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DEVELOPMENT OF COMMON BEAN GENOTYPES WITH HIGH IRON AND ZINC SEED CONCENTRATIONS AND SUPERIOR CANNING AND AGRONOMIC QUALITY TRAITS

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ABSTRACT

Iron deficiency anemia is prevalent worldwide but mainly affects children under five years of age and women of reproductive age. One of the main causes of anemia in these groups is diet incapable of meeting daily iron requirements. Biofortification of staple foods is an approach aimed at contributing to reduction of anemia in Africa, and common bean (*Phaseolus vulgaris* L.), one of the leading staple foods in East and Central Africa, has gained attention as a valuable source of iron (Fe) and zinc (Zn). Its usefulness in improving iron status of iron anemic women and children is documented. Natural variation in iron and associated micronutrients like zinc exists in beans but their concentrations are below the target levels to meet daily requirements. This study aimed to develop and identify potential bean genotypes that surpass the HarvestPlus threshold of 90 ppm seed iron for possible promotion as high iron and zinc beans, and utilization in hybridization programs targeting these minerals, productivity and market traits. Advanced 578 genotypes were evaluated in five genotype groups planted in three locations from 2016 to 2018. Genotypes significantly differed ($P \leq 0.05$) in Fe, Zn, cooking time, canning quality and yield. Iron and Zn varied highly, ranging between 44-118 and 25-50 ppm, respectively, across the five-genotype groups. Cooking time ranged from 29-118 minutes and majority of the genotypes expressed good to excellent canning quality based on visual assessment (4-5) and hydration coefficients (2.1-2.2). Mean yields for bush beans and climbers were 1674-1977 kg ha^{-1} and 2204-3160 kg ha^{-1}, respectively. The most outstanding genotypes that combined above average yield with high Fe/Zn were CMKN1109 (96/ 43 ppm), SMR103 (92/ 43 ppm), SMC12 (90/ 43 ppm), and NUS16 (91/ 48 ppm). In addition, NUA127 (84/ 42 ppm), SMR53 (84/ 42 ppm), SMC160 (84/ 43 ppm) and NUA595 (83/ 42 ppm) yielded above average and expressed high canning quality. The genotypes that combined high Fe/ Zn, canning quality, and yield are potential genotypes for further improvement or evaluation for possible release.

Key words: Common bean, biofortification, iron, zinc, canning, cooking time

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INTRODUCTION

Anemia affects all age groups worldwide, with more pronounced effects in children under five years, and women of reproductive age, causing major implications on health and capabilities [1]. Such effects include reduced fetal brain maturation, pediatric cognitive defects, maternal depression, lowered immunity and tiredness [2, 3]. The common underlying causes of anemia are nutritional deficiencies, diseases or genetic haemoglobin disorders [3]. In 2011, children in Africa represented the highest proportion (62.3%) of individuals with anemia but the greatest number of children (96.7 million) and women of reproductive age (202.0 million) were in the South-East Asia [4]. Iron (Fe) deficiency anemia, a condition in which the body has a low level of Fe in the bloodstream to meet Fe needs, is commonly attributed to menstruation, pregnancy, poor diet and/ or poor absorption of Fe due to medication or lack of vitamin C, folic acid and vitamin B12 [5].

Solutions for Fe deficiency anemia should address poor dietary intake of haematopoietic nutrients such as Fe, and infant feeding practices combined with basic causes of anemia including disease control especially malaria and intestinal helminths [3, 6]. The use of diverse micronutrient-rich diets is effective in treating Fe deficiency anemia. However, many people in developing countries may not afford a diverse nutritious diet, or possess limited knowledge of nutrition [7]. In addition, behavioral change towards consumption of diverse foods to improve dietary quality may take a long time due to traditional attachments to certain foods [8]. Biofortification, the process of improving the nutritional quality of frequently consumed crops, provides a more feasible and sustainable option among such groups of people [7, 9].

Common bean (*Phaseolus vulgaris* L.) is an advantageous legume for biofortification of Fe because the baseline grain Fe concentration is high at 55 ppm with variability of up to 110 ppm [10]. The crop feeds over 400 million people in Africa (International Center for Tropical Agriculture [11]). It is very popular because it is affordable, palatable and has a long shelf life that makes it an insurance food. Beans are also a healthy food option reported to reduce development of heart disease, and breast /colon cancer [12, 13]. In addition to Fe, the crop provides quality protein (20-28%), energy (32%), fibre (56%) and micronutrients, especially zinc (33 ppm) and vitamin A, which enhance normal body and mental growth and development. Hence, it has the potential to alleviate malnutrition and hunger related problems. The attention of biofortification in beans has been on seed Fe concentration and bioavailability [10]. Recent studies report positive effects of consuming biofortified beans on university women's hemoglobin levels, total body Fe, physical activity and cognitive performance [14, 15, 16, 17].

The daily requirement for Fe varies from 7-8 mg/day in children aged 1-4 years, 12-28mg/day in adults, and 30-38mg/day in pregnant or lactating women [18, 19]. Beans with the full target level of 94 ppm of Fe is estimated to provide 127% and 80% of daily estimated average requirements of children and women, respectively [20]. A threshold for high Fe beans set by HarvestPlus is 90 ppm [21]. However, majority of the popular beans in East Africa still fall below the target of 94 ppm [20, 22]. More

recently, released varieties in Rwanda, Uganda and Congo were above the HarvestPlus threshold in seed Fe concentration and several breeding lines are in the pipeline [20, 23].

