the MSU Food Processing and Innovation Center (FPIC), Okemos, Michigan, USA. Water analysis report by Wayne Chemical Inc. shows that the tap water at FPIC contains 56 ppm calcium and 48 ppm of magnesium.

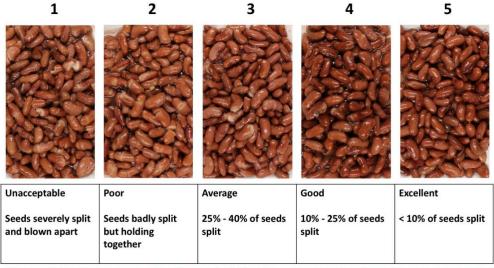
- i. Group samples by market class. Place 25 mesh bags in a larger mesh bag to ensure sufficient soaking for the samples.
- ii. Fill one large plastic bucket for each large bag of samples with tap water containing 100ppm added calcium in the form of CaCl₂. "Overnight soak" (ambient temperature) kidney (except white kidney), cranberry and pinto beans for 12 hours. Ideally, beans segregated by color classes for soaking.
- iii. Set up tables with cans placed upside down. Place labels on each can in numerical order.
- iv. On the day of canning, prepare soaking water (with 100 ppm calcium) for the samples that need hot soak at 125°F (51.7°C) (Table 2.2). Two kettles may be needed to separate the colors. Start soaking and take samples out from kettle after 30 min.
- v. Change the water for blanching (as described in Figure 1a) and increase the temperature to 200°F (93.3°C). Blanch the lighter colored beans first and followed by beans with darker color for the time length described in Table 2.2. Submerge each large mesh bag in the blanching water and start the timer.
- vi. Place bags on table to cool down. Remove small mesh bags from larger bags. Measure the soaked weight for samples if needed.
- vii. Match each small mesh bag to pre-numbered can. Transfer contents of bag into can, fold bag, place on top of the can to keep sample moist (and allow for double checking sample number if needed).
- viii. Prepare brine water with 1.5% sucrose, 1.2% salt (NaCl), 100 ppm calcium (CaCl₂). Heat brine to 200°F (93.3°C). Once all cans are filled with beans, collect bags. Fill one can to within ~1/4 inch of top with gently boiling brine. About one gallon (3.79L) of brine is needed for every ten cans.
 - ix. Closure of cans through automatic seamer, followed by inversion of cans (code label up) and stacking into basket for retort.
 - x. Retort process the cans for 19 min at 250°F (121.1°C) with a 180° inversion of the cans during the cooling cycle (a single retort holds 522 cans). Store cans for at least 2 weeks at

room temperature before opening them for evaluation. (Note: the time of the retort process was determined specifically to the retort size and based on can internal temperature for food safety.)

Post-canning evaluation

I. Appearance and color evaluation

Rating of canned bean appearance and color is conducted by a group of trained panelists. The panelists are trained via a training video that describes the dry bean market classes, rating scales, and attributes. A canning quality evaluation training video and an exercise video provides further instructions on rating (Supplemental video files). The samples are rated by at least 10 panelists on a scale of 1 to 5 (Figure 2.2). Canning quality evaluation by trained panelists may be conducted in person or through live/prerecorded videos whereby the trained panelists evaluate the samples by watching a video recording of samples (Supplementary figure 2.2). The use of video to conduct evaluations is a recent addition to the method and offers multiple benefits over in person evaluations including that each sample is presented under consistent lightning conditions and the video can be sent out to a broader group of participants.



* Color, seed size and shape are NOT rated specifically, but it is recommended to note in the comments section if non-uniform/off color, or abnormal seed size/shape are observed.

Figure 2.2 Canning quality sensory evaluation scale (1-5) for rating the appearance of all bean market classes.

In person

- i. Open cans and pour the content of each sample into labeled paper food trays (Figure 2.1b; Figure 2.5).
- ii. A group of trained panelists (at least 10 people) rate each sample with a scale of 1-5 (1 = worst and 5 = best) as described in Figure 2.2.
- iii. Evaluate black beans for appearance and color (darkness) according to Figure 2.3.



Figure 2.3 Canning quality sensory evaluation scale (1-5) for rating appearance and color of black beans (reproduce from Mendoza et al., 2017, permission requested).

Video recording

- i. Open can and pour the content into a sieve to drain out the brine. Pour each sample into its labeled paper food tray.
- ii. Record video for each sample using the "camera box" (Mendoza et al., 2017).
- iii. Trained panelists watch the recorded video and rate the samples with the evaluation scales (Figures 2.2&2.3).
- II. Canning quality related traits measurement

Digital high-quality image

- i. Drain and rinse the beans with tap water after sensory evaluation.
- ii. Take a high-quality image of each sample with the camera box.
- iii. Images can be used for further analysis such as automatic bean split detection with computer program (Long et al., 2019).

Water uptake

- i. Measure the washed drained weight for each sample.
- ii. Calculate hydration coefficient (HC) as HC = weight of soaked bean (g)/ fresh weight (g) of beans equivalent to 100g solid. This ratio indicates the weight increase of beans after soaking relative to the initial fresh weight.
- iii. Calculate washed drained coefficient (WDC) as WDC = washed-drained weight (g)/ soaked bean weight (g). WDC indicates the water uptake of the beans during the canning process.

Texture

- iv. Weigh 100g of subsample for each sample.
- v. Use the texture analyzer with the Kramer shear cell attached to measure the maximum force (kg) to cut through the subsample. This measurement is an indicator of firmness and softness of the beans.

Color

- i. Use Hunter Labscan XE colorimeter to measure CIE (International Commission on Illumination) L*, a*, and b* values for each sample. L* measures the lightness or darkness on a scare of 0 (pure black) to 100 (pure white). a* measures the redness or greenness of a sample where 0 to +50 is redness and 0 to -50 is greenness. b* measures the yellowness or blueness of a sample where 0 to +50 is yellowness and 0 to -50 is blueness.
- ii. Calculate Hue angle (°) as Arc tan (b*/a*). Each market class of thermally processed beans has a specific range of Hue angle (Table 2.3).
- iii. Calculate chroma as Chroma = $[(a^*)^2 + (b^*)^2]^{1/2}$. The higher the chroma, the brighter the color.

Table 2.3 The expected hue angles* for thermally processed beans of ten major US dry bean market classes.

Market Class	Hue Angle
Market Class	Range
Dark Red Kidney	10° to 25°
Light Red Kidney	35° to 50°

Small Red	35° to 45°
Pink	40° to 50°
Table 2.3 (cont'd)	
Cranberry	50° to 60°
Pinto	50° to 60°
Navy	70° to 80°
Great Northern	70° to 80°
White Kidney	70° to 85°
Black	20° to 40°

250 / 450

Statistical analysis

a 11 D 1

Post-canning evaluation data can be analyzed using linear mixed model with market class as fixed variable, and genotype and panelist (for sensory evaluation data only) as random effect variable to detect the difference between market classes. Use simple linear model with genotype as fixed variable to detect the variation among genotypes for canning quality measurements. Use linear mixed model with genotype as fixed variables and panelist as a random effect variable for sensory data.

Use Tukey's Honestly Significant Difference (Tukey's HSD) post-hoc test to run pairwise comparisons for market class or genotype. Use Pearson's correlation analysis to detect the correlation between each two measured traits.

Results and Discussion

A total of 28 bean varieties from the ten major US dry bean market classes were canned using the pilot-scale canning method. These varieties were obtained from dry bean farms in Michigan from 2020 growing season. The canning quality related traits measurement and sensory evaluation results of the 28 canned varieties are shown in Table 4. The difference of each trait among the tested market classes is shown in Figure 2.4.

^{*} Hue angle is calculated with the a* and b* values from colorimeter measurements, which converts the surface color values of an object to an expression of human perception for the primary colors.

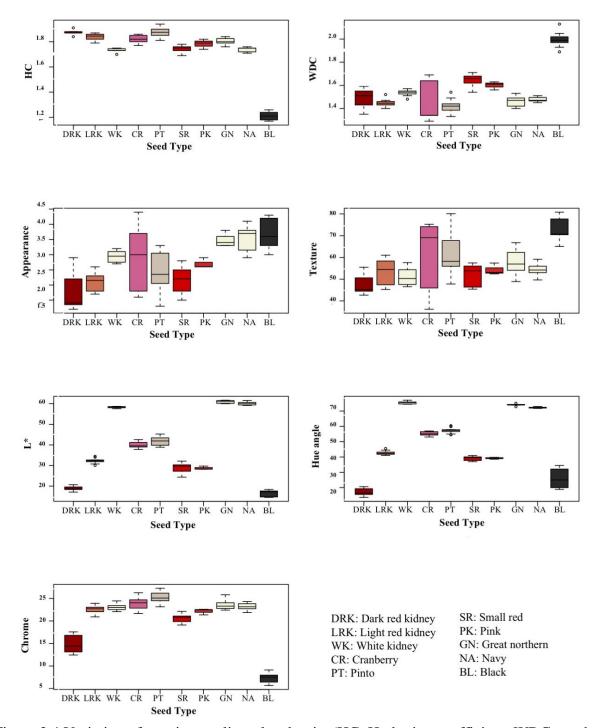


Figure 2.4 Variation of canning quality related traits (HC: Hydration coefficient; WDC: washed and drained coefficient; Appearance; Texture; Color measurements: L*, Hue angles and chrome) among ten dry bean market classes.

