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# The role of genotype and production environment in determining the cooking time of dry beans (*Phaseolus vulgaris* L.)

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## Abstract

Dry bean (*Phaseolus vulgaris* L.) is a nutrient-dense food rich in proteins and minerals. Although a dietary staple in numerous regions, including Eastern and Southern Africa, greater utilization is limited by its long cooking time as compared with other staple foods. A fivefold genetic variability for cooking time has been identified for *P. vulgaris*, and to effectively incorporate the cooking time trait into bean breeding programs, knowledge of how genotypes behave across diverse environments is essential. Fourteen bean genotypes selected from market classes important to global consumers (yellow, cranberry, light red kidney, red mottled, and brown) were grown in 10 to 15 environments (combinations of locations, years, and treatments), and their cooking times were measured when either presoaked or unsoaked prior to boiling. The 15 environments included locations in North America, the Caribbean, and Eastern and Southern Africa that are used extensively for dry bean breeding. The cooking times of the 14 presoaked dry bean genotypes ranged from 16 to 156 min, with a mean of 86 min across the 15 production environments. The cooking times of the 14 dry bean genotypes left unsoaked ranged from 77 to 381 min, with a mean cooking time of 113 min. The heritability of the presoaked cooking time was very high (98%) and moderately high for the unsoaked cooking time (~60%). The genotypic cooking time patterns were stable across environments. There was a positive correlation between the presoaked and unsoaked cooking times ( $r = .64$ ,  $p < 0.0001$ ), and two of the fastest cooking genotypes when presoaked were also the fastest cooking genotypes when unsoaked (G1, Cebo, yellow bean; and G4, G23086, cranberry bean). Given the sufficient genetic diversity found, limited crossover Genotype  $\times$  Environment interactions, and high heritability for cooking time, it is feasible to develop fast cooking dry bean varieties without the need for extensive testing across environments.

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**KEYWORDS**cooking time, dry beans, end-use quality, Genotype  $\times$  Environment, heritability, *Phaseolus vulgaris*

## 1 | INTRODUCTION

Dry beans (*Phaseolus vulgaris* L.) are an important food crop, accounting for over 30% of the total pulse production worldwide (Joshi & Rao, 2016). As a dietary staple in sub-Saharan Africa, Latin America, and the Caribbean, dry beans provide food and nutritional security (Joshi & Rao, 2016; Ojiewo et al., 2015). Sub-Saharan Africa has the greatest per capita bean consumption worldwide, and the top two countries, Burundi and Rwanda, receive 55% of dietary protein from beans (Akibode & Maredia, 2011). Dry beans are rich in protein with dry weight concentrations ranging from 20% to 29% after cooking (Katuuramu et al., 2018). They are also an excellent source of folate, potassium, iron, zinc, and dietary fiber (Havemeier, Erickson, & Slavin, 2017).

To access the dry beans' rich nutritional value, long cooking times are generally required. The cooking process is necessary to gelatinize starch, enhance protein digestibility, and inactivate lectins and trypsin inhibitors (Genovese & Lajolo, 1998; Liener & Thompson, 1980; Thompson, 2019). Beans are commonly prepared by cooking in boiling water. In some cultures, beans are soaked in water overnight (8–12 hr) prior to cooking, and in other cultures, they are cooked without soaking (Borchgrevink, 2013). Boiled whole beans are typically consumed as a whole seed in soups, stews, or baked dishes, as well as pureed into a paste (Albala, 2007).

Bean cooking times matter to consumers for two major reasons: time availability and cooking fuel scarcity. Consumer-eating patterns have shifted away from home-prepared foods to more easily prepared, convenient foods, and the long cooking times required for dry bean preparation is not viewed favorably by many consumers (IPSOS, 2010; Karlsen, Ellmore, & McKeown, 2016; Rööös et al., 2018). Fuelwood scarcity is a concern in regions such as sub-Saharan Africa where 80% of the population uses biomass for cooking (Buruchara et al., 2011; Foell, Pachauri, Spreng, & Zerriffi, 2011; Mohammed, Bashir, & Mustafa, 2015). Gathering wood for cooking is a time-consuming and labor-intensive task that typically is done by women and children (Schlag & Zuzarte, 2008). Consumers, especially in Eastern and Southern Africa markets, are often willing to pay a premium for bean varieties recognized as fast cooking (Katungi, Farrow, Chianu, Sperling, & Beebe, 2009; Mishili, Temu, Fulton, & Lowenberg-DeBoer, 2011).

Cooking time of dry beans is influenced by many factors, such as environmental conditions during production and storage, seed age, cooking method, and genetics (Stanley, 1992a). As bean seeds age, their cooking time increases. Freshly harvested beans have been shown to cook two to four times faster than beans stored for 6 months (Coelho, de Mattos Bellato, Santos, Ortega, & Tsai, 2007). In addition, when dry beans are stored in unfavorable conditions, specifically at high temperature and high humidity, cooking time greatly increases, known as the “hard-to-cook” effect (Liu & Bourne, 1995; Reyes-Moreno, Paredes-López, & Gonzalez, 1993).

Wide genetic variability for cooking time has been documented for dry beans. Under optimal growing, storage, and cooking conditions, the cooking time of over 200 dry bean lines from the *P. vulgaris* Andean Diversity Panel ranged from 16 to 90 min (Cichy, Porch, et al., 2015; Cichy, Wiesinger, & Mendoza, 2015). The few studies that have examined the genetic control of this trait suggest that it is highly heritable and controlled by a small number of genes (Cichy, Wiesinger, & Mendoza, 2015; Elia, Hosfield, Kelly, & Uebersax, 1997; Jacinto-Hernandez, Azpiroz-Rivero, Acosta-Gallegos, Hernandez-Sanchez, & Bernal-Lugo, 2003). More evidence, however, is needed on the stability of the cooking time trait in dry beans across environments encompassing diverse agroecological zones. Such information would be useful to plant breeders interested in developing fast cooking bean varieties for their local constituents. The objectives of this research were to assess the phenotypic stability of cooking time in dry bean germplasm and to characterize the role of genotype, environment, and the Genotype  $\times$  Environment interaction on the cooking time of dry beans. This study was conducted with 14 dry bean genotypes grown across 10 to 15 environments using the two soaking methods of either presoaked or unsoaked prior to cooking.