Conventional and modern breeding methods have been utilized in biofortification of beans [24]. A meta-QTL analysis across seven studies discovered eight QTL associated with combined high Fe and zinc (Zn) that could be relevant in marker-assisted breeding of Fe and Zn concurrently [25]. Improvement of Fe is also achievable without the general negative effect on yield and its concentration is unlikely influenced by water stress [26, 27, 28, 29]. Nonetheless, previous studies reported significant influence of environment on seed Fe and Zn concentrations but understanding how this interaction influences genotype ranks in different environments is what is important in making decisions [30, 31]. Bean genotypes that maintain relatively high micronutrient levels in comparison to others in varying environmental conditions are generally preferred.

Traits such as long cooking time and susceptibility to common diseases like anthracnose (*Colletotrichum lindemuthianum*), angular leaf spot (*Pseudocercospora griseola*), and bean common mosaic virus and its necrotic strain, limit bean utilization. Cooking soaked or unsoaked regularly consumed beans in Africa takes 1 to 3 hours, requiring more fuel and water [32]. These inconveniences reduce *per capita* consumption of beans thereby limiting the health benefits especially of Fe to the anemia prone children and women. Strong negative correlations of cooking time and bioavailable Fe in yellow beans cooked when unsoaked [r = -0.76] or soaked, [r = -0.65] indicated that Fe becomes less available with prolonged cooking [33]. High genetic diversity for cooking time occurs among bean germplasm for crop improvement, and several studies have identified genotypes that cook in less than 30 minutes [33, 34, 35, 36]. Six significant QTLs on chromosomes Pv01 and Pv09 were detected in two environments from evaluation of 140 F2:4 families for cooking time, and a few others on chromosomes Pv02, Pv03, and Pv06 from 206 accessions of the Andean beans [34, 37]. Narrow sense heritability of 0.53 and 0.74 for the F2:5 families evaluated at the two different locations, and 0.74 for a RIL population at F6, F7 and F8 indicated that it was easy to select for the trait during breeding [34, 38].

There is also a rising market for precooked beans in Africa [39] that require phenotyping for other quality traits in addition to short cooking time. In Kenya and Uganda, the potential market share for precooked beans was projected at 44% of bean consumers [32]. Canned beans are one of the precooked products that require additional quality traits such as high can yield, ability to retain colour, clear brine, and minimal bean splitting and starch/clumps [40]. Factors like genetics, environment, genotype by environment interactions, seed handling after harvest, and the processing method influence canning quality [41, 42]. The identification of QTL for quality traits to facilitate near future marker-assisted breeding is in progress [43, 44]. A yield drag is also unlikely in breeding for canning quality [41]. This study sought to develop and identify potential genotypes that surpass the HarvestPlus threshold of 90 ppm seed iron, with superior agronomic and canning quality, for possible promotion as high iron and zinc beans, and utilization in hybridization programs targeting high iron and zinc, productivity and market traits.

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MATERIALS AND METHODS

Trial site characteristics

Trials were established at Kawanda located at 32° 31'E, 0°25′ N with an altitude of 1,190 m above sea level (asl), in central Uganda, Kachwekano located at 1°15′ S, 29°57′ E at an elevation of 2,200 m asl in southwestern Uganda and at Kitengule prison farm in Karagwe. Kitengule is located at 2°08' S, 33°26' E at an elevation of 1,320 m asl in western Tanzania. Facilities at the International Centre for Tropical Agriculture (CIAT), Kawanda were utilized for cooking time and canning quality analysis. The three locations have a bimodal rainfall pattern represented by "a" and "b" for the first (March-June) and second (September-December) rainy season.

Genetic materials assessed

Large and small seeded bean genotypes of different seed colours (Fig. 1) were grouped into five sets based on breeding history, target agro-ecologies and growth habits (Table 2). Set1 (CMKN) consisted of 240 genotypes targeting agro-ecologies of climbing beans, combining micronutrient density with traits relevant for variety adoption, that were developed from single crosses of three iron dense parents; MAC42, Gitanga and NGWINxCAB2 selected from previous evaluations at CIAT [22]. The other sets were developed from multiple parent crosses of diverse backgrounds. Set2 (NUV, NUC, MNC) were 54 climbing beans selected from previous trials. Set3 (MIB, SMC, SMB, SMN, SMR, DAN) were 144 bush beans biofortified to target drought prone areas from which 75 genotypes were selected based on Fe, Zn and yield. Set4 (NUA, NUAK) were 108 nutritionally enhanced large-seeded bush beans (NUA) selected from previous yield trials, and Set5 (NUS) were 36 medium and large seeded bush beans (Table 2).