Hydration coefficient (HC) measures the water-uptake during soaking. The HC values for black beans were much lower than other types since the black beans were not soaked but only received a 90-second blanch. The HC values of other types of beans varied from 1.72 to 1.92 which means the beans took up about 1.8 times of water of their dry seed weight. There was no significant difference of HC among varieties, with the exception of black beans. Washed drained coefficient (WDC), which measures weight gain during canning process can vary between and within market classes with the tested varieties. The average WDC value was about 2 for black beans which indicates that black beans gained 2 times of its blanched seed weight in the can. The WDC values varied from 1.32 to 1.68 for other market classes. The appearance score reflects the overall appearance and bean splits of each genotype in the sensory evaluation as described in Figure 2.2. Scores ranged from 1.3 to 4.2 in the tested varieties. A score of 3 is usually considered acceptable. Texture, which measures the firmness of the canned beans, reflects the effort required to chew the beans and the mouthfeel (Mendoza et al., 2018). The texture values of these lines can be significantly different within market class or between market classes. The black bean samples were the firmest and dark red kidney samples were the least firm among the varieties. Cranberry bean samples had highest variability for texture (Figure 2.4). Hue angles, chroma and L* are seed coat color indicators of the varieties. Hue angle converts the surface color values of an object on the a* and b* scales to an expression of human perception for the primary colors. The hue angles of the tested varieties match with the expected values (Table 2.3) and beans with darker seed coat color (dark red kidney and black beans) have lower chroma and L* values than the other market classes (Table 2.4).

A correlation between HC and WDC was identified with R^2 = -0.9, which means the seeds that take up more water during soaking will take up less water in the cooking process in the cans. A significant correlation between appearance and texture was also identified (R^2 = 0.5), which indicates that the samples with higher appearance score tend to be firmer in texture. In general, variations were seen in most of the tested dry bean market classes (market classes) for WDC, appearance and texture. Genetic variability in canning quality could be identified using this canning protocol which is important for the selection of germplasm with improved canning quality.

Conclusion

Canning quality is a quantitatively inherited trait, and the assessment of canning quality combines a series of sensory and physical measurements. It is known that the quality of canned bean products can be affected by market class, storage time and condition, and processing method (Davis, 1976; Nordstrom and Sistrunk, 1979; Junek et al., 1980), while environmental factors and genotype x environment interactions can also have significant effects (Khanal et al., 2014; Miklas et al., 2020).

Although canning quality evaluation is challenging, a standardized and reliable canning protocol can help researchers to get consistent results and assist breeders to make selections for ideal bean varieties. The protocol presented in this paper inherited the major steps developed by Hosfield and Uebersax (1980) with adjusted soaking and blanching conditions, and the solid fill weight per can specified for different market classes. The thermal process (retort) condition was adjusted to 19 min at 121.1°C. This updated canning processing protocol enables detection of variation among bean varieties for their water up-take ability, appearance, texture and color. The modified soak and blanch method specified for each market class has minimized the effects from the canning process on the quality of canned beans, so that their genetic variations can be reliably studied. The shortened retort time at a higher temperature makes the process more efficient and improves the general texture and appearance of the canned products.

This small-scale canning protocol is useful for breeding, however there are still limitations and opportunities for improvement. It is noted that a relatively large sample size (100-115g solid weight, depending on market class) is needed for each genotype to fill the standard can using this protocol. Only a single retort time and temperature is implemented for all cans regardless of market class in this protocol. Research by Durance (1997) indicated that loss of food quality during heat sterilization can happen and the time/heat required for sterilization is usually more than needed for inactivation of deteriorative enzymes or modification of texture and flavor. Bassett et al. (2020) also found that canning quality of fast-cooking yellow beans was improved by reducing retort processing time. However, it is challenging to set varied retort time and temperature based on market class due to that a single retort holds 522 cans and our goal is to maximize efficiency with fewer retort runs. The canning and evaluation process is reserved for advanced breeding lines for which 250 g of seed is available. Earlier generations are not well suited for this analysis due to lack of seed and the likelihood that there is still residual

heterozygosity (~12.5% in an F₄ generation) that may influence the seed characteristics related to canning quality.

It can be challenging to work with trained panel in a sensory evaluation since people's perception varies and can be affected by multiple factors (Ares, 2015). A standard training video for every participant in the sensory evaluation can minimize the variation caused by human error. An increased panel size is also useful for more precise sensory results. With the virtual evaluation method that the samples are rated through recorded video, and can potentially involve trained panelists from anywhere in the world over an asynchronous schedule.

Adjustments are still been made to this protocol from year to year according to experience and canning results. The objective has been to optimize the canning procedure so that it can best fulfill the specific needs for canning quality research.

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APPENDIX



1. Manual seed cleaning.



Moisture content measurement.



3. Individual subsample preparation.



4. Soaking and/or Blanching.



5. Seeds transferred to cans.



6. Hot brine added to cans.



7. Cans sealed with an automatic sealing system.



8. Cans cooked in retort.

Figure 2.5 The flow chart of dry bean seed preparation and canning process for small-scale canning quality research.



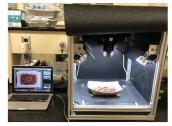
1. Cans are matched with labeled paper trays.



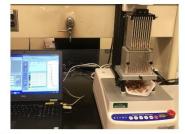
2. Sensory evaluation for appearance by a trained panel in person or through recorded video.



3. Samples weighted for drained weight.



4. High quality images are taken with a machine vision system.



5. Texture is measured as the maximum force (kg) to cut through a subsample.



6. Color is measured as CIE L*, a*, and b* values.

Figure 2.6 The flow chart of post canning evaluation for appearance and objective canning quality measurements.

Table 2.4 Canning quality related traits measurement and sensory evaluation results of varieties from ten major US dry bean market classes.

Market Class	Variety	HC ¹	WDC ²	Appearance ³	Texture(kg)	Hue angle (°)	Chroma	\mathbf{L}^*
Dark Red	Dynasty	$1.87a^{5}$	1.40a	2.4b	52.3b	19.7b	17.1b	19.3a
Kidney	Talon	1.88a	1.55b	1.3a	44.2a	15.2a	13.3a	18.6a
Liabt Dad	California Early	1.84a	1.49b	1.8a	46.4a	41.9a	23.5a	32.0a
Light Red	Coho	1.84a	1.41a	2.2b	59.6c	44.4b	21.9a	33.7a
Kidney	Rosie	1.84a	1.45ab	2.3b	54.0b	41.9a	22.3a	31.5a
White Videou	Beluga	1.73a	1.54a	3.1b	52.4a	75.8a	23.0a	58.2a
White Kidney	WhiteTail	1.74a	1.52a	2.8a	49.9a	75.2a	23.1a	58.3a
	Bellagio	1.85a	1.32a	4.0c	71.2b	56.8b	24.9a	40.0a
Cranberry	Chianti	1.80a	1.34a	3.0b	72.9b	53.9a	24.4a	38.8a
-	Etna	1.80a	1.66b	1.7a	41.5a	55.3ab	22.3a	41.5a
	Charro	1.87a	1.38a	2.9bc	67.4cd	57.3b	25.7ab	43.2a
	LaPaz	1.85a	1.43ab	2.1ab	59.0bc	57.2b	24.7ab	41.5a
Pinto	ND Falcon	1.92a	1.49b	1.7a	49.3a	57.4b	24.1a	41.3a
	ND Palomino	1.90a	1.37a	3.1c	56.9ab	60.2c	25.8ab	43.5a
	Windbreaker	1.83a	1.42ab	2.4ab	75.9d	54.7a	26.5b	39.4a
	Caldera	1.75a	1.59a	2.7b	57.0c	37.2a	19.5a	25.8a
Small Red	Ruby	1.76a	1.68a	2.1ab	45.8a	39.8ab	21.5b	30.2b
	Viper	1.72a	1.67a	1.7a	54.0b	40.2b	20.8b	30.7b
Pink	Cayenne	1.78	1.60	2.7	54.3	39.2	22.1	28.8
Cmoot	ND Pegasus	1.78a	1.51b	3.4a	50.7a	74.0a	23.0a	60.8a
Great	Powderhorn	1.82a	1.40a	3.6a	66.3c	74.7a	25.2a	60.5a
Northern	Aires	1.81a	1.47ab	3.5a	57.2b	73.7a	22.9a	61.1a
	Merlin	1.72a	1.48a	3.1a	53.0a	72.0a	23.9a	59.9a
Navy	Nautica	1.74a	1.48a	3.8b	53.7a	72.5a	22.8a	60.8a
	Medalist	1.75a	1.47a	3.8b	57.8a	72.0a	22.7a	59.4a
Market Class	Variety	нс	WDC	Appearance (Color ⁴)	Texture(kg)	Hue angles	Chroma	\mathbf{L}^*

Table 2.4 (cont'd)

	Zorro Average	1.21b 1.18	2.02a 1.84	3.4a (3.2a) 8.25 (6.74)	71.3a 4.10	24.6b 1.91	7.3b 3.52	17.3b 2.85
Black		1.18a 1.21b	2.03a 2.02a	4.2b (4.5b) 3.4a (3.2a)	68.5a 71.3a	19.3a 24.6b	6.1a 7.3b	14.6a 17.3b
	Eclipse	1.25c	1.93a	3.3a (3.0a)	78.8b	33.6c	8.4b	17.8b

^{1.} HC: Hydration Coefficient = Soak Weight/ Dry Solid Weight.
2. WDC: Washed Drained Coefficient = Drained Weight/ Soak Weight.
3. Appearance rating scale: 1-5 (5 = Excellent).
4. Color score is for black beans only, with rating scale1-5 (5 = Very dark).
5. Different letters indicate statistically significant difference within each market class, p=0.05.