## 2 | MATERIALS AND METHODS

### 2.1 | Plant material

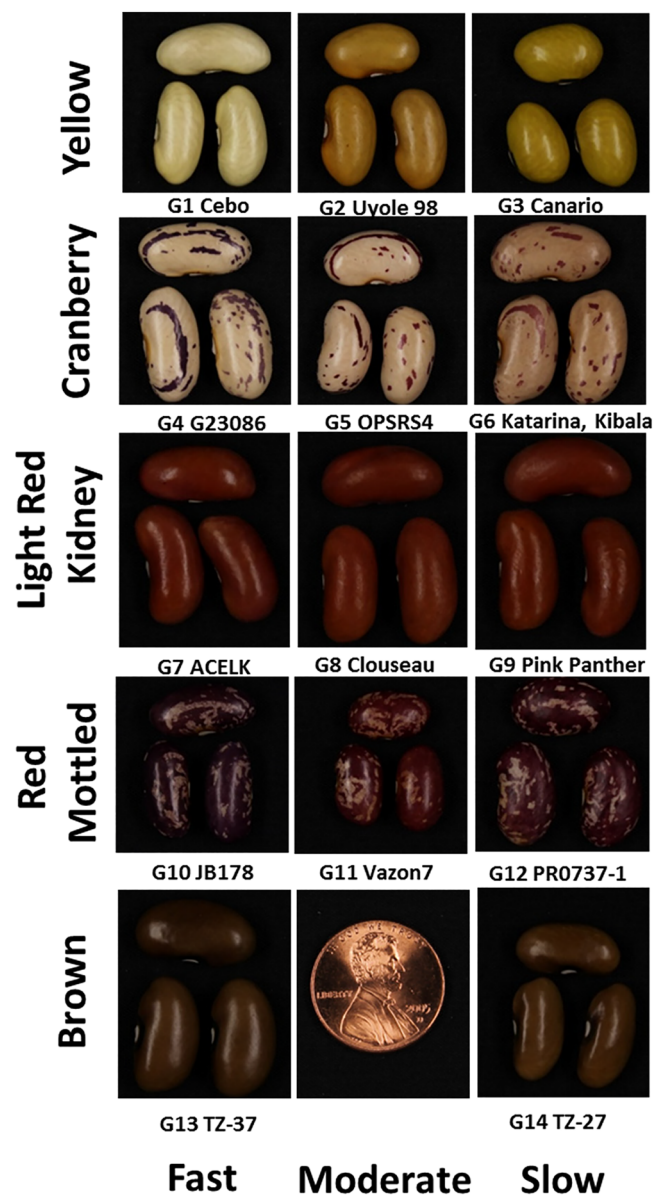
Fourteen dry bean genotypes were evaluated in this study, and these genotypes were initially identified based on cooking time evaluation of over 200 bean genotypes of the Andean Diversity Panel (Cichy, Porch, et al., 2015; Cichy, Wiesinger, & Mendoza, 2015). This set of 14 genotypes encompasses four dry bean market classes with

commercial importance in Africa, the Caribbean, and North America including yellow, cranberry (a.k.a. sugar), light red kidney, and red mottled (Figure 1). Within each of the market classes, a fast, moderate, and slow cooking genotype was identified based on data from the 2012–2013 field seasons at Montcalm Research Farm, near Entrican, Michigan (Table 1). A full description of the cooking times and nutritional composition of the materials grown in Michigan has been previously published (Wiesinger et al., 2016). A fast and slow cooking brown bean genotype was also included. Although not a widely recognized commercial market class, they are consumed in East Africa, and these particular genotypes are the parental lines of a recombinant inbred line population developed to identify quantitative trait loci for cooking

time (Table 1). All genotypes are from the Andean gene pool except for the red mottled landrace, Vazon 7 (G11), which is from the Middle American gene pool.

## 2.2 | Production environments

Fifteen environments were utilized for cooking time evaluation (Table 2). These consisted of nine field locations across the United States, Tanzania, South Africa, Puerto Rico, and Ethiopia. Multiple years were included for the Othello, WA, and Entrican, MI, USA, locations. At the Entrican location, the genotypes were grown under low soil nitrogen conditions in 2012 and 2013 (E1 and E2; Kamfwa, Cichy, & Kelly, 2015). In 2015, there was severe



**FIGURE 1** Seed images of the dry bean genotypes evaluated for cooking time across production environments. Genotypes are organized from top to bottom by market class and cooking class from left to right

**TABLE 1** Description of the dry bean genotypes evaluated for cooking time across production environments

Genotype ID	Genotype name <sup>a</sup>	ADP ID <sup>b</sup>	Seed type	Region of origin	Cooking class <sup>c</sup>
G1	Cebo	ADP 521	Yellow	Southern Africa	Fast
G2	Uyole 98	ADP 111	Yellow	East Africa	Moderate
G3	Canario	ADP 513	Yellow	Southern Africa	Slow
G4	G23086	ADP 367	Cranberry	Southern Africa	Fast
G5	OPS-RS4	ADP 113	Cranberry	Southern Africa	Moderate
G6	Katarina	ADP 515	Cranberry	Southern Africa	Slow
G7	AC ELK	ADP 618	LR kidney	North America	Fast
G8	Clouseau	ADP 680	LR kidney	North America	Moderate
G9	Pink Panther	ADP 687	LR kidney	North America	Slow
G10	JB178	ADP 436	Red mottled	Caribbean	Fast
G11	Vazon 7	ADP 443	Red mottled	Caribbean	Moderate
G12	PR0737-1	ADP 434	Red mottled	Caribbean	Slow
G13	TZ-37 (W616488)	ADP 037	Brown	East Africa	Fast
G14	TZ-27 (PI146755)	ADP 027	Brown	East Africa	Slow

Abbreviation: LR, light red.

<sup>a</sup>G number represents an entry from the CIAT Bean Germplasm Collection, and PI or W6 represents entries from the U.S. Bean Germplasm Collection; genotypes without these designations are not available in either collection.

<sup>b</sup>ADP is the Andean Diversity Panel of *Phaseolus vulgaris* (described in Cichy, Porch, et al., 2015).

<sup>c</sup>The cooking class determination was based on the presoaked cooking time data from the Andean Diversity Panel grown in Montcalm, MI, in 2012 and 2013 (Cichy, Wiesinger, & Mendoza, 2015).