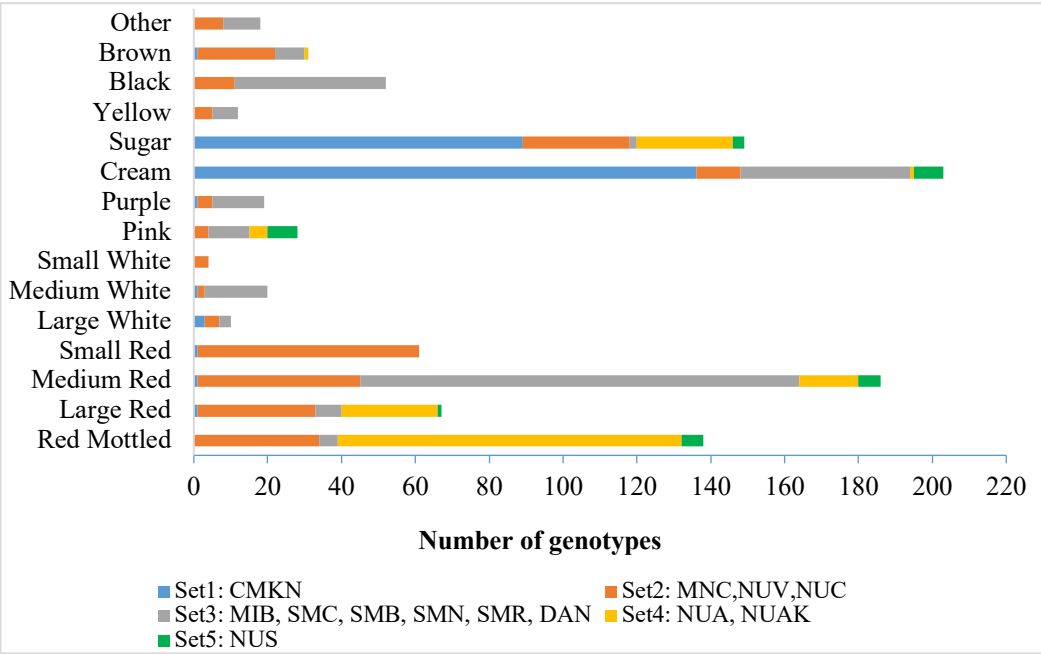

Figure 1: Market class groups for the evaluated genotypes

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Trial establishment and management

The climbing and bush bean checks included in the trials were high (RWV1129, RWR2245) and low (Decelaya, DOR500) iron beans, and high yield (Vunihing, CAL96) varieties [22]. The canning (NABE12C, Awashmelka) and cooking time (Awashmelka) checks were selected from previous evaluations at CIAT. Trials were replicated twice in an alpha lattice design. Plots within a replication were of 3 rows by 3 m in length; row and plant spacing were 50 cm and 10 cm for bush, and 60 and 10 cm for climbers. Each trial was weeded twice and an insecticide (Dimethoate) and two fungicides (Mancozeb and Ridomil) applied using the manufacturers' rates. Granular NPK 17:17:17 fertilizer was hand applied just before planting at the rate of 125 kg ha^{-1}.

Data collection

Agronomic performance

Diseases including angular leaf spot, common bacterial blight, bean common mosaic virus, rust and aschochyta blight on leaves were assessed on a 1 to 9 scale [45]. Days to flowering (DF) and physiological maturity (DPM) were recorded as the number of days from planting to when 50% of plants had at least one flower, and when the first pods began to discolour, respectively [45]. Harvested seeds were sun dried to ≤ 13% moisture content before recording plot seed weight (g).

Iron and Zinc evaluation

Samples were analyzed using the Oxford instruments X-Supreme 8000 energy dispersive X-ray Fluorescence (XRF) model in Rwanda, at Rubona Agriculture Research Station. Properly filled 10-15 pods hanging above the soil were randomly sampled per plot, and placed in new clean envelops, before the main harvest, hand threshed, wiped with distilled water to remove any soil contamination prior to packing 100 g in new paper bags. The seed samples were prepared and analyzed as described by Mukamuhirwa *et al*. [46].

Cooking time assessment

Randomly sampled, less than three-month old seeds free of damage and of moisture contents 10-13%, were soaked in distilled water at room temperature (22 ± 2 ◦C) for 12 hours, drained and kept in sealed bottles [37, 47]. Seeds per genotype were positioned into each of the 25 holes of the Matson cooker so that the piercing tip of the 90 g rod was in contact with the surface of the bean prior to placing in a five-litter beaker containing boiling distilled water [47]. The optimum cooking time was defined as the time required for 80% of the plungers to penetrate the seeds [47].

Canning quality assessment

The protocol involved cold and hot soaking of bean samples, brine preparation, autoclaving, storage and evaluation for consumer traits on freshly harvested and damage free seeds [40]. Moisture content (% MC) for each sample was obtained using a SINAR Model 6095 AgriPro moisture analyzer, and the dry bean weight (DBW) for

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canning, which is the fresh weight of beans equivalent to 90 g of total solids at a given MC calculated as $\frac{90\ g\ (i.e. solids\ required)}{1-(\frac{MC\,\%}{100})\ (i.e. MC = moisture\ content)}$. During the canning process, the soaked bean weight (g), which is the measure of both the weight of water and total solids in the sample, was recorded after cold and hot soak. Hydration coefficient (HC) $= \frac{Weight\ of\ soaked\ beans\ (g)}{Dry\ bean\ weight\ \ (g)}$. Canned beans were stored in boxes at room temperatures for two weeks, and then visually assessed for colour, appearance, brine clarity, bean splitting, and free starch/clumps using a 7 point scale, where; 1=Unacceptable, 2=Very bad, 3=Bad, 4=Fair, 5=Good, 6=Very good and 7=Excellent. A 'can' score for each sample was obtained by averaging values for all the above-mentioned visual traits from five evaluators prior to data analysis.

Data analysis

Data were analyzed separately in GenStat for windows 20th Edition [48] to assess within trial variability before performing combined analysis of variance (ANOVA). Each season was considered as an environment for the combined analysis. The linear model for individual and across environments were $Y_{ijk} = GM+R_i+B_j+G_k+e_{ijk}$, and $Y_{ijk} = GM+R_i+E_j+G_k+GE_{jk}+e_{ijk}$. Y_{ijk} described the observed value, GM the Grand Mean, R_i the Replication effect, B_j the Block effect, G_k the Genotype effect, R_i/E_j the effect of replications nested within environment, E_j the environment effect, GE_{jk} the Genotype x Environment effect and e_{ijk} the error [49]. Replications, blocks, and environments were random factors while genotypes were fixed. The null hypothesis (H_o) was: no differences existed among genotypes. Broad sense heritability was calculated as: $\frac{VC_G}{VC_G+VC_{e/r}}$ for mean within environment, $\frac{VC_G}{VC_G+VC_{GE}+VC_{e/r}}$ for mean within environment, including GE effect and $\frac{VC_G}{VC_G+VC_{GE/E}+VC_{e/rE}}$ for mean across environments. Yield stability by joint regression analysis characterized the sensitivity of each genotype to environmental effects by fitting a regression of the environment means for each genotype on the average environment means [50]. Low sensitivity values represent more stable genotypes with respect to changes of environment [50]. Correlations between traits were analyzed using means from across environments.