SUPPLEMENTAL VIDEO FILES

- 1. Canning quality evaluation training video: https://mediaspace.msu.edu/media/t/1_9wvq41vg
- 2. Canning quality evaluation exercise video: https://mediaspace.msu.edu/media/t/1 budo7emj

CHAPTER 3: DRY BEAN POUCH PROCESSING METHOD AND QUALITY EVALUATION

Dry Bean Pouch Processing Method and Quality Evaluation

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Abstract

Flexible retort pouches have been gaining popularity as a food package to replace cans. There are many benefits of using pouches for processed bean products, but a research-scale pouch processing protocol for beans is needed. In this paper, a pouch processing method for dry beans and the subsequent quality evaluation are described. The beans used in this study included black and kidney bean genotypes from two organic field trials and two threshing methods. Pouch processing was conducted, and processing quality including texture, color, and appearance as well as cooking time of the samples were evaluated. It was found that, in black beans, genotype and year had a significant impact on the water uptake and all processing quality traits. In kidney beans, genotype had a significant impact on water uptake and processing quality traits, while year had a asignificant impact on water uptake, texture and color. Threshing method had a significant impact on appearance of processed kidney beans while had no significant impact on canning quality of black beans. This study provides a detailed methodology for pouch processing of dry beans and useful information for researchers and processors in future applications of using pouches as alternative packaging for beans.

Introduction

Dry beans are recognized as a nutritionally rich whole food and are a good source of fiber, protein, and iron, (Bouchenak & Lamri-Senhadji; 2013). However, dry bean consumption is low in many developed countries and one of the reasons is the unfamiliarity and lack of knowledge on the preparation method of beans (Schneider, 2002; Hughes et al., 2022). Nowadays, consumers also have a preference for convenient food that saves time and effort, and fits their busy lifestyles (Jackson & Viehoff, 2016). Many consumers are also willing to pay a premium for convenience foods (Swoboda & Morschett, 2001; Brunner et al., 2010).

Ever since the invention of canned food in the early twentieth century, consumers have had access to fully cooked and shelf-stable whole beans through canned bean products.

However, flexible retort pouches as an alternative food packaging that can provide the same shelf-stability of metal cans have gained more popularity in recent years and have been used in multiple types of food (Potter, 2008; Majumdar et al., 2017; Martinez-Ceniceros et al., 2022). A study has found that the demand for metal cans is experiencing slow demand growth while significant demand growth is expected for other retort packaging including pouches and cartons (Research and Markets, 2022). Retort pouches are a flexible multi-layered laminated package that can withstand thermal processing. There are many benefits of using pouches as a new form of package of processed food. They are lighter in weight, require less time and energy in thermal processing due to their thin profile, require less energy for transportation, and are easier for the consumer to open and cook (Jun et al., 2006; Potter, 2008). With less heat exposure of food in pouches during retort, it can improve the taste, color, and flavor of the product, and reduce nutrient losses (Coles and Kirwan, 2011; Featherstone, 2015). Therefore, the development of a pouch processing method for beans can potentially improve the product quality and accessibility and promote bean consumption.

The processing quality of whole beans is a measure of how well they withstand the thermal process and is usually evaluated by the appearance, color, and texture. There are many factors that affect processing quality of beans. The appearance is mainly affected by the splits of the seeds. Research has found that splits in processed whole beans are affected by seed size, seed coat cracks, and seed moisture content (Forney et al., 1990; Nordstrom and Sistrunk, 1979). The blanching methods and thermal processing methods also affect processing quality and can be adjusted to optimize the product quality (Davis, 1976; Uebersax and Hosfield, 1985). Variations in processing quality of beans are known by researchers and processors, but the organic processors are facing further challenges as the limitation on food additives and the potential inferior seed quality of organic beans. For example, EDTA that is typically used as a food preservative in canned beans is not allowed in the organic products. Calcium chloride, which is usually used in canned beans to promote a firmer texture and reduce splits of the beans, is not prohibited in organic products (The National List of Allowed and Prohibited Substances). But the organic processed bean products on the market usually avoid any additives and keep the label simply with just water and bean, because people usually think food additives are harmful to human health (Kaptan & Kayisoglu, 2015).

Despite all the benefits of using pouches to produce processed bean products, there has been a lack of protocol on research-scale bean processing in flexible pouches. The objectives of this study were to 1) establish a pouch processing method for dry beans; 2) test the pouch processing method with organic beans from our organic trials; 3) evaluate the pouch processing quality of select dry bean germplasm.

Materials and Methods

Dry bean seeds

Two organic field trials with black and kidney beans (including breeding lines and commercial varieties) were conducted in the growing seasons in 2018 (Akron, MI) and 2019 (Unionville, MI) on certified organic plots provided by Everbest Organics, the largest commercial organic bean producer in Michigan. The trials were planted as a randomized complete block design with three field plot replications and followed the same row spacing and management as the rest of the commercial field. The bean plants from 2018 field trial were hand harvested and threshed with a belt thresher (Almaco BT-14) at Michigan State University (MSU). The bean plants from 2019 field trial were hand-harvested and half of each field plot was threshed with belt thresher (Almaco BT-14) and the other half was threshed with combine thresher using a research plot combine (Hege 140). The belt threshing is a gentle process to remove seeds out from pods while the combine threshing is a more severe process that can cause mechanical seed coat damage for the seeds.

Two of the three field replications of both black and kidney beans were used for pouch processing. A total of nine black and nine kidney bean genotypes were selected from 2018 field trial and nine black (seven of them were the same genotypes as 2018 genotypes) and nine kidney (eight of them were the same genotypes as 2018 genotypes) bean genotypes were selected from 2019 trial to use in the pouch processing. The names of the black and kidney bean lines are listed in Table 3.4 and 3.5. All genotypes used for pouch processing were measured for seed coat mechanical damage and presented as seed coat check (SCC) severity score and SCC percentage. *Pouch processing*

The pouch processing method followed the major steps in our canning protocol (Wang et al., 2022) with cans being replaced by flexible pouches and some processing details being adjusted accordingly. The flexible pouches used in this study are made of four layers of materials including polypropylene, nylon, aluminum foil, and polyester from inside to outside (Figure 3.1).

There is another type of retort pouch that does not have the aluminum foil layer, which makes it microwaveable. Unlike the pouches with a metal layer that need to be sealed with a specific countertop vacuum sealer, the microwaveable pouches are easier to seal as shown in Figure 3.5.

There were three major steps of the pouch processing: 1. Seeds preparation; 2. Packaging and thermal processing; 3. Quality evaluation (Figure 3.2 and Figure 3.5). The seed preparation included cleaning the seeds, equilibrating seed moisture content to be in the range of 16-18%, and soaking and blanching of the seeds. The solid weight (moisture-free basis) of beans in each pouch was set as 90g regardless of market class. A target sample weight was calculated based on the moisture content of the seeds: Target Sample Weight (g) = Target solid weight (90g) / (1-(Moisture Content/100)). Seeds from each sample were weighed to be within +/- 0.5g of the target sample weight and the actual sample weight was recorded. Cleaned and weighed seeds were stored in a moisture room to equilibrate moisture for a week. The day before the pouch processing, the light red kidney (LRK) and dark red kidney (DRK) beans were soaked overnight (12 hours) in tap water with 100ppm added calcium (in the form of CaCl₂). On the day of pouch processing, white kidney (WK) beans were hot soaked (30 min at 125°F/51.7°C), and all samples were blanched in tap water with 100ppm added calcium with varied time length according to market class (Wang et al., 2022). Samples were then cooled to room temperature and filled in the pouches. Brine (tap water with added sugar, salt, and calcium) was prepared and cooled to < 45°C to use, and 270ml/pouch of brine was used for each pouch. Pouches in this study were sealed with a countertop vacuum sealer (Sammic SE-310 countertop vacuum packing machine) one pouch at a time.

Thermocouples (Ecklund-Harrison, Fort Myers, FL) were attached to several pouches before filling the beans in order to record the internal temperature during the retort process. The thermal process (retort) was required to achieve at least six minutes of sterilization at $250^{\circ}\text{F}/121^{\circ}\text{C}$ (F₀ = 6) to ensure the kill of all bacteria for food safety. The retort temperature for samples in this study was $245^{\circ}\text{F}/118^{\circ}\text{C}$ with an 11min cooking time to achieve the F₀ = 6. The pouch processing was conducted in November 2020 at MSU Food Processing and Innovation Center (FPIC).