Fusarium root rot disease in Entrican (E3). In Othello, WA (E4–E7), and Juana Diaz, Puerto Rico (E13 and E14), the genotypes were grown side by side under terminal drought and water-sufficient growing conditions. Table 2 provides information on the treatment, location, climate, and soil properties of each of the 15 environments.

The genotypes were planted in a randomized block design. All locations included two field replications per genotype except for Othello 2015, Cedara 2014, and Potch 2014, which each had three replications. Hawassa had a single field replicate. The number of seeds planted per replication and the plot size varied across location and experiment ranging from 50 to 160 seeds planted per plot. Agronomic data, including days to flower, days to maturity, and seed yield, were recorded at each location. A subset of approximately 200 seeds from each field replicate of each genotype were shipped to the USDA-ARS Food Legume Genetics Laboratory located at Michigan State University, East Lansing, Michigan, for cooking time analysis after harvest.

### 2.3 | Cooking time

Cooking measurements were conducted within 6 months of harvest. Prior to cooking, seeds were equilibrated in an

atmospheric cabinet (Storage Control Systems, Inc. Sparta, MI) to a moisture content of 10–14%. Raw seed weight was measured on 100 moisture-equilibrated seeds. A Mattson pin drop cooker was used to measure cooking times (Wang & Daun, 2005). The base plate of the cooker contains 25 wells, and each well holds an individual bean seed. A 70-g piercing rod rests on the center of each bean. The cooker containing 25 seeds from a single sample was placed in a metal beaker with boiling distilled water set on a hot plate. Individual beans were considered cooked when the piercing rods had passed through a seed. A sample's cooking time was recorded when 80% of the pins pierced the beans. The 80% cooking time is an approximation of a fully cooked sample at the optimal texture for consumption, which was validated with a trained sensory panel (Bassett, Cichy, & Ambechew, 2017). The presoaked cooking time was determined on beans that were soaked for 12 hr in distilled water at room temperature (pH 7 ± 0.5). The unsoaked cooking time was determined on beans that received no precooking treatment. The unsoaked cooking time was measured in only 10 of the 15 environments due to seed limitations. The unsoaked cooking time was not conducted in E1 (Entrican, 2012), E2 (Entrican, 2013), E13 (Puerto Rico, nonstress), E14 (Puerto Rico, drought), and E15 (Hawassa).



**TABLE 2** Location profiles, climate classification, and soil properties of production environments where dry bean genotypes were grown for the evaluation of cooking time

Env. ID	Location	Field site	Treatment	Year	Climate	Elevation (m)	Latitude (°)	Longitude (°)	Soil type	Soil pH
E1	USA	Entrican, MI	Nonstress	2012	Temperate	290	43.35	-85.18	Loamy sands	5.8
E2	USA	Entrican, MI	Low soil N	2013	Temperate	290	43.35	-85.18	Loamy sands	5.8
E3	USA	Entrican, MI	Nonstress	2015	Temperate	290	43.35	-85.18	Loamy sands	5.8
E4	USA	Othello, WA	Nonstress	2014	Semi-arid	110	46.79	-119.04	Coarse silty	5.9
E5	USA	Othello, WA	Drought	2014	Semi-arid	110	46.79	-119.04	Coarse silty	5.9
E6	USA	Othello, WA	Nonstress	2015	Semi-arid	110	46.79	-119.04	Coarse silty	6.6
E7	USA	Othello, WA	Drought	2015	Semi-arid	110	46.79	-119.04	Coarse silty	6.6
E8	Tanzania	Mbeya	Nonstress	2014	Subtropical highland	1,750	-8.92	33.53	Mineral	6.3
E9	Tanzania	Arusha	Nonstress	2014	Tropical savanna	1,387	-3.37	36.62	Loamy-medium texture	7.3
E10	Tanzania	Morogoro	Low soil N	2014	Tropical	500	-7.40	38.09	Clay loamy	7.1
E11	South Africa	Cedara	Nonstress	2014	Mediterranean	1,053	-29.53	30.28	Hutton	5.3
E12	South Africa	Potch	Nonstress	2014	Steppe	1,335	-26.72	27.10	Avalon	6.4
E13	Puerto Rico	Juana Diaz	Nonstress	2014	Tropical	21	18.02	-66.37	Clay loam	7.1
E14	Puerto Rico	Juana Diaz	Drought	2014	Tropical	21	18.02	-66.37	Clay loam	7.1
E15	Ethiopia	Hawassa	Drought	2015	Tropical savanna	1,708	7.05	38.47	Clay loam	7.0

Cooking time data presented in this manuscript have been previously presented in other contexts. These include genome-wide association analysis for cooking time and the relationship between cooking time and seed nutrient retention for E1, E2, and E3 (Cichy, Wiesinger, & Mendoza, 2015; Wiesinger et al., 2016), high throughput phenotyping for cooking time on E1, E2, E3, E4, E5, E6, and E7 (Mendoza et al., 2018), and the relationship between cooking time and seed carbohydrate profile in E1, E2, E3, E9, and E10 (Hooper et al., 2017).

## 2.4 | Data analysis

Levene's test for equal residual variance (Brown & Forsythe, 1974) was conducted in SAS 9.4 to check assumptions for analysis of variance (ANOVA) and other parametric tests. The residuals were not equal for all the genotype means for the soaked cooking time ( $p = .0089$ ), so the data were log transformed. Using the log transformed data, the residuals were equal for all the genotype means, and the log transformed data were used for ANOVA and heritability. The unsoaked cooking time met the assumption of equal variance of residuals, so untransformed data were used for all analyses.

ANOVA was conducted in SAS software version 9.4 of the SAS System for Windows, Copyright 2018, SAS Institute Inc. (SAS Institute, Cary, NC, USA) using the residual maximal likelihood method with the proc mixed command. For each trait evaluated, genotype and environment were considered fixed effects, and replication was a random effect. Phenotypic correlations with the presoaked cooking time and days to flowering, days to maturity, seed yield, and raw seed weight, as well as between the presoaked and unsoaked cooking times, were calculated with Spearman's rank correlation (XLSTAT, 2017). Boxplots for cooking time of individual genotypes across environments and of individual environments across genotypes were developed in R x64 3.4.3 using the boxplot command (RCoreTeam, 2018).