RESULTS AND DISCUSSION

Soil analysis

There was sufficient iron concentration in the soil for beans but zinc was limited at all sites (Table 1). The pH at Kawanda was the lowest and below the critical value of 5.2 whereas that of Kitengule was the highest ranging from 5.9 to 6.1. Nitrogen (N) was within acceptable levels but Potassium (K) and Phosphorus (P) were generally limited especially at Kawanda.

Broad sense heritability (H^2)

The total variations due to genetic factors among the fixed genotypes developed from different crosses were estimated based on genotype means across environments. The broad sense heritability (H^2) ranged from 0.12-0.55 (YDHA), 0.37-0.74 (Fe), 0.36-0.51 (Zn), 0.50-0.75 (HC) and 0.58-0.68 (can score) in the five sets (Table 3). This showed

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that the larger part of the variation was genetic for Fe (except in set4), canning quality and Zn for all sets. Thus, selection of genotypes for hybridization purpose or further evaluation for possible release was effective to realize moderate genetic gain for these traits. However, emphasis was on selection of genotypes that were relatively stable across environments. The variations observed in yield were majorly due to the differences in microenvironments in genotype set1, set3 and set4 (Table 3). Hence, selection in one environment would not produce much gain in different environments. More efficient yield selection was possible in genotype sets 2, 3_selected and 4, but testing in more replications or seasons could have improved selection efficiency. The H^2 for cooking time showed that selection would be ineffective across environments. From individual environments, only set3 could be selected effectively (H^2= 0.83) using the environment means (Fig. 2).

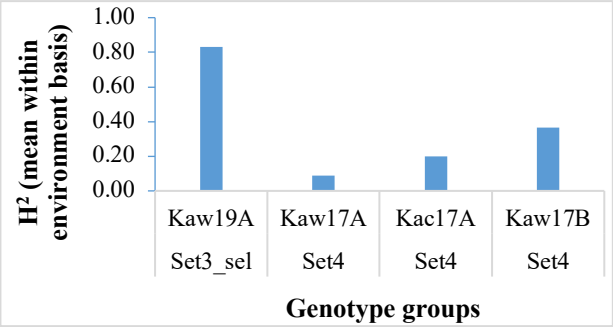

Figure 2: Broad sense heritability (H^2) for cooking time based on means within environment. Kaw= Kawanda, Kac = Kachwekano, 17a, 17b, 19a= 2017 or 2019 season "a" or "b"

Genetic estimates like broad sense heritability apply strictly to the analyzed populations, the environmental conditions and the estimation method [51]. The estimates in this study provided useful information on whether the differences observed among genotypes were due to genetic composition or microenvironments. Due to high variation from extraneous factors that interacted with the genotypes (GE), the inherent attributes for cooking time and yield in some sets were minimized. The H^2's in this study were classified as low (≤0.30), moderate (0.31-0.60) and high (>0.60) [52]. Higher H^2 for cooking time (0.70) and yield (0.69) from on farm multi-location trials in Uganda were recently reported with a recommendation for effective selection for both traits, and evaluation for cooking time in few environments since GE interaction was not important [35]. In this study, cooking time was not widely evaluated to make effective conclusions on the relevance of GE but set4 recorded large and significant GE. The H^2 of 0.70 and 0.86 for Fe and Zn were reported in a study carried out in two locations in Uganda [53]. While similar H^2 for Fe was obtained in this study, all the H^2 for Zn were moderate due to relatively higher magnitudes of GE. Insufficient zinc concentration in the soils (Table 1) could have been a contributing factor.

Iron (Fe) and zinc (Zn) seed concentration

Significant differences (P≤0.001) existed among genotypes for both Fe and Zn in all sets except for Zn in set1 (Table 4). Genotype x environment interaction for Fe and Zn were also significant (P≤0.001) in all sets except in set 5 and set1 for Zn (Table 4). Iron

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and Zn seed concentrations varied highly among the genotypes within environments ranging between 44-118 and 25-50 ppm, respectively, across the five-genotype groups. Thirty and 12 genotypes like CMKN1109 (96 ppm of Fe, 43 ppm of Zn), SMB15 (94, 44 ppm), SMN57 (94, 47 ppm) accumulated significantly different levels of Fe and Zn from the high iron checks (Table 5). Despite the fluctuations in the actual levels of these minerals in different environments, superior and stable genotypes maintained consistent performance above the high checks. Genotypes were grouped according to the levels of Fe and Zn and out of 578 genotypes, 12% and 6% accumulated 5 and 10 ppm, respectively, higher iron than the concentrations in high check genotype (Fig. 3). The Zn levels for only 2% of the genotypes were 5 ppm higher than the high check. To achieve higher progressive gains, it is necessary to select new high checks in each breeding cycle. Ranked according to different levels, 75% and 61% of the genotypes accumulated 70-90 ppm of Fe and 35-40 ppm of Zn. The genotypes that surpassed these levels were 1% and 26% for Fe and Zn, respectively (Fig. 3).