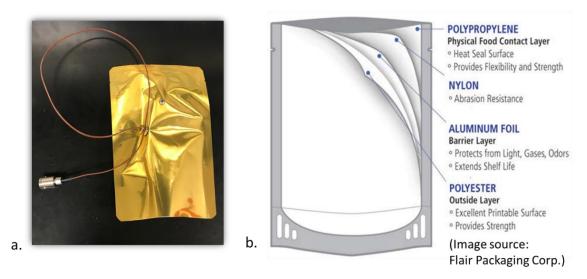


Figure 3.1 Retortable flexible pouch with thermocouple sensor attached (a) and graphical illustration of the layers and materials of the pouch(b).

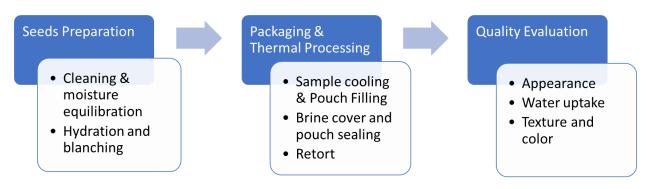


Figure 3.2 Flow chart of the major steps in the pouch processing methods.

Processing quality evaluation

The pouches with cooked samples were cooled and stored in room temperature for at least two weeks for equilibration. The quality evaluation was conducted by following the post-canning sensory evaluation protocol (Wang et al., 2022). A digital high-quality image was taken for each of the sample and used for sensory rating by a group of trained panelists. Kidney bean samples were rated for appearance with a scale of 1 - 5 (in which 1 = 1 - 5 (in which 1

USA) was used to measure the sample texture as the kg force to cut through 100g of sample. Color Hunter Labscan XE colorimeter (Hunter Associates Laboratory Inc., USA) was used to measure the color of samples with L*, a*, and b* values and interpreted as Hue angle (Arc tan (b^*/a^*)) and Chroma ($[(a^*)2 + (b^*)2]^{1/2}$). Hydration coefficient (HC) and washed drained coefficient (WDC) were calculated using the sample weight, soaked weight (measured after blanch) and drained sample weight to indicate the water uptake of beans during soaking and thermal processing, respectively.

Cooking time

The seeds of all genotypes from two years were cooked with Mattson cookers (Wang & Daun, 2005) to measure their cooking time. Seeds were equilibrated for moisture content in a cold room (4°C, 75% relative humidity) to achieve 10-14% moisture content before soaking and cooking. A total of 30 seeds were soaked for each replicate of each genotype and 25 seeds were used to cook in boiling distilled water. Two replicates of each genotype were cooked and the average of the two replicates was used as the cooking time of the genotype in each year. Data analysis

The data of black beans and kidney beans were analyzed separately. ANOVA was used to test for the statistical significance of variations caused by genotype, year, and threshing methods. The statistical analysis was performed using R software with "lme4" package for linear mixed-effects models (R Core Team, 2020).

Results and Discussion

The ANOVA results (Table 3.1) of black beans samples indicated that genotype and year both played important role in the variations in water uptake and canning quality of tested samples. There was a significant difference in cooking time over two years.

Table 3.1 ANOVA results of the impact of genotype, year, and threshing method on pouch processing quality traits including water uptake (WDC), texture, appearance, color (hue angle, chroma, and color score), and cooking time in black bean samples.

	WDC	Texture	Appearance	Hue angle	Chroma	Color	Cooking Time
Genotype	***	***	*	***	*	***	-
Year	***	***	*	***	***	***	***
Threshing Method	-	*	-	-	-	*	NA

Significant Levels: * p<0.05, ** p<0.01, *** p<0.001, - Not significant.

The black bean seeds from 2019 absorbed more water than seeds from 2018 during the retort process (Figure 3.3). Therefore, the texture of 2018 samples were significantly firmer than 2019 samples. The appearance and color scores of 2018 samples were also higher than 2019 samples. Similar finding was identified in previous chapter (Chapter one) as well as previous research (Forney et al., 1990) that the texture had a positive correlation with canning quality appearance. The threshing method (belt threshing vs. combine threshing) did not have significant effect on the water absorption or canning quality of the black bean samples in this study. It was because the seed coat damage induced by the two threshing methods was not significantly different for the black bean samples in this study (Table 3.4). The texture, appearance and color scores, cooking time, and SCC severity score and percentage data of each genotype of the black beans were listed in Table 3.4. Variations were observed for the individual genotypes in processing quality among genotypes and from year to year, but not between the two threshing methods.

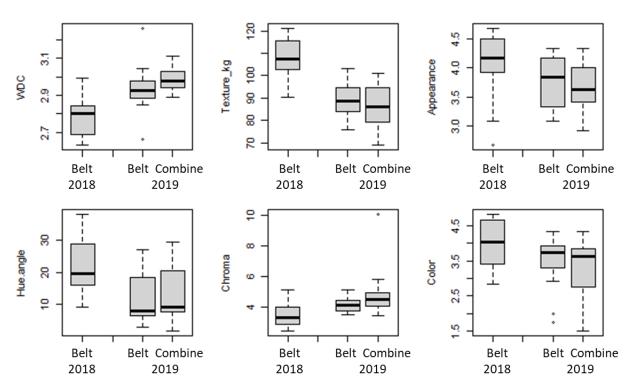


Figure 3.3 Comparing the processing quality characteristics of black beans in two years and two threshing methods (2019 only).

The ANOVA results of kidney bean samples (Table 3.2) indicated that genotype had a significant effect on water uptake (HC and WDC), texture, appearance and color. Year had a

significant effect on water uptake in the soaking process (HC) but not in the retort process. Year also had significant effect on texture, color and cooking time, but not on appearance. Threshing method had significant effect on appearance.

Table 3.2 ANOVA results of the impact of genotype, year, and threshing method on pouch processing quality traits including water uptake (HC and WDC), texture, appearance, color (hue angle and chroma), and cooking time in kidney bean samples.

	НС	WDC	Texture	Appearance	Hue angle	Chroma	Cooking Time
Genotype	***	***	***	***	***	***	-
Year	*	-	***	-	**	**	***
Threshing Method	-	-	-	**	-	-	NA

Significant Levels: * p<0.05, ** p<0.01, *** p<0.001, - Not significant.

The kidney bean samples from 2019 absorbed more water than 2018 samples during the soaking process (Figure 3.4). Like the black bean samples, 2018 kidney bean samples had firmer texture than 2019 samples. There was no significant difference in appearance scores comparing the two years, but the belt threshed samples generally had higher appearance scores than combine threshed samples. The SCC severity score and SCC percentage of each genotype (Supp. Table 3.2) indicated that the more seed coat damage was induced by combine threshing in the kidney bean samples and it resulted in more splitting seeds in the thermal processed products. One outlier was the white kidney bean 'Snowdon' which had lower appearance score in the belt threshed samples (Table 3.5), but the appearance score of both belt threshed and combine threshed samples were in the lower end of the range (being 1.5 and 2.4 respectively) which indicated an unacceptable and poor processing quality.

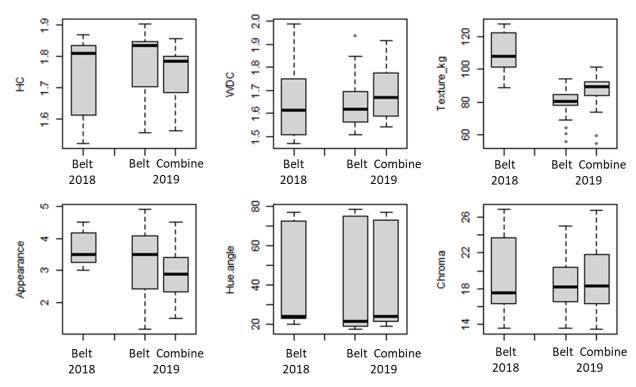


Figure 3.4 Comparing the processing quality characteristics of kidney beans in two years and two threshing methods (2019 only).

Both black beans and kidney beans had longer cooking time for 2018 samples than 2019 samples (Table 3.3). It has been well reported in previous research that environment can have a significant effect on the cooking time of dry beans (Marbach & Mayer, 1974; Chu et al., 2020). Besides the year-to-year growing environment differences, the longer storage time for 2018 samples could also affect the water absorbance and cooking time of those samples. Previous studies have shown that storage time and conditions can affect the cooking time and the hard-to-cook phenomenon in beans happens under unfavorable storage conditions (Jackson & Varriano-Marston, 1981; Reyes-Moreno et al., 1993; Coelho et al., 2007). In this study, the seeds were stored in a cool and low-humidity environment but obviously some changes still happened in the seeds and led to chemical components changes in the seeds that resulted to longer cooking time.

Table 3.3 Cooking time (range and mean) of all black beans and kidney beans in two years.

Market class		Cooking Time (min)			
Market Class		2018	2019		
Black Bean	Range	36.9 - 54.9	27.3 - 45.3		

Table 3.3 (cont'd)

	Mean	44.7	34.7
Kidney Beans	Range	37.5 - 55.3	33.7 - 45.5
	Mean	43.8	38.1

Conclusion

This study identified the variations in pouch processing quality in the organic black and kidney beans. Genotype had significant effect in all processing quality traits, thus, the selection of appropriate bean genotype is important for improvement of processing quality. The year-to-year difference was also significant in both black beans and kidney beans for processing quality and cooking time. The threshing methods did not have significant effect in the processing quality of black beans but had some effect in kidney beans. The kidney beans in this study were more susceptible than the black beans to mechanical seed coat damages happened during the more severe combine threshing.