Broad sense heritability ( $H^2$ ) was determined on a family mean basis by the equation  $Var(G)/Var(P)$  where  $Var(P) = Var(G) + (Var(GXE)/no. env) + (Var(error)/no. env * rep)$  where  $Var$  is variance,  $G$  is genotypic, and  $P$  is phenotypic. The variance components were determined in SAS 9.4 using the proc varcomp statement with method = restricted maximum likelihood method (reml). The number of environments ( $no. env$ ) was determined as the harmonic mean of the number of environments that each of the 14 genotypes were grown in. The number of environments \* number of reps ( $no. env * rep$ ) was

determined by the harmonic mean of the total number of data points for each of the 14 genotypes (Holland, Nyquist, & Cervantes-Martínez, 2003).

Agglomerative hierarchical clustering dendrograms were developed with Euclidean distance to determine dissimilarity between each environment and each genotype (XLSTAT, 2017). With the agglomerative hierarchical clustering method, each individual group is first considered dissimilar from all others, and the clustering determines which groups should be combined. Ward's (1963) method was used as the agglomerative hierarchical clustering method. Missing data were estimated using the mean. There was an outlier in the unsoaked cooking time (G11 and E10: 380.6 min) that was not included in the clustering analysis because it deviated markedly from the other samples and skewed the results so as to cause them to be misleading for this analysis. The reason for the long cooking time of this genotype is likely because of hardshell, which is addressed in Section 4.

Genotype and Genotype  $\times$  Environment (GGE) biplots were developed using a data matrix of the mean cooking times for each genotype–environment combination with the GGEBiplotGUI package in R (Frutos, Galindo, & Leiva, 2014). The GGE biplot approximates the data matrix with singular value decomposition (Yan & Tinker, 2006). Missing data were imputed via the SVDImpute algorithm in Package “bcv” in R (Perry, 2015; Troyanskaya et al., 2001). For the presoaked cooking time, there were 8% missing data (17 values total) across 15 environments and 14 genotypes, and for the unsoaked cooking times, there were 7% missing data (10 values total) across 10 environments and 14 genotypes. In the GGEBiplotGUI R package, data were test centered ( $G + GE$ ), column metric conserved, which ranks the environments on the genotype axis, and were scaled using the standard deviation of the environment (Frutos, Galindo, & Leiva, 2014). All values were converted to negatives prior to developing the biplots because a faster cooking time is preferred and the default GGE biplot considers a larger value superior. GGE biplot figures were edited in Excel to enlarge and highlight data labels and to increase contrast to improve visualization.

Phenotypic stability was determined as the coefficient of variation (CV %) of data points from all production environments for an individual genotype calculated as the standard deviation divided by the mean and multiplied by 100. The use of the environmental variance as a measure of stability is referred to as “static phenotypic stability” and is most applicable for end-use quality traits where a constant value is sought after (Becker & Leon, 1988).



### 3 | RESULTS

#### 3.1 | Phenotypic variability for cooking time

Large phenotypic variability for cooking time was observed among the 14 dry bean genotypes evaluated in this study. The cooking times of presoaked dry beans ranged from 16 to 156 min, with a mean of 86 min and a median of 41 min across the 15 production environments. The unsoaked cooking times ranged from 77 to 381 min, with a mean of 113 min, and a median of 108 min among the 14 genotypes across 10 of the production environments (Table S1).

The cooking time variances were partitioned into genotype, production environment, and Genotype  $\times$  Environment interactions ( $G \times E$ ) to understand how each of these factors influenced the phenotypic variability observed. In the case of presoaked beans, genotype was the greatest contributor to cooking time variance (47%) followed by environment (38.6%) and lastly  $G \times E$  (11.9%; Table 3). For unsoaked beans, the greatest proportion of the variance was explained by  $G \times E$  (55.5%) followed by environment (22.2%) and finally genotype (20.2%; Table 3).

The heritability of cooking time was calculated as the ratio of the total genetic variance to the phenotypic variance (broad sense heritability). The presoaked cooking time heritability was 98% and the unsoaked cooking time heritability was 60% when all data points were included and 85% when data from G11 grown in E10 (an outlier

with a markedly longer unsoaked cooking time) were excluded (Table 3).

#### 3.2 | The genotype in relation to cooking time

The 14 dry bean genotypes were specifically selected to capture the known range of cooking time variability within globally important market classes of the Andean gene pool (Table 1 and Figure 1). The presoaked cooking times of the individual genotypes (Figure 2a) exhibited variability both among and within each of the five market classes. The within market class genotypic variability was most pronounced among the three red mottled genotypes (G10, G11, and G12) where there was a difference of 40 min between the fastest and the slowest genotypes. The among market class genotypic variability was exhibited by the generally longer cooking times of the light red kidney and the red mottled genotypes as compared with the yellow, cranberry, and brown market classes. The three fastest cooking genotypes were fully cooked on average in 30 min or less and included G1 (yellow, Cebo, 28 min), G4 (cranberry, G23086, 29 min), and G13 (brown, TZ-37, 30 min). The slowest cooking genotype was G12 (red mottled, PR0737-1, 80 min).

The unsoaked cooking times of individual genotypes were 1.7 to 3.3 times longer than their presoaked counterparts (Table S1). The unsoaked cooking times of individual genotypes (Figure 2b) showed larger interquartile ranges as compared with the presoaked beans indicating

**TABLE 3** Analysis of variance and broad sense heritability of cooking time for presoaked and unsoaked bean genotypes across production environments

Cooking time (presoaked)						
Effect <sup>a</sup>	Num DF <sup>b</sup>	Den DF <sup>c</sup>	% Phenotypic variation	F value	<i>p</i> > <i>F</i>	Broad sense heritability (H <sub>2</sub> )
Genotype (G)	13	238	47.0	334.91	<0.0001	
Environment (E)	14	238	38.6	255.57	<0.0001	98%
G $\times$ E	165	238	11.9	6.67	<0.0001	
Cooking Time (unsoaked)						
Effect	Num DF	Den DF	% Phenotypic variation	F value	<i>p</i> > <i>F</i>	Broad sense heritability (H <sub>2</sub> )
Genotype (G)	13	129	20.2	85.59	<0.0001	
Environment (E)	9	129	22.2	135.55	<0.0001	60% (with outliers)
G $\times$ E	107	129	55.5	28.51	<0.0001	85% (no outliers)

<sup>a</sup>Type 3 tests of fixed effects in proc mixed with REML estimation method.