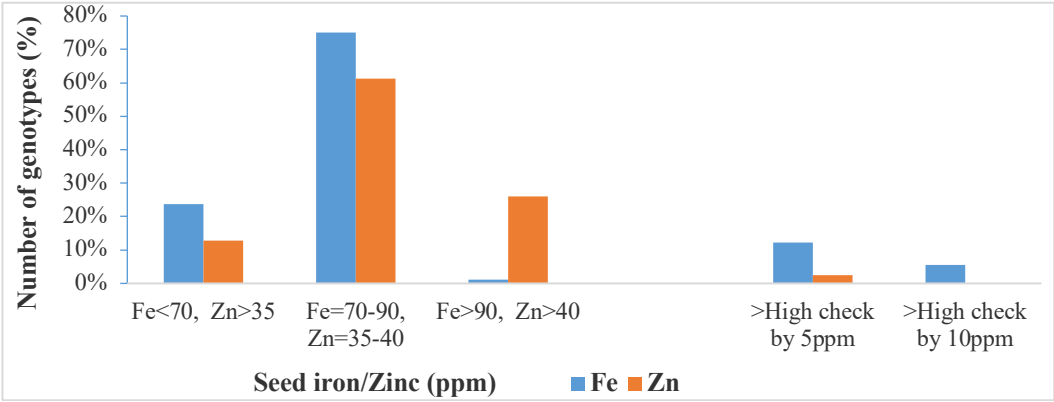

Figure 3: Number of genotypes (%) belonging to different levels of seed iron and zinc concentration

In consideration of the target threshold of 90 ppm of iron by HarvestPlus [21], the realized improvement was small based on mean performance across environments because only 1% of 578 genotypes surpassed the threshold. Significant influence of environment on seed Fe and Zn concentrations were reported [30, 31]. Thus, selection for superior and stable genotypes across environments are important for consistency. Evaluation of genotypes in more environments could have allowed grouping genotypes into target environments and possibly improved selection efficacy. The comparison of test materials with the check genotypes was useful in identifying more stable genotypes that performed consistently better than the high check despite the fluctuations in different environments. Recent studies report seed iron concentrations of above 100 ppm in genotypes like BCB11-145 (136 ppm), BF-08-13-181 (106 ppm), IBC2 (107 ppm) and NUA 66 (112 ppm) [23, 54], which could be useful parents for further improvement of the iron concentrations. Beans with 94 ppm of iron, provide 127% and 80% of daily estimated average requirements of children and women, respectively [20], and several of the evaluated genotypes that accumulated seed iron close to this level are potential genotypes for further evaluation for possibly adoption. Nonetheless, there is need to effectively predict genetic gains in early breeding to increase genetic variation and improve selection accuracies to realize the target gains.

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Canning quality of genotypes in set 2, set 3 and set 4

The observed genetic differences were adequate to make effective selection as shown by moderate to high broad sense heritability (Table 3). Genotypes significantly differed (P≤0.001) in hydration coefficient (HC) and visual canning quality (can score) in all the three sets except in HC in set 2 (Table 4). On average, beans increase in weight by 80% when fully hydrated, which converts to a HC of 1.8 [55]. The HC of 75-100% of the 210 genotypes in the evaluated sets were higher than 1.8, and 33-98% of the genotypes hydrated betted than the canning quality checks, NABE12C or Awashmelka (Fig. 4). A high HC implies heavy weight, which is attractive for the canning industry regarding profitability. Based on visual assessment for colour, appearance, brine clarity, bean splitting, and free starch/clumps, 11-28% of the 210 genotypes like NUC173, SMC14, NUAK291, and NUAK291 were very good (6) or excellent (7) (Fig. 4). Despite the various traits for assessing canning quality [56], consumers assess quality visually making these traits very useful criteria for routine breeding evaluation. Compared to the checks, only 0-3% of the genotypes from each set were visually better (Fig. 4). This showed the existence of nutrient dense genotypes with acceptable canning quality, which could be promoted for further evaluation or used for breeding.

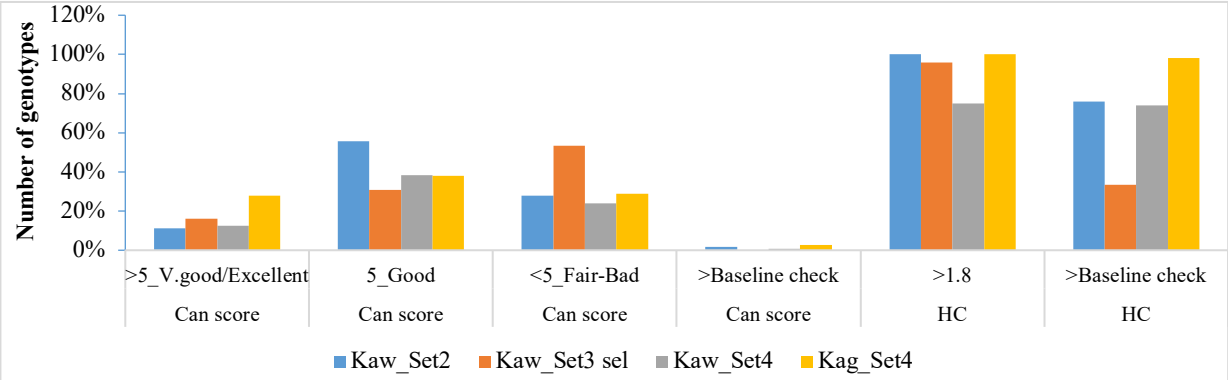

Figure 4: Classification of genotypes in set2, set3 sel and set4 for visual canning quality (can score) and hydration coefficient (HC)

Cooking time for set3 and set4

The variance component for genotype by environment interaction was large compared to that of the genotypes to make effective selection of stable genotypes in set4. Consequently, the cooking time of genotypes in set4 were not significantly different (P≤0.05) (Table 4), but a few genotypes like NUAK143 and NUA633 that had similar cooking times of 42-46 mins in locations (Fig. 5) are potential breeding materials. The mean performance of the genotypes in set4 were 72±20 and 50±10 mins for Kawanda and Kachwekano. Set 3 genotypes generally expressed short cooking time with a mean of 49±5 mins. The check, Awashmelka, was cooked in 41 mins, and nine genotypes had less cooking time (Fig. 5) although they were not significantly different (P≤0.05).