The pouch processing method of organic dry beans in this study is innovative. The method can successfully differentiate the varieties for processing quality. The amount of seeds required for the pouch processing in this protocol is 90g, compared to 250g required for the cans. It makes the processing quality evaluation more accessible for the genotypes with less available seeds. The pouches were also easier to open and to pour the content out as compared to cans.

There are improvements can be applied to the pouch processing method in this study. The pouches used in this study had an aluminum foil layer which makes them hard to seal. The difficulty in sealing made preparation of the pouches before retorting very time-consuming. The retort temperature used in this study was 245°F/118°C compared to 250°F/121°C for cans, which leaded to a retort processing time of 11min. The retort processing time can be further shortened if the temperature was adjusted higher.

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APPENDIX



Figure 3.5 Flow chart of pouch processing and quality evaluation method with the microwaveable retort pouches.

Table 3.4 Canning quality (texture, appearance and color scores), cooking time and seed coat check (SCC) severity of each genotype of black beans in this study.

ID	Year	Threshin g Method	Texture (kg)	Appearanc e	Color	Cookin g Time (min)	SCC Severit y Score	SCC Percentag e
	2018	Belt	111.4	4.6	4.4	49.3	1.01	1.0%
B16501	2019	Belt	83.8	4.2	4.1	38.0	1.08	2.7%
	2019	Combine	82.8	4.0	4.0		1.04	2.5%
	2018	Belt	108.6	3.9	3.9	40.6	1.00	0.3%
B16504	2019	Belt	93.0	4.1	3.7	35.7	1.08	2.7%
	2019	Combine	82.3	3.4	2.2		1.01	1.0%
	2018	Belt	117.0	4.4	4.7	41.4	1.00	0.3%
B17220	2019	Belt	85.9	4.0	3.9	27.3	1.02	1.5%
	2019	Combine	99.2	3.9	3.7		1.21	10.0%
	2018	Belt	103.0	3.9	3.4	42.7	1.01	0.7%
B18504	2010	Belt	91.7	3.6	1.9	32.5	1.00	0.0%
	2019	Combine	92.1	3.6	1.9		1.04	2.0%

Table 3.4 (cont'd	Table	3.4	(cont'd)
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DI 1402 -	2018	Belt	102.4	4.4	4.6	54.9	1.01	0.3%
BL1402 15	2019	Belt	81.2	4.1	4.3	29.0	1.05	2.3%
	2019	Combine	90.1	3.8	4.0		1.01	1.0%
_	2018	Belt	90.5	3.6	4.7	53.8	1.02	1.7%
Zenith	2019	Belt	77.2	3.1	3.6	34.7	1.01	0.3%
	2019	Combine	77.5	3.7	3.8		1.04	1.7%
	2018	Belt	103.0	4.0	3.7	39.6	1.00	0.0%
Zorro	2019	Belt	101.9	3.5	3.0	29.6	1.05	2.0%
	2019	Combine	96.2	3.7	2.8		1.07	2.0%
B17922	2019	Belt	91.0	4.0	3.6	39.8	1.00	0.0%
D17922	2019	Combine	85.8	3.9	3.9		1.04	2.0%
B18204	2019	Belt	91.6	3.7	3.9	45.3	1.02	0.7%
D10204	2019	Combine	72.1	3.0	3.3		2.03	46.0%
Black	2018	Belt	111.6	3.6	2.8			
Bear	2010	Beit	111.0	3.0	2.0	43.3	1.01	1.0%
Eclipse	2018	Belt	119.7	4.2	3.7	36.9	1.00	0.3%

Table 3.5 Canning quality (texture and appearance scores) and cooking time of each genotype of kidney beans in this study.

		Threshing			Cooking	SCC	
Genotype	Year	Method	Texture	Appearance	Time	Severity	SCC
		Wichiou			(min)	Score	Percentage
	2018	Belt	104.3	4.2	37.5	1.02	1.7%
Beluga	2019	Belt	79.0	4.0	35.8	1.01	0.7%
	2019	Combine	90.3	3.5		1.16	10.0%
	2018	Belt	103.8	3.0	44.4	1.00	0.3%
K16131	2010	Belt	82.3	3.1	43.3	1.03	1.3%
	2019	Combine	90.4	2.3		1.19	11.5%
	2018	Belt	109.7	3.9	42.1	1.01	1.0%
K16136	2019	Belt	82.2	3.9	35.3	1.02	1.3%
	2019	Combine	96.4	4.3		1.17	10.7%
	2018	Belt	93.3	3.0	44.9	1.05	1.7%
K16957	2019	Belt	64.5	2.5	34.2	1.05	2.7%
	2019	Combine	75.2	2.9		1.14	8.7%
	2018	Belt	101.1	3.3	44.7	1.04	1.3%
Montcalm	2010	Belt	79.8	3.8	38.9	1.03	1.7%
	2019	Combine	89.1	3.1		1.09	6.0%
Red	2018	Belt	110.4	3.4	41.5	1.00	0.0%
Cedar	2019	Belt	92.9	4.0	34.7	1.01	1.0%

Table 3.5 (cont'd)

		Combine	89.4	2.1		1.29	16.7%
D. I	2018	Belt	123.4	4.2	55.3	1.01	0.7%
Red Hawk	2010	Belt	81.8	4.9	41.6	1.04	3.0%
Hawk	2019	Combine	87.3	3.4		1.25	14.5%
	2018	Belt	118.3	3.4	43.0	1.06	3.7%
Snowdon	2019	Belt	60.0	1.5	33.7	1.02	1.3%
	2019	Combine	56.7	2.4		1.52	28.5%
Talon	2018	Combine	125.6	4.2	40.8	1.00	0.3%
Coho	2010	Belt	88.8	2.2	45.5	1.00	0.3%
Coho	2019	Combine	97.3	1.7		1.10	6.0%

CHAPTER 4: EFFECT OF CALCIUM ON POUCH PROCESSING QUALITY IN ORGANIC DRY BEANS AND COMPARISON OF PROCESSING QUALITY IN CANS AND POUCHES

Effect of Calcium on Pouch Processing Quality in Organic Dry Beans and Comparison of Processing Quality in Cans and Pouches

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Abstract

With the increasing demand for organic dry beans, the desire for better seed and end-use product quality of organic beans is also rising. Pouch-processed bean products as a popular alternative to cans, are convenient for consumers, but the inferior product quality (such as mushy seed texture) has been a major concern, especially in organic products due to the limitation of additives. Calcium chloride is commonly used in the canning process to maintain firmness and reduce splitting of the beans, but consumers are less interested in additives and prefer a clean label. This study tested the effect of calcium usage on the quality of pouch-processed organic beans with one black bean and one kidney bean variety. The processing quality of canned and pouch-processed bean products were compared with in total of six black bean, seven kidney bean and four yellow bean samples from either organic field or conventional field. It was found the most significant impact of calcium in processing quality was on the texture while no significant impact was found on appearance scores. Firmer seed texture can be achieved by adding calcium in hydration process but not in brine so that the additives are limited in the final product. It was also found that samples with better canning quality also had better pouch processing quality (R= 0.73, p < 0.001), which means the varieties previously selected for canning quality can be directly adopted for pouch processing products. Overall, the results of this study can benefit the organic dry bean industry by providing methods to enhance the processing quality of organically produced dry beans.

Introduction

Ever since the initiation of USDA National Organic Program (NOP) standards in 2002, which started the federal regulation of organic agriculture practice and products, organic food sales have been continuously increasing in the U.S and the consumer demand for organic foods

are predicted to keep growing (Kuepper, 2010; Peng, 2019). Consumers are motivated to purchase organic food for food safety and nutrition concerns as well as a desire to protect the environment (Chekima et al., 2017; Azzurra et al., 2018, Ahmed et al., 2021). The consumer demand for organic dry beans has also been rising and more farmers have transformed their conventional farms to organic farms for the price advantage of organic products. With the establishment and growth of the organic food industry, the need for research and educational services relevant to the organic sector has increased. However, organic dry bean production is facing challenges and the inferior quality of processed organic beans is one of the major concerns. More efforts are needed to improve the end-use quality of organically produced beans.

Calcium chloride is commonly used in the canning process and is added at various stages of preparation (soak water, blanch water and brine) to maintain firmness and reduce splitting of the beans. This firmness is attributed to the pectin-calcium complexes formed during soaking (Uebersax and Bedford, 1980), and firmness increases with the increase of calcium concentration (Balasubramanian et al., 2000). Uebersax and Bedford (1980) also found that calcium in soak water had a greater effect on the processed bean firmness than calcium in brine for navy beans. Calcium chloride as a byproduct in the production of sodium carbonate (soda ash, Na₂CO₃) and a joint product from natural salt brines, is allowed to be used in organic products (The National list of allowed and prohibited substances). However, consumers today are less interested in additives and prefer a clean label, which is also a reason people choose organic products (Kaptan and Kayısoglu, 2015; Naspetti and Zanoli, 2009). Thus, the processed organic bean products on the market usually try to avoid any additives including calcium chloride.