<sup>b</sup>Numerator degrees of freedom for the *F* value calculation.

<sup>c</sup>Denominator degrees of freedom for the *F* value calculation.

larger cooking time variability of a given genotype across environments (Figure 2a,b). This was also reflected with a lower heritability for unsoaked as compared with the presoaked cooking times (Table 3). The two fastest unsoaked cooking genotypes, G1 (Cebo, 91 min) and G4 (G23086, 96 min), were also identified as fastest in the presoaked cooking evaluations. The red mottled genotypes showed the greatest within market class variability for the unsoaked cooking times, and one outlier was detected in this market class, G11 (Vazon 7), which took 381 min to cook in E10 (Morogoro, Tanzania).

### 3.3 | Cooking time classification

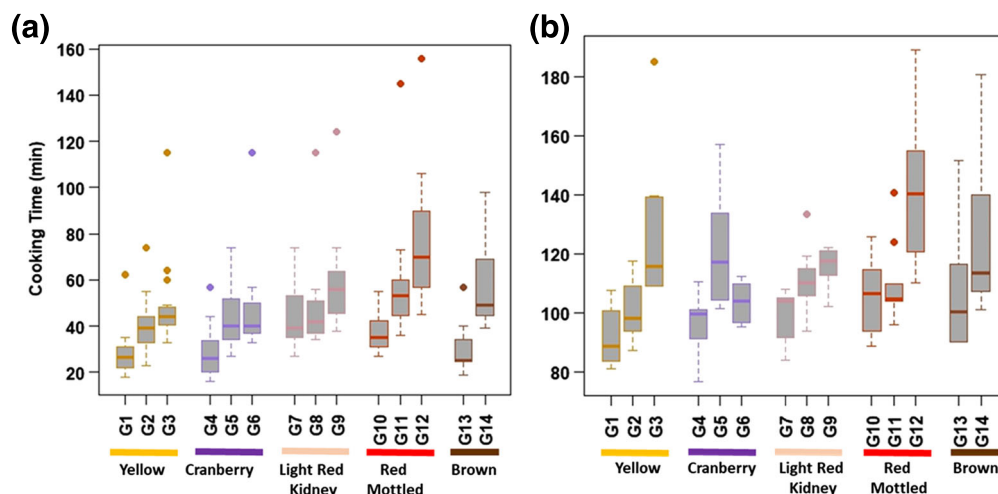
Cooking time classification into fast, moderate, and slow is a potentially useful tool to describe cooking trends across environments or multiple experiments. The original selection and classification of the 14 genotypes used in this study (Table 1 and Figure 1) were based on the evaluation of the presoaked cooking times of the Andean Diversity Panel with over 200 genotypes grown for 2 years in Montcalm, MI (E1 and E2; Cichy, Wiesinger, & Mendoza, 2015). With the additional 13 environments, and the inclusion of the unsoaked cooking time data, the genotypes were reclassified using agglomerative hierarchical clustering with Wade's method. The results of this reclassification within the presoaked cooking times were the grouping of seven genotypes into a fast cooking group, six genotypes into a moderate cooking group, and one genotype into a slow cooking group (Figure 3 a). This reclassification is different from the original

classification by extending the number of genotypes in the fast and moderate groups, while reducing the number of genotypes in the slow group. The reclassification still contains the original four fast cooking genotypes but now also includes three additional lines that were originally classified as moderate cooking. The moderate group contains four genotypes previously classified as slow cooking. With this cooking time reclassification using the complete set of environments, it is important to note that there were no changes in rank resulting in a fast cooking reclassified as slow cooking.

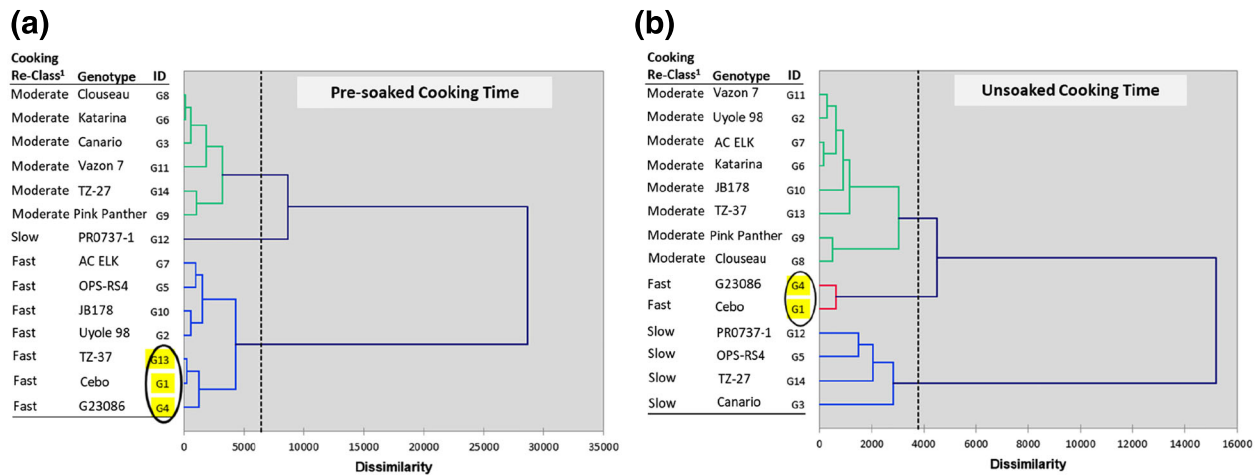
The unsoaked cooking times were classified with two fast cooking, eight moderate cooking, and four slow cooking genotypes. The two genotypes that were classified as fast cooking when unsoaked were also the two fastest cooking genotypes when presoaked. They are highlighted in yellow in Figure 3a,b. There was only one change of rank in the unsoaked as compared with the soaked groups. In the cranberry class, G5 is ranked as slow when unsoaked, but when presoaked, G5 is ranked as fast. This is an indication of a crossover Genotype  $\times$  Cooking method interaction. In this case, selecting genotypes based on the presoaked cooking times would not have been the best strategy if the consumers decided not to presoak.