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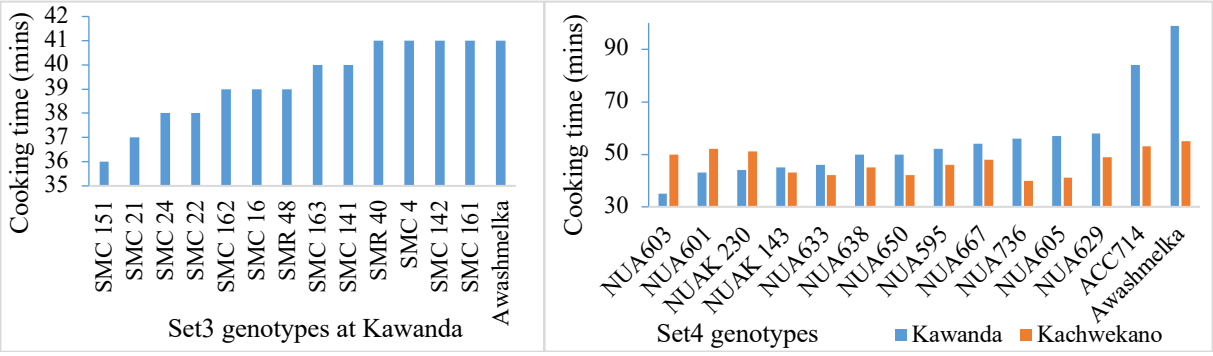

Figure 5: Cooking time of selected genotypes in set3 and set4
Range (set3=36-82, set4=25-118 at Kawanda, 27-84 at Kachwekano), *coefficient of variation* (set3=10%, set4=27%, 19%), *standard error of the mean* (set3=3.6, set4=11, 7) and *least significant difference* at P≤0.05 (set3=10)

Previous studies reported cooking time for Awashmelka as 35-41 mins showing that its cooking duration is stable [36, 54]. Several ranges including: 14-75, 19-271 and 31-130 mins for cooking time determined by Mattson cooker method exist in African germplasm [35, 36, 54]. The identified genotypes like Awash-1~14 mins, and Cebocela ~19 mins [35, 54] are useful breeding germplasm.

Yield performance and other traits
Genotypes, and genotype x environment interaction significantly differed (P≤0.05) in all sets except set 4 and set 2, respectively (Table 4). Yield sensitivity analysis showed that environments were significantly different (P≤0.001) but genotype sensitivities were only significant (P≤0.01) in set1 and set 3 (Table 6). This showed that the rank of genotypes based on yield in these environments significantly varied and selection of stable genotypes instead of the most superior in yield was a better option. Genotypes such as CMKN1139 (2918 kg ha^{-1}), CMKN1945 (3014 kg ha^{-1}), SMR125 (2128 kg ha^{-1}), ACC714 (2237 kg ha^{-1}), NUA647 (1461 kg ha^{-1}), NUS26 (2050 kg ha^{-1}) and RWR2245 (2324 kg ha^{-1}) combined stability and yield superiority because they were among the 16 least sensitive genotypes and yielded above group average (Table 7). Such genotypes are expected to maintain high yield performance during further testing. The DF and DPM for bush/ climbing beans ranged from 45-65/ 49-64 and 67-104/ 97-114, respectively, at Kachwekano, 27-46/ 43-46 and 60-85/ 83-85 at Kawanda and 41-49/ 34-48 and 80-87/ 74-87 at Kitengule (Data not presented). Angular leaf spot (ALSF) and common bacterial light (CBBFL) were the major field diseases observed across all the five sets of germplasm (Data not presented). In set1, up to 89% and 23% of the germplasm expressed intermediate response to CBBFL and BCMV, respectively, in at least a season, and 4% were susceptible to CBB. Similarly, 70% and 80% of set2 germplasm expressed intermediate response of ALSF and CBBF, respectively. Susceptible disease response was also observed in 1% (ALSF) and 2% (BCMV) in set3, 2% (BCMV) in set 4, and 14% (ALSF), 8% (CBBFL) and 2% (rust) in set5. Intermediate response to ascochyta blight (ASCFL) was observed in 59% and 5% of set3 and set 4, respectively (Data not presented).

Relatively large GE for common bean yield was previously reported [35, 36]. When GE is large and consistent over seasons, it is important to define target environments for multi-location yield evaluations to make effective selection. In this study, evaluations were performed in utmost two seasons so it was not informative to group environments. The yields of recently released varieties in Tanzania and Uganda were >1500 kg ha^{-1} and 1500-2200 kg ha^{-1} for bush, and >2000 kg ha^{-1}, and 2500-3700 kg ha^{-1} for climbers, with DPM of 67-90 and 58-68 (bush), and 90-110 and 82-96 (climbers), respectively [57, 58]. The selected stable genotypes yielded within the range of these recently released varieties, which increases their value to farmers. Average yields across environments ranged from 932-2893 kg ha^{-1} for bush, and 853-4972 kg ha^{-1} for climbers showing better yielding, but environment sensitive genotypes existed in the study and could be useful.