Flexible retort pouches provide convenient and shelf-safe processed food products. Pouches are also lighter in weight and require less energy in thermal processing and transportation (Jun et al., 2006). There is a trend that pouches are replacing some canned products on the market. With the benefits of flexible pouches, preparation of organic dry beans using pouch cooking technology to deliver ready-to-eat bean products with optimized quality attributes is appealing. In-pouch cooking technology for beans will enhance the commercial viability of organic dry beans for the industry.

The objectives of this study were to 1) explore the effect of calcium usage on the quality of pouch-processed organic beans; 2) compare the processing quality of canned and pouch-processed bean products.

Materials and Methods

Dry bean seeds

Organic black bean "Adams" and dark red kidney (DRK) bean "Dynasty" were obtained from a commercial organic dry bean farm in Michigan in 2022. These two varieties were used to test the effect of changed calcium usage on the pouch processing quality of the beans.

A total of six black bean samples, seven kidney bean samples, and four yellow bean samples were collected from organic fields in 2020 and 2022 or conventional fields in 2022. They were used to compare the difference in processing quality of dry bean canned products and pouch-processed products. Organic black bean "Adams" and organic kidney bean "Dynasty" were obtained from an organic farm in 2022. Organic black bean "Black Beard" and a sample of mixed varieties, and an organic dark red kidney bean of unknown variety were purchased from an organic farm in 2020. The conventional black bean samples including "Adams", "Zenith" and "Zorro" and kidney bean samples including DRK varieties "Montcalm" and "Red Hawk", light red kidney (LRK) variety "Coho", and white kidney (WK) varieties "Beluga" and "Snowdon" and four yellow bean advanced breeding lines of the USDA-ARS EL dry bean breeding program, including Y1608-14, Y1609-14, Y1610-01 and Y1702-22 were obtained from MSU Montcalm research farm (Entrican, MI) in 2022.

A commercial pouch packaged organic black bean (Net wt. 284g) and a pouch packaged organic kidney bean product (Net wt. 284g) were purchased from Thrive Market (An American e-commerce retailer offering natural and organic food products) to compare the processing quality with our samples.

Changing the usage of calcium

Calcium chloride was used as the source of calcium in the hydration process and brine of the products. 100ppm of calcium was the amount used which is 0.37g of CaCL₂·2H₂O per liter of water. Changes in the usage of calcium in the pouch processing procedure were designed to explore the effect of calcium on the processing quality. The different calcium treatments for black beans and kidney beans were listed in Table 4.1, which included 1) adding calcium in soaking and blanching water, as well as in brine; 2) adding calcium in brine only; 3) adding calcium in soaking and blanching water only; 4) no calcium in any step.

Table 4.1 List of the different calcium usage in the pouch processing methods for the black bean and kidney bean samples.

	Code	Treatment	Description			
	A	Regular	Calcium added in blanching water and brine			
Black	В	Calcium in brine only	Calcium added in brine, not in blanching water			
B	C	No calcium in brine	Calcium added in blanching water, not in brine			
	D	No calcium	No calcium added in any step			
	A	Regular	Calcium added in soaking and blanching water, and brine			
	B Calcium in brine		Calcium added in brine, not in soaking nor blanching			
>		only	water			
Kidney	C1	No calcium in brine	Calcium added in soaking and blanching water, not in			
Kid		1	brine			
	C2	No calcium in brine	Calcium added in soaking water only, not in blanching			
		2	water nor in brine			
	D	No calcium	No calcium added in any step			

Pouch processing and canning

The pouch processing method followed the method in the previous chapter with the pouches being replaced with the microwavable retort pouches. These microwavable retort pouches were courtesy samples obtained from Flair Packaging (Appleton, WI).

The seeds cleaning and moisture equilibration were the same as in the previous chapter. The solid weight of seeds for each sample was 90g regardless of market class. The regular pouch processing method included soaking and blanching of beans in tap water with 100ppm added calcium. DRKs and LRKs were soaked overnight (12h at room temperature) and blanched for 5min. WK and yellow beans were hot soaked (30min at $125^{\circ}F/52^{\circ}C$) and blanched for 15min. The black beans were not soaked but blanched (90sec at $200^{\circ}F/93^{\circ}C$). The brine for the samples had 1.5% sucrose, 1.2% salt, 0.01% (100ppm) calcium chloride in tap water and was cooled to room temperature for use. The pouches were sealed eight at a time with an industrial vacuum sealer. The pouches with different calcium treatments were prepared separately according to the descriptions in Table 4.1. All pouches with special calcium treatments and regular treatment were processed together in the retort with temperature adjusted to $250^{\circ}F/121^{\circ}C$ (instead of $245^{\circ}F/118^{\circ}C$) and cooking time adjusted to 7.5min (instead of 11min) to achieve $F_0 = 6$ (which is at least 6min at $250^{\circ}F/121^{\circ}C$) to ensure the kill of all bacteria for food safety.

The same bean varieties used for pouch processing were also canned with a modified canning method. The black beans were not soaked but blanched for 90sec at 185°F/85°C. DRKs

and LRKs were hot soaked (instead of overnight soak) for 2hr at 130°F/54°C with 125ppm added calcium (instead of 100ppm added calcium) and blanched at 190°F/88°C for 5min. White kidney beans were hot soaked for 30min at 125°F/52°C with 100ppm added calcium and blanched at 190°F/88°C for 15min. Yellow beans were hot soaked for 30min at 125°F/52°C with 100ppm added calcium and blanched at 200°F/93°C for 5min. The brine for the cans was prepared with 1.5% sucrose, 1.2% salt, 0.005% calcium chloride (instead of 0.01%), and 0.02% EDTA. The retort was conducted at 250°F/121°C with 19min of cooking time.

Processing quality evaluation

The processing quality of both pouches and cans were evaluated following the same protocol in previous chapters. Black beans were rated for appearance and color with a scale of 1-5 (Appearance: 1 = unacceptable and 5 = excellent; Color: 1 = light brown and 5 = dark black). Kidney bean and yellow bean samples were rated for appearance only with the 1-5 scale. Texture of all samples was measured with the TA.XTPlus texture analyzer (Texture Technologies Corp., USA). Hydration coefficient (HC) and washed drained coefficient (WDC) were calculated to indicate the water uptake of beans during soaking and thermal processing, respectively. *Data analysis*

The results data for comparing calcium treatments and data for comparing pouches and cans were analyzed separately. ANOVA was used to test the impact of calcium treatments on processing quality traits, and the impact of variety and processing methods (Cans vs. Pouches) on quality traits in each market class. Pearson's correlation analysis was used to detect the correlation between two traits. All statistical analysis was performed in R with "lme4" package for linear mixed-effects models (R Core Team, 2020).

Results and Discussion

Effect of calcium in processing quality

The ANOVA results showed that different calcium treatments had significant impact on water uptake in the tested black bean but not in the dark red kidney bean. The most significant impact of calcium treatment was on texture (Table 4.2). However, the appearance score of pouch-processed products of both market classes was not affected by the use of calcium.

Table 4.2 ANOVA results of the impact of calcium treatments on processing quality traits including water uptake (HC and WDC), texture, appearance, and color (black beans only) of organic black bean and dark red kidney bean.

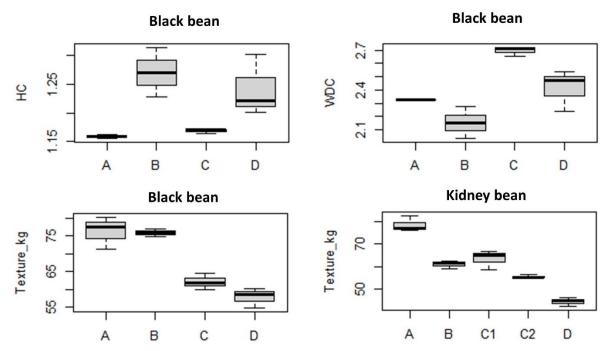
	НС	WDC	Texture	Appearance	Color
Black bean	*	**	***	-	-
Dark red kidney	-	-	***	-	NA

Significant Levels: * p<0.05, ** p<0.01, *** p<0.001, - Not significant.

The canning quality traits that were significantly affected by calcium treatments, including HC, WDC for black beans and texture value for both black beans and kidney beans were shown in Figure 4.1. It was found that black beans absorbed much more water during blanching (higher HC) when there was no calcium added in the blanching water (treatment B and D). The water uptake of black beans was significantly decreased in the retort process (lowest WDC in treatment B) when calcium was added in the brine. But with no calcium added in the brine, the black beans absorbed more water in the retort process (higher WDC in treatment C and D) regardless of the water absorption in the soaking process. Soaking and blanching is the process that allows beans to absorb water before thermal processing in order to reduce cooking time (Wainaina et al., 2021). The water uptake results of black and kidney beans under different calcium usage in this study agreed with previous research that water absorption has a significant positive correlation with the softness of the water (Del Vall et al., 1992; Thanos, 1998).