### 3.4 | The production environment in relation to cooking time

The 15 environments selected to produce the dry beans for cooking time evaluation include a combination of locations, years, and field treatments. They include nine unique locations in the United States, the Caribbean, East



**FIGURE 2** Boxplots of cooking times. (a) Presoaked cooking time genotype boxplot: cooking times (presoaked) with genotype as the main effect. Boxplot depicts 14 dry bean genotypes across 15 environments. (b) Unsoaked cooking time genotype boxplot: cooking times (unsoaked) with genotype as the main effect. Boxplot depicts 14 dry bean genotypes across 10 environments. There was an unsoaked cooking time outlier of 381 min for G11 grown in E10 that is not shown. Information on individual genotypes is available in Table 1



**FIGURE 3** Clustering and reclassification<sup>(1)</sup> of genotypes into fast, moderate, and slow cooking across all environments. Agglomerative hierarchical cluster dendrograms of 14 dry bean genotypes: (a) the presoaked cooking time across 15 environments and (b) the unsoaked cooking times across 10 environments. Genotypes highlighted in yellow are the fastest cooking. The dotted lines indicate the point at which the number of clusters was determined. The x axis is the dissimilarity between genotypes, and it is a squared Euclidean distance and should not be interpreted as a metric but as an indication of divergence

Africa, and South Africa. All nine locations are research stations used by breeders in the development and evaluation of new dry bean varieties. Six of these locations are in regions important for commercial dry bean production (Michigan, USA; Mbeya and Arusha, Tanzania; Potch and Cedara, South Africa; and Hawassa, Ethiopia).

Although some variability across environments for the presoaked cooking times was observed, there was minimal variability within locations across years and/or treatments (Figure 4a). For example, all 3 years of data for Entrican, USA (E1–E3), had very similar boxplot distributions. The four Othello, USA, environments (E4–E7) had similar boxplot distributions, although there appears to be a trend such that the drought stress (E5 and E7) had slightly longer cooking times than the nondrought stress environments (E4 and E8).

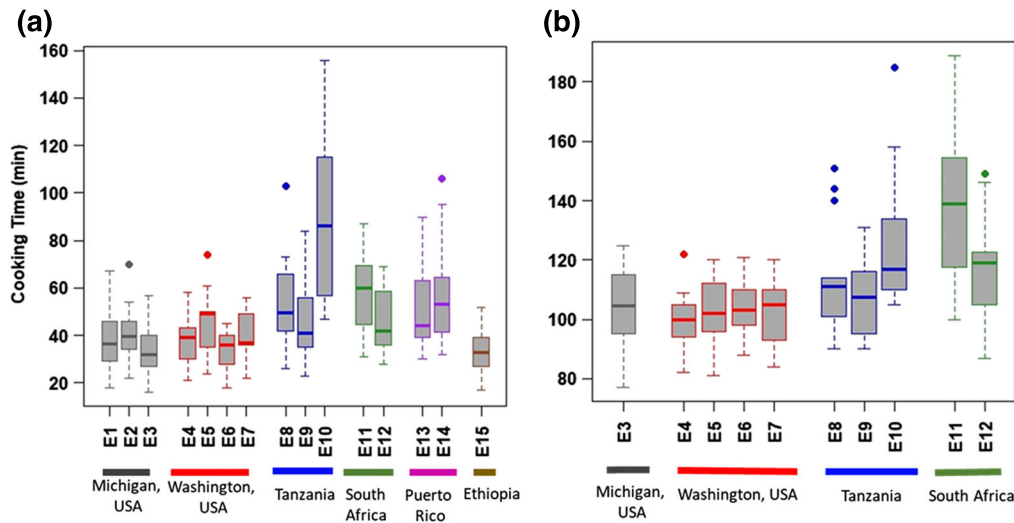
Variability among locations indicated that Entrican and Othello, USA, and Hawassa, Ethiopia, environments had similar “shorter” cooking times, whereas Morogoro, Tanzania, Cedara, South Africa, and Juana Diaz, Puerto Rico, environments have similar “longer” cooking times. The environment with the longest cooking times was E10 (Morogoro, Tanzania). Genotypes grown in Morogoro took 1.6–2.4 times longer to cook as compared with the genotypic averages across all environments. Morogoro has a tropical climate with high day and nighttime temperatures, which could explain the longer cooking times. Juana Diaz, Puerto Rico (E13 and E14), also has a tropical climate, and although the average cooking times observed in Puerto Rico were long (51 and 58 min, respectively, for E13 and E14), they were still less than Morogoro, suggesting that other factors are

contributing to the long cooking times of the beans grown in Morogoro.

The unsoaked cooking times of individual environments followed a similar pattern as the presoaked times (Figure 4b). Beans grown in Morogoro (E10) and Cedara and Potch, South Africa (E11 and E12), exhibited longer cooking times than beans grown in Entrican, MI (E3), Othello, WA (E4–E7), Mbeya (E8), and Arusha (E9). One interesting difference between the presoaked and the unsoaked was that relative to Morogoro (E10), Cedara, South Africa (E11), exhibited elevated unsoaked cooking times more so than the presoaked beans.

### 3.5 | Genotype × Environment

Knowledge of Genotype × Environment interactions can help to develop effective breeding strategies that use resources efficiently. With that goal, the cooking time data were projected onto GGE biplots to aid in interpretation of GGE interactions (Figure 5). For the presoaked cooking time, approximately 84% of the total GGE variation was explained in the biplot, and PC1 explained 72.8% and PC2 explained 10.8% of the observed variation (Figure 5a). The cooking times of individual genotypes generally maintained their rank across environments, and Figure 5a shows that the fastest cooking genotypes (G1, G13, and G4) were the fastest in nearly all environments. The presence of all environments on one side of the biplot indicates a lack of crossover G × E. Although any of these environments would perform well for cooking time breeding and selection, the environments