Considering trait combinations, these bush and climbers yielded above average and cumulated high Fe/Zn; SMR103 (92 / 43 ppm), SMC12 (90/ 43 ppm), NUS16 (91/ 48 ppm) and NUS18 (87/ 41 ppm), CMKN1109 (96/ 43 ppm), CMKN898 (84/ 38 pm), NABE29C (83/ 38 ppm), MNC554 (88/ 44 ppm) and NUC76 (82/ 47 ppm). Similarly, these bush beans yielded above average, expressed good (5) or very-good (6) canning quality and accumulated high Fe/Zn; NUA127 (84/ 42 ppm), SMR53 (84 / 42 ppm), SMC160 (84/ 43 ppm), SMR128 (82/ 41 ppm) and NUA595 (83/ 42 ppm); NUA595 was also cooked in 49 mins. The response of genotypes to field diseases showed that majority of the genotypes possess broad resistance to intermediate response considering that only 0-4% of the genotypes expressed susceptibility to a disease. Thus, selection of genotypes with broad resistance in different environments during further phenotyping should be possible. The response of the selected genotypes to diseases were 1-4 (SMR103, NABE29C, MNC554, NUC76, SMR53, SMC160, CMKN1109), 1-7 (SMC12; 7 = BCMV), 2-6 (NUS16, NUS18 and CMKN898; 6=CBB), 1-6 (SMR128; 6 = ALSF) and 2-3 (NUA595). Genotypes like SMC12 and NUS16 are recommended for improvement for BCMV and CBB resistance, respectively.

Correlation of traits

Most of the trait associations were weak and not significant (Table 8). Although weak, a trend suggested that yield was negatively correlated to FESEED (r=-0.21 to r=0.04), ZNSEED (r=-0.14 to r=0.07) and canning quality (r=-0.28 to r=-0.19) (Table 8). The weak associations showed that selection for superiority in these traits and yield was possible. Generally, weak positive and negative association of yield to FESEED (r=-0.23, r=-0.39, r=0.25, r=0.40) [22, 26 27, 36], yield to ZNSEED (r=-0.30, r=0.20, r=0.35) [36, 27, 22],[22,27,36] and yield to cooking (r=-0.06, r=0.16) [36, 27] [27,36] have been reported by several authors. The studies showed that yield penalty is unlikely to occur during improvement of these traits. Significant (P≤0.001) and moderate positive correlation coefficients of r=0.46 to r=0.66 were recorded between Fe and Zn. This association was reported at a genetic level and could be very useful in marker-assisted breeding for both traits [25]. Modern breeding tools coupled with quality phenotypic data are essential to achieve higher gains.

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CONCLUSION

The study aimed to develop and select beans with >90 ppm of iron that possessed additional useful traits for farmers, processors and consumers, for promotion in the Pan African Bean Research Alliance for further evaluations and breeding. Genotypes that combined quality traits for canning, micronutrient density and yield are potential candidates. Those that accumulated iron levels above the threshold of 90 ppm are recommended for further improvement by parents that are significantly higher in iron. The genotype by environment interactions influenced the heritability estimates most especially for yield and cooking time, which showed the relevance for considering the important interactions in selecting best genotypes for use in many environments.

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Table 1: Soil analysis from soil and plant analytical laboratories at NARL-Kawanda

Site	season	pH	OM	N	P	Ca	Mg	K	Fe	Zn
			----%---		----------ppm----------					
Kawanda	2016a	4.9	4.4	0.2	4.3	1134.4	386.5	75.6	227.1	3.1
	2016a	4.8	4.0	0.2	3.1	973.5	362.9	79.4	256.1	2.6
Kachwekano	2016a	5.5	10.6	0.4	15.9	2033.9	581.9	232.3	223.9	1.8
	2016a	5.1	8.8	0.4	8.2	1191.6	483.3	300.1	208.4	1.2
Kitengule	2016b	6.1	7.0	-	15.0	1894.5	411.0	547.5	131.5	2.7
	2016b	5.9	8.0	-	23.0	2537.5	483.5	581.0	151.0	14.4
Critical values		5.2	3	0.2	5	350	100	150	-	-
Sufficient levels		5.2-7.0	6	0.3	20	2000	600	500	50	20

Table 2: Groups of genotypes evaluated for micronutrient concentration, canning quality, cooking time and yield

Sets of genotype	Growth habit	Number of genotypes	Trial location	Trial environment/ seasons	Analyzed Data
Set1: CMKN	Climber	240	Kawanda Kachwekano	2016a_Kawanda, Kachwekano 2016b_Kawanda, Kachwekano	Yield, Fe, Zn_4 sets
Set2: NUV, NUC, MNC	Climber	54	Kawanda Kitengule	2017b_Kawanda 2017b_Kitengule	Yield, Fe, Zn_2 sets Canning quality_1 set
Set3: MIB, SMC,SMB, SMN, SMR, DAN	Bush	144	Kawanda Kachwekano	2016a_Kawanda, Kachwekano 2016b_Kawanda, Kachwekano 2018b_Kawanda 2019a_Kawanda	Yield_6 sets Fe, Zn_4 sets Canning quality_2 sets Cooking time_1 set
Set4: NUA, NUAK	Bush	108	Kawanda Kachwekano Kitengule	2017a_Kawanda, Kachwekano 2017b_Kawanda, Kachwekano, Kitengule	Yield, Fe, Zn _5 sets Canning quality_2 sets Cooking time_3 sets
Set5: NUS	Bush	36	Kawanda, Kitengule	2017b_Kawanda, Kitengule 2018a_Kawanda, Kitengule	Yield_4 sets Fe, Zn _2 sets

Table 3: Broad sense heritability (H^2) on mean within environment, including GE effect and mean across environment basis

Genotype	YDHA		Iron		Zinc		HC		Can score	
	within	across	within	across	within	across	within	across	Within	across
Set1	0.08	0.21	0.41	0.74	0.16	0.44				
Set2	0.29	0.44	0.41	0.58	0.22	0.36				
Set3	0.04	0.15	0.30	0.63	0.21	0.51				
Set3_selected	0.38	0.55					0.33	0.50	0.41	0.58
Set4	0.03	0.12	0.10	0.37	0.15	0.46	0.60	0.75	0.52	0.68
Set5	0.12	0.35	0.37	0.54	0.28	0.43				