For both black and kidney bean samples, samples with the regular treatment which contained added calcium in the water in every step had the firmest texture. The texture dropped with the decrease in the use of calcium for both market classes and the samples with no calcium added at any step had the softest texture (Figure 4.1). Even though the purpose of thermal processing is to soften beans for human consumption, a certain level of firmness is still preferred (Howard et al., 2018). Previous research revealed that the calcium added in water contributed to the formation of pectin-calcium complexes which resulted in a firmer texture of the beans and the firmness of beans increased with the increase of calcium concentration (Uebersax & Bedford, 1980; He et al., 1989; Balasubramanian et al., 2000). The results of texture measurements of both the black bean and kidney beans in this study agreed with previous studies. The results also indicated that the samples had no calcium brine but were blanched or soaked in water with calcium turned to be firmer than samples being processed with no calcium at all. It indicates that

organic bean processors have the choice to use calcium in the preparation of seeds to avoid calcium in final product while improving the firmness of the beans and to avoid the mushiness texture.



A: Regular; B: Calcium in brine only; C: No calcium in brine; D: No calcium in any step C1: No calcium in brine; C2: No calcium in blanching and brine;

Figure 4.1 Boxplot of quality characteristics that significantly affected by different calcium treatments in black bean samples and kidney bean samples.

Texture of food is an important factor that affects consumer choice and acceptability. The oral processing and sensory perception of food is a very complex topic that has been specifically studied (Guinard & Mazzucchelli 1996; Stokes et al., 2013; Chen, 2014). The texture value obtained from texture analyzer for the processed beans as part of the quality evaluation only describes the hardness. This value can only partially explain the acceptability of the product since there is a lack of standards for ideal texture of processed beans in each specific market class. The texture that matches with consumer preference can be explored in future studies through sensory taste evaluation (Civille & Seltsam, 2014). Then the processing methods and the use of calcium can be further adjusted accordingly.

Comparing processing quality in cans and pouches

The processing quality traits for the five market classes in cans and pouches are compared in Table 4.3. The water uptake during soaking and blanching (HC) for cans and pouches were different in all market classes except the light red kidney beans. The difference was caused by the different soaking and blanching methods for cans and pouches. The water uptake during the retort process (WDC) was not significantly different in all market classes except dark red kidney beans. The statistical significantly different texture in two package types was found for black beans and yellow beans. But in all market classes, the pouch-processed samples had firmer texture than the canned samples. This could be the result of the reduced amount of calcium added in the cans compared to the pouches as well as the reduced thermal processing time for pouches (9min) compared to cans (19min). The appearance of black, dark red kidney and yellow bean samples of canned and pouch-processed products had statistically significant differences. The pouch-processed samples of black and yellow beans had better appearance than their canned products besides the firmer texture. This result aligns with previous chapters that texture and appearance have a positive correlation.

Previous research found that better canning quality could be achieved through reduced retort processing time for fast-cooking yellow beans (Bassett et al., 2020). The time/heat required for sterilization is usually more than needed for inactivation of deteriorative enzymes or modification of texture and flavor, and thermal processing can have undesirable influences on food quality when the process overcooks the food (Durance, 1997; Kadam et al., 2015). Therefore, with the much shorter retort processing time required for pouches than cans, pouch processing is suitable for faster cooking genotypes. It will be appealing to use fast cooking genotypes to process in pouches to obtain optimized quality for processed bean products.

Table 4.3 Comparing the canning and pouch processing quality traits including water uptake (HC and WDC), texture, appearance scores and color scores (black bean only) for the five market classes.

Market class	Package Type	нс	WDC	Texture (kg)	Appearance	Color
Black	Can	1.22a*	2.13a	63.2a	2.8a	3.4a
Diack	Pouch	1.31b	2.16a	76.2b	4.1b	4.4b
Dark Red	Can	1.87a	1.33a	67.6a	3.2a	
Kidney	Pouch	1.96b	1.42b	71.2a	2.5b	
Light Red	Can	1.93a	1.38a	70.7a	2.0a	
Kidney	Pouch	2.00a	1.49a	73.7a	1.5a	

Table 4.3 (cont'd)

White Vidney	Can	1.45a	1.83a	51.3a	1.8a
White Kidney	Pouch	1.74b	1.73a	57.2a	2.2a
Yellow	Can	1.68a	1.64a	39.8a	1.9a
I ellow	Pouch	1.90b	1.55a	55.7b	2.2b

^{*}Different letters indicate the statistically significant difference of the trait in each market class.

The appearance score of canned samples was found to be positively correlated with appearance of pouch-processed samples (R = 0.73, p < 0.001) in all five market classes (Figure 4.2). It indicated that the samples with better canning quality will have better pouch processing quality. Thus, the varieties that were selected for good canning quality can be directly used in pouch processing to achieve a better product quality.

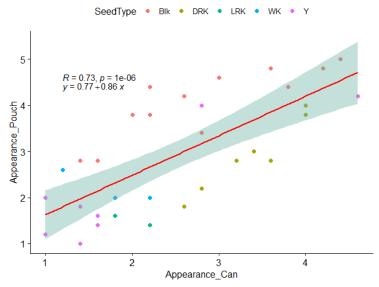


Figure 4.2 The correlation between the appearance scores of canned vs. pouch-processed samples of all tested varieties in this study.

The pouch-processed quality of different varieties of black and kidney beans are listed in Table 4.4 and was compared to the commercial product purchased from market. It was observed that the pouch processing quality varied among different varieties in both black and kidney beans. In black beans, the samples from conventional system performed better than the samples from organic system. But in kidney beans, the conventional 'Dynasty' performed the same as the organic sample. The commercial product samples in both black and kidney beans performed

poorly for the processing quality and had very mushy texture. This revealed the significant potential in the improvement of pouch processing quality of organic beans.

Table 4.4 Comparing the quality of the pouch-processed samples with regular processing method (calcium was added in water in every step) in this study and the commercial sample of black and kidney beans.

Market class	Variety	Organic vs. Conventional	Texture	Appearance	Color
	Black Beard	Organic	75.5	2.8	4.8
	Adam	Organic	76.3	3.9	3.7
	Adam	Conventional	70.8	4.5	3.6
Black	Zenith	Conventional	73.8	4.6	5
	Zorro	Conventional	88.9	4.9	4.8
	Commercial Product	Organic	47.0	1.3	1.5
	Dynasty	Organic	78.5	1.9	
	Dynasty	Conventional	63.9	1.7	
Dark Red	Montcalm	Conventional	69.8	2.9	
Kidney	Red Hawk	Conventional	68.0	3.9	
·	Commercial Product	Organic	51.2	2.2	

Conclusion

The most significant impact of calcium in processing quality is on the texture, while the impact on appearance was not significant. The samples with calcium treatment in all hydration procedure and brine had firmest texture while the samples with no calcium had the softest texture. The processors of organic pouch bean products can choose to use calcium in hydration process and avoid it in the brine to achieve firmer texture of the beans while eliminating the chemical compound in the final products.

Beans with better canning quality were found to also have better pouch processing quality. The pouch-processed samples of black and yellow beans were found to have better appearance than their canned products in this study, and the other market classes did not have significant difference in appearance in pouch and canned products. Therefore, the pouch processing method in this study can be successfully applied in the production of self-stable and ready for consumption bean products.

The retort processing time in this study was 7 min instead of 11min due to the retort temperature being adjusted from 245°F to 250°F(121°C). The shortened processing time reduces the energy consumption in the thermal processing of bean products as well as potentially improves pouch-processed the product quality.

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CHAPTER 5: MECHANICAL SEED COAT DAMAGE IN MICHIGAN DRY BEANS AND ITS EFFECT ON END-USE PRODUCT QUALITY: A HARVEST SURVEY

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Mechanical Seed Coat Damage in Michigan Dry Beans and Its Effect on End-use Product Quality: A Harvest Survey

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Abstract

The use of modern equipment in harvest and post-harvest handling of dry beans can cause seed coat checks (SCC) which results in breakage and splits in downstream product quality. SCC has been a major seed quality concern for commercial bean growers, and it can cause direct economic consequences. A survey was conducted with participation of 48 Michigan bean growers to investigate the mechanical seed coat damage occurring in Michigan-grown dry beans during harvest. Variation was identified in the collected bean samples for the SCC severity level and 9 out of 22 black bean samples had a higher than 20% SCC percentage which is considered unacceptable in the industry standard. In general, mechanical seed coat damage is a result of a series of factors including the genetics of the seeds, the seed moisture content, the environmental condition, and the operation of the combine. The choice of the right bean variety, the right equipment and timing for harvest, the suitable environment condition, the appropriate operation of harvester are all important methods to prevent severe seed coat damage.

Introduction

With the use of modern equipment in harvest and post-harvest handling of dry beans, the seed coat of beans can be damaged. The damage can result in mild to severe **seed coat checks** (**SCC**) and cause split seeds in cooked beans, therefore, affecting the quality of end-use products such as canned bean products (Figure 5.1).





Figure 5.1 Examples of dry bean seeds with mild to severe seed coat checks (SCC) and split seeds in canned product.