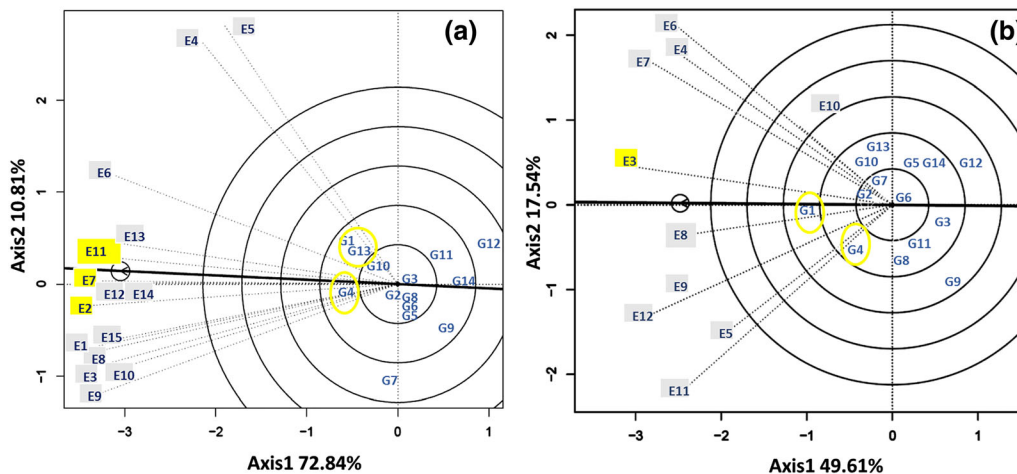


**FIGURE 4** Boxplots of production environments. (a) Presoaked cooking time environment boxplot: cooking times (presoaked) with environment as the main effect. Boxplot depicts 15 environments across 14 dry bean genotypes. (b) Unsoaked cooking time environment boxplot: cooking times (unsoaked) with environment as the main effect. Boxplot depicts 10 environments across 14 dry bean genotypes. There was an unsoaked cooking time outlier of 381 min for G11 grown in E10 that is not shown. Information on individual environments is available in Table 2

with the longest vector have the strongest discrimination potential for differentiating genotypes (Yan & Kang, 2002). The ideal environments for selection are those in proximity to the open circle (E7, E11, and E2).

For unsoaked cooking time, approximately 67% of the total GGE variation was explained by the biplot,

and PC1 explained 49.6% and PC2 explained 17.5% of the variation (Figure 5b). The fastest cooking genotypes were G1 and G4 in most environments, and E3 was the ideal environment for selection of cooking time because it had the longest vector and was in proximity to the open circle.



**FIGURE 5** Production environment influence of cooking time: Genotype and Genotype  $\times$  Environment (GGE) biplot for (a) the presoaked cooking time of 14 dry bean genotypes across 15 environments with column metric preserving, tester centered  $G + GE$ , scaled by standard deviation. (b) The unsoaked cooking time of 14 dry bean genotypes across 10 environments with column metric preserving, tester centered  $G + GE$ , scaled by standard deviation. The fastest cooking genotypes are circled in yellow, and the optimal selection environments are highlighted in yellow. The GGE biplot figures can be interpreted as follows: Similarity between individual genotypes, and individual environments, is estimated by separation across each axis. In panel (a), genotypes that are farther apart (i.e., G1 and G12) are the most different from each other for cooking time. Generally, the genotypes in proximity to an environment on the biplot are fastest cooking in that particular environment. The environments with the longest vectors have the strongest discrimination potential for differentiating genotypes (Yan & Kang, 2002). The ideal environments for selection of fast cooking genotypes are those in proximity to the open circle

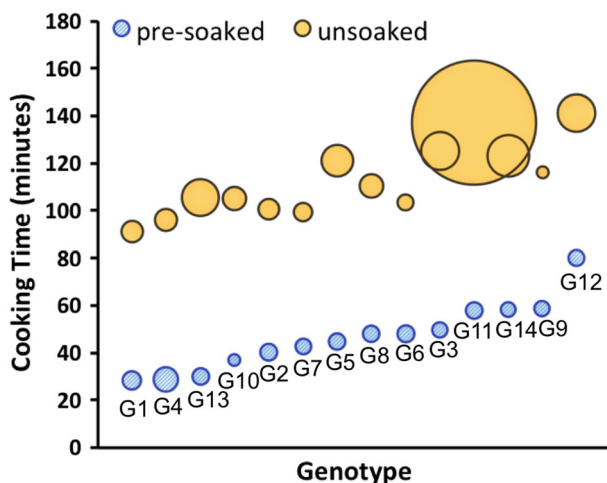


### 3.6 | Phenotypic stability

The phenotypic stability for cooking time was calculated as the CV for a genotype across all environments. The circle size for each genotype depicted in Figure 6 is proportional to CV. In general, cooking time was stable across environments for presoaked beans. G11 (Vazon 7) had the least stability for the unsoaked cooking time. These data include the long cooking time in E10, which reduced the stability of this genotype (Figure 6).

### 3.7 | Phenotypic correlations

Cooking time is only one trait among many important for a finished variety. Acceptable agronomic characteristics and seed yield are essential for any breeding program. To understand their possible interactions with cooking time, agronomic characteristics including days to flower, maturity, seed yield, and seed weight were measured for each genotype and environment (Table S2). Each of these characteristics was negatively correlated with cooking time (Table 4). Those with the largest correlations to cooking time were days to flower ( $r = -.475$ ,  $p < .0001$ ) and days to maturity ( $r = -.435$ ,  $p < .0001$ ), which can be seen as unfavorable correlations because early maturing varieties are often preferred by farmers. The presoaked and unsoaked cooking times were positively correlated ( $r = .638$ ,  $p < .0001$ ).



**FIGURE 6** Mean cooking time and phenotypic stability of 14 dry bean genotypes across 15 environments (presoaked) or 10 environments (unsoaked). Genotypes are sorted from fastest to slowest presoaked cooking times. The bubble size is based on the percent coefficient of variation of the mean across all environments, and a smaller bubble size indicates a smaller percent coefficient of variation and in turn a more stable cooking time across environments. The stability comparisons are relative to one another, as specific thresholds have not yet been established

## 4 | DISCUSSION

Genotypic variability for cooking time was observed both within and among market classes. Within market class variability can be especially useful to make breeding gains in cooking time while maintaining the seed characteristics expected by consumers. Genotypic variability among market classes can serve as a catalyst for breeders, farmers, and processors to differentiate seed types based on unique cooking attributes.