The most significant mechanical seed coat damages usually happen during harvest and warehouse cleaning and handling. While a series of post-harvest cleaning methods are used to remove debris and contaminants, it is difficult to separate and remove seeds with SCC. SCC has been a major seed quality concern for commercial bean growers in both organic and conventional systems and it can cause direct economic consequences. At the local elevators, over 10% of checked seeds in a dry sample (or over 20% in a soaked sample) is considered unacceptable and a lower price (discount) may be provided or even the entire load may get rejected depending on the general seed quality when they arrive (Personal conversation with the grower relations manager at ADM seed company in Michigan).

Harvest Survey Identified Variations in SCC Severity Level in Michigan Dry Beans

A survey (MSU Study ID: STUDY00006603) was conducted during the 2021 growing season to investigate the mechanical seed coat damage occurring in Michigan-grown dry beans during harvest. A total of 48 Michigan bean growers participated in the survey. The collected samples were tested for seed coat check severity and evaluated for canning quality at Michigan State University (MSU) and USDA-ARS facilities. The number of bean samples collected in each market class in this survey is shown in Figure 5.2.

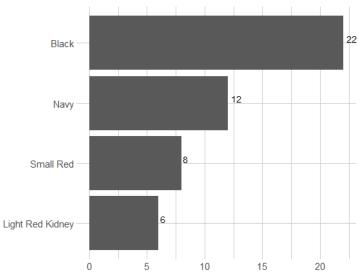


Figure 5.2 Number of samples in each market class in the survey.

The varieties of collected black beans included "Black Beard", "Specter", "Zenith", "Zorro" and "Nimbus". The varieties of navy beans included "Medalist", "Bounty" and "Merlin". The small red beans were all "Viper". The light red kidney beans included "California Early", "Big Red" and "Pink Panther". The agronomic information of the collected samples is summarized in Table 5.1 below.

Table 5.1 Summary of days to harvest, usage of harvest methods, header type, usage of harvest aid, and seed moisture at harvest of four market classes of dry beans collected in this study.

		Black	Navy	Small Red	Light Red Kidney
Days (Planting to Harvest)		93-114	92-117	92-102	92-108
Harvest Method	Direct Harvest	100%	92%	88%	
narvest Method	Windrowed		8%	13%	100%
Header Type (Direct	Auger Head	55%	73%	43%	
Harvest Only)	Draper Head	45%	27%	57%	
Harvest Aid		100%	92%	83%	83%
Seed Moisture at Harv	12.9-17%	13.2-18.8%	13.4-16%	15-19.7%	

A staining test (Figure 5.3) was used to measure the SCC of collected samples, in which seeds were soaked and then sorted into five groups. An SCC severity score was calculated accordingly to indicate the severity level of seed coat checks. SCC severity score can range from 1-5 (1 = all seeds have no seed coat damage, 5 = all seeds split). SCC percentage (%) was also calculated to indicate the number of seeds with seed coat checks in every 100 seeds.

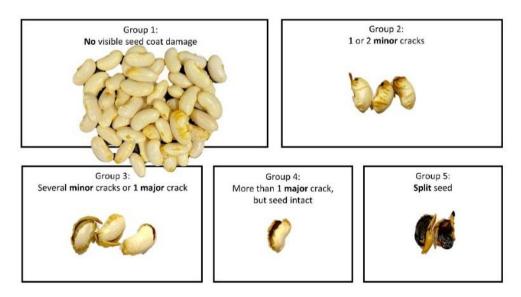


Figure 5.3 Staining test to measure the seed coat checks (SCC). Seeds were stained and grouped according to the level of SCC.

Variation was identified in the collected bean samples for the severity level of SCC (Table 5.2). The black bean samples had the biggest range of SCC severity scores (1.08 - 2.04) and SCC percentage (4.5% - 51%), and there were 9 out of 22 black bean samples had a higher than 20% SCC percentage which is considered unacceptable in the industry standard.

Table 5.2 SCC severity score and SCC percentage in each market class.

Market Class	SCC severity score*	SCC percentage	Percentage of SCC > 20%
Black	1.08 - 2.04	4.5 - 51%	40.9%
Navy	1.03 - 1.61	1.0 - 30.5%	8.3%
Small Red	1.05 - 1.51	3.0 - 22%	12.5%
Light Red Kidney	1.09 - 1.42	6.5 - 33%	33.3%

^{*} SCC severity score (1 - 5): 1 = no seed coat check and 5 = all seeds split.

Factors Affect SCC and Canning Quality

Seed Moisture

It was found that seed moisture content at harvest had a significant negative correlation (r=-0.39, P<0.01) with the SCC severity score among all the samples. The seeds with higher seed moisture at harvest tended to have less seed coat damage (lower SCC severity score) being observed in the staining test (Figure 5.4). A significant negative correlation (r=-0.66, p=<0.001) was found between the SCC severity score and appearance score in black bean samples. It indicates that the less seed coat damages the seeds had at harvest, the better they will appear in canned products. But this correlation was not identified in other market classes, probably because of the limited number of samples we were able to collect.

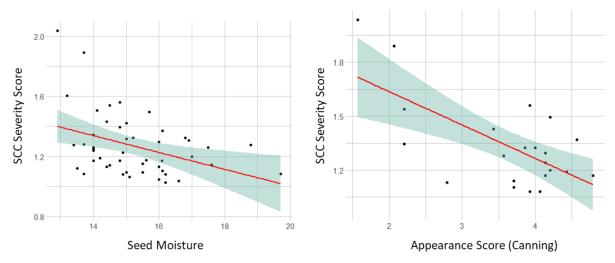


Figure 5.4 The correlation between SCC severity score and seed moisture at harvest (left), and the correlation between SCC severity score and canning quality appearance score of black beans (right).

Harvest Methods

When the samples were grouped by harvest methods (auger head combine, draper head combine, or windrowed harvest), there was no statistically significant difference among the harvest methods for SCC severity score. There were two black bean samples harvested by auger head combine that had the highest SCC severity score and were indicated as outliers in the boxplot. But these two samples also had the lowest moisture content at harvest, being 13.7% and 12.9%. There was no sample in the draper head group that had moisture content lower than 14%. Thus, we cannot conclude that the auger head combine would lead to more seed coat damage according to the data in this study.

Varieties

The SCC severity score and canning quality (appearance and color) scores can vary within the same variety (collected from different farms) in the black bean samples (Table 5.3). One "Black Beard" sample had the lowest SCC severity score of 1.08 while another had the highest SCC severity score of 2.04. The SCC severity score of "Specter" also ranged from 1.13 – 1.89. Significant differences were also identified in appearance and color scores among the 5 different varieties. The canning quality (appearance and color) of "Black beard" and "Zenith" was significantly better than "Specter".

Table 5.3 SCC severity score and canning quality scores of the different varieties in black bean samples.

Variety Name	SCC Severity Score*	Appearance	Color
Black Beard	1.08 - 2.04	1.57 - 4.79	3.29 - 4.71
Specter	1.13 - 1.89	2.07 - 2.79	1.71 -2.93
Zenith	1.11 - 1.19	3.71 - 4.43	4.21 - 4.43
Zorro	1.43	3.43	2.79
Nimbus	1.54	2.21	1.64

^{*} SCC severity score (1 -5): 1 = no seed coat check and 5 = all seeds split; Appearance (1-5): 1 = Unacceptable and 5 = Excellent; Color (1-5): 1 = light brown and 5 = dark black.

Conclusions

- Seed moisture is a very important factor affecting seed coat damage at harvest according to our survey results. In this study, a seed moisture content close to the lower end (12%) led to severe seed coat damage while seeds with moisture content at the higher end (18%) had significantly less seed coat damage. Therefore, seed moisture content of 14-18% at harvest is recommended with 16% at delivery being optimal.
- The impact of the machinery operation is another important factor for seed coat damage during harvest. The draper head has a reputation for the reduction of seed coat damage.
 However, our data do not show a statistically significant difference in SCC severity score between auger head and draper head.
- The canning quality of beans can vary among varieties as well as within the same variety.
 Samples with lower SCC severity scores received less seed coat damage during harvesting and thus tended to have better appearance scores in canning quality

evaluation. When we compared varieties in black beans, a significant difference in canning quality was identified, which indicated that variety choice is important in terms of achieving a better quality of canned bean products.

In general, mechanical seed coat damage is not a result of any single factor, but a series of factors including the genetics of the seeds, the seed moisture content, the environmental condition, and the operation of the combine. Dry beans are highly susceptible to mechanical damage and the seed coat damage may result in not only inferior end-use product quality but also impact germination and field production that leads to economic consequences for growers. Thus, extra care is always needed during the harvest and post-harvest process to maintain the integrity of the seeds. The choice of the right bean variety, the right equipment and timing for harvest, the suitable environment condition, the adjustment of the operation according to the environment, and even the right operators are all important to prevent severe seed coat damage.

APPENDIX

Farm-to-fork infographic

An educational infographic has been made on the topic of "Farm to fork: a story of beans", which demonstrates the process beans experience from farm to consumers tables. The target audience of this infographic are kids aged from 0-12. The idea is to educate them about the process that beans go through from farm to our tables in an interesting way. We believe that kids can influence their parents' purchase of food. This infographic will help us to connect with our potential bean consumers and promote the appreciation and consumption of beans.



Figure 5.5 Infographic of "Farm to fork: a story of beans" which demonstrates the process beans experience from farm to consumers tables.