A handful of especially fast cooking landraces from Africa have previously been identified (Cichy, Wiesinger, & Mendoza, 2015). There is a need to transfer this fast cooking trait to higher yielding, photoperiod-insensitive varieties. The high heritability estimates measured in this study suggest that the phenotypic differences observed for the presoaked cooking time between genotypes are highly controlled by genetic factors and that phenotypic selection will be an effective means to develop fast cooking bean lines (Dudley & Moll, 1969). The lower heritability values for the unsoaked cooking time suggest that this trait is more sensitive to other nongenetic factors. One of the causes of the higher cooking time variability of the unsoaked beans was the appearance of hardshell in one of the genotypes resulting in a prolonged cooking time. Hardshell is a defect of the seed coat of some bean germplasm typically exhibited when the plants are water stressed during seed filling and when seeds are not stored in optimal conditions. Hardshell is exhibited by seeds not imbibing sufficient water during soaking and/or cooking (Stanley, 1992a). Hardshell can be managed and often reversed after harvest by maintaining sufficient moisture during seed storage (Castellanos, Guzmán-Maldonado, Acosta-Gallegos, & Kelly, 1995). Hardshell is easy to detect

**TABLE 4** Spearman correlation and  $p$  values between the presoaked cooking time (actual mean values) and agronomic, seed, and the unsoaked cooking time characteristics<sup>a</sup>

Trait	Presoaked cooking time		
	$r$	$p$ value	$N^a$
Days to flower	-.475	<0.0001	350
Days to maturity	-.435	<0.0001	350
Seed yield	-.212	<0.0001	416
Raw seed weight <sup>b</sup>	-.153	<0.0001	433
Unsoaked cooking time	.638	<0.0001	259

<sup>a</sup> $N$  is the number of samples used in the correlations. Individual replications were included separately.

<sup>b</sup>The weight of 100 seeds at approximately 10% to 14% moisture.

during the soaking process, and consumers can easily pick out hardshell beans after soaking. When cooking unsoaked beans, however, it is not possible to identify and remove hardshell prior to cooking. This could be a potential reason for the high variability in cooking times among the unsoaked beans in this study.

Beans grown in certain production environments had longer cooking times than in other environments. The tropical environments of Morogoro and Juana Diaz and the Mediterranean climate of Cedara appeared to induce the longest cooking times. When beans are stored in tropical conditions (e.g., high temperature and high humidity), they develop prolonged cooking times, and the cotyledons are difficult to soften during cooking (Stanley, 1992a). Our data suggest that growing beans under these conditions also induce longer cooking times. Future research is needed to clearly separate the effect of the growing and storage environment. That was not possible in this study because of the inability to account for environmental conditions immediately following harvest and during shipping to Michigan for cooking analyses. There is a need for future dry bean variety development to identify fast cooking genotypes suited to heat stress, tropical production and storage conditions, and changing climates.

Crossover Genotype  $\times$  Environment interactions for cooking time were minimal.  $G \times E$  for cooking time (such as observed in the cranberry market class) suggest multiple genetic mechanisms at work, and some are expressed differently in one environment than another. For example, when beans are stored under high temperature and humidity, tannins in their seed coat leach into the cotyledon and cause lignification of the cell wall, thereby increasing cooking times (Del Valle & Stanley, 1995; Stanley, 1992b). Beans with dark seed coats have higher tannin concentrations than beans with light colored seed coats and are more susceptible to high temperature and humidity storage conditions (Del Valle & Stanley, 1995; Stanley, 1992b). Therefore, looking at multiple genotypes within a seed color/market class is useful to understand the  $G \times E$  response.

Soaking beans prior to cooking reduces the cooking time as compared with cooking unsoaked beans. The soaking process softens beans and activates cell wall enzymes (Martínez-Manrique et al., 2011; Miano & Augusto, 2018). Despite the time-saving benefits of presoaking beans, many cultures and people prefer to cook unsoaked beans, mainly due to flavor differences (Borchgrevink, 2013; Castellanos et al., 1995). There are clear time-saving opportunities to use presoaked beans for genetic selection of fast cooking germplasm. A significant correlation ( $r = .638$ ) between the

presoaked and unsoaked cooking times was found, suggesting that it may be sufficient to select fast cooking lines based on the presoaked cooking times. The two genotypes, G1 and G4, were the fastest cooking whether presoaked or not. It was clear, however, that some genotypes that cook fast when presoaked do not cook fast unsoaked. An example of this is G13 (TZ-37). This finding suggests that there are additional genetic mechanisms at play, such as enzyme activation during hydration that can be further explored (Martínez-Manrique et al., 2011).

The cooking work presented here was conducted with distilled water, whereas in real-world scenarios, people often cook with hard water, which is rich in calcium and magnesium. Hard water will cause prolonged cooking times (Liu & Bourne, 1995), and future studies should be conducted to determine how fast cooking varieties will fare when cooked in hard water. As beans age, cooking time is prolonged, so it will also be important to conduct shelf-life studies with fast cooking bean varieties to characterize their stability over time.

The development of dry bean cultivars with fast cooking times that are stable and predictable across a wide range of production environments will have great appeal to processors and consumers. Fast cooking cultivars could be marketed as dry pack, thereby appealing to many consumers who prefer the taste of home cooked dry beans to canned beans and to consumers who do not have access to canned beans (Winham, Tisue, Palmer, Cichy, & Shelley, 2019). Fast cooking cultivars may also appeal to canners looking to increase processing efficiency. With the growing human population and changing climate, scientists are advocating for diets that are both nutritious and environmentally sustainable (Foyer et al., 2016; Willett et al., 2019). There is evidence that beans with shorter cooking times retain more nutrients during the cooking process and have more bioavailable iron than beans that take longer to cook (Wiesinger et al., 2016; Wiesinger, Cichy, Tako, & Glahn, 2018).

Given the sufficient genetic diversity found, limited crossover Genotype  $\times$  Environment interactions, and high heritability for cooking time, this study shows that it is feasible to develop fast cooking dry bean varieties without the need for extensive testing across environments. In the case of cooking time, it is important to offer consumers a standard value (phenotypic stability) for meal preparation. Because dry beans are a minimally processed agriculture product that is directly exposed to the environment, standardization of cooking quality has been difficult to achieve. However, the data presented in this study indicate that some genotypes exhibit stable cooking times across production



environments. Selecting high phenotypic stability is a key component for a breeding program to introduce consumer-focused traits, such as fast cooking dry bean varieties.

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