YDHA Yield estimated in kg ha^{-1}, *HC* Hydration coefficient, *Can score* Visual canning quality

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Table 4: Analysis of variance for across environment analysis for yield, iron, zinc, cooking time and canning quality

	Set1				Set2							
Change	d.f.	YDHA	Iron	Zinc	d.f.	YDHA	Iron	Zinc	Change	d.f.	HC	Can score
Env't	3	844541936**	43584.2**	2750.6*	1	120166194	4574.1*	4751.6**	Rep	1	0.0003	0.08
Rep/ Env't	4	31244084***	2564.0***	277.2***	2	12891340***	79.7	5.6	Block/rep	16	0.0073*	0.39***
Genotype	232	1909377*	236.6***	33.6***	53	1155424*	178.9***	36.7	Genotype	53	0.0048	0.77***
GenotypexEnv't	696	1562639***	62.5*	18.9**	53	641400	74.5*	23.6	Residual	37	0.0038	0.11
Residual	931	1072931	54.2	15.9	106	483838	48.5	16.9	Total	107	0.0048	0.48
Total	1863	2784795	155.6	24.2	215	1360313	108.4	45.4				
	Set3				Set3_selected							
Change	d.f.	YDHA	Iron	Zinc	d.f.	YDHA	HC	Can score	Change	d.f.	COOKT	
Env't	3	145793262**	11639.8**	673.8	1	5687965	1.319**	49.94**	Rep	1	67.0	
Rep/ Env't	4	5459268***	301.9	111.9***	2	2061406***	0.002	0.20	Block/rep	8	87.5**	
Genotype	143	640495	364.1***	47.3***	74	719782***	0.034**	4.30***	Genotype	74	156.3***	
GenotypexEnv't	429	544411**	135.1***	23.2***	74	326971**	0.017***	1.80***	Residual	66	26.3	
Residual	575	440115	94.8	15.1	149	191994	0.007	0.71	Total	149	94.4	
Total	1151	901326	174.3	24.2	299	387551	0.021	2.03				
	Set4				Set4				Set5			
Change	d.f.	YDHA	Iron	Zinc	d.f.	COOKT	HC	Can score	d.f.	YDHA	Iron	Zinc
Env't	4	18324547*	29579.4***	2662.8***	2 (1)	48011.7	0.020	0.93	3 (1)	22981651*	2693.8	1052.0**
Rep/ Env't	5	2374112***	239.9**	42.1**	3 (2)	17444.3***	0.007*	0.18	4 (2)	2283647***	1458.5***	5.4
Genotype	111	496972	173.6***	39.9***	111 (107)	310.6	0.012***	1.62***	35 (35)	525987*	139.2***	51.6**
GenotypexEnv't	444	436386***	110.0***	21.5***	222 (107)	358.3*	0.003***	0.51*	105 (35)	340949*	42.1	20.4
Residual	559	290187	64.0	12.7	335 (215)	285.1	0.002	0.36	143 (71)	252321	71.0	24.6
Total	1119	443523	199.65	28.6	671 (431)	533.4	0.005	0.71	287 (143)	586658	118.9	37.3

d.f. Degree of freedom, *Env't* Environment, *Rep* Replication, *,**,*** Significant at P≤0.05, P≤0.01, P≤0.001, *YDHA* Yield estimated in kg ha^{-1}, *HC* Hydration coefficient, *Can score* Visual canning quality, *d.f.* in parentheses are for *HC*, *Can Score* (set4) and Iron, Zinc (Set5)

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Table 5: Iron and zinc seed concentration of some selected genotypes that accumulated more iron or zinc than the high iron check

Set1	Fe ppm	Zn ppm	Set2	Fe ppm	Zn ppm	Set3	Fe ppm	Zn ppm	Set4	Fe ppm	Zn ppm	Set5	Fe ppm	Zn ppm
CMKN1109	96	43	NABE 29C	89	43	SMB 15	94	44	NUA595	83	42	NUS16	91	48
CMKN1484	88	36	MNC554	88	44	SMN57	94	47	NUAK512	82	34	NUS18	87	41
CMKN1551	85	38	NUV2	87	41	SMR103	92	43	NUA642	81	39	NUS6	83	42
CMKN 898	84	38	*RWV1129*	85	39	SMN 10	92	45	NUA607	81	38	*RWR2245*	82	40
NABE29C	83	38	NUC174	84	41	SMC 12	90	43	NUA641	81	41	NUS24	80	48
CMKN 948	82	39	NUC76	82	47	SMN 6	89	45	NUAK143	80	38	NUS17	80	41
CMKN2169	82	41	NUV134	80	44	SMC 4	89	46	NUAK 99	79	36	NUS27	80	43
CMKN 819	82	38	NUC176	79	43	SMR113	89	43	NUAK346	79	37	NUS31	79	41
RWV3006	81	42	NUV17	78	44	SMR102	89	44	*RWR2245*	79	40	NUS5	78	45
RWV1129	79	37	MNC370	78	42	SMR107	89	44	NUAK243	79	44	NUS33	76	40
MAC42	79	39	NUC428	77	40	SMC 7	89	40	NUAK364	77	41	NUS10	76	45
Gitanga	75	36	NUC173	73	48	SMN61	88	44	NUAK536	77	40	NUS13	74	40
NgwinxCAB2	62	34	NABE12C	73	40	*RWR2245*	74	41	NUAK359	75	40	NUS4	71	41
Yunihing	60	32	*DECELAYA*	71	38	CAL96	69	35	CAL96	73	36	CAL96	73	38
DECELAYA	57	34	Yunihing	66	41	*DOR500*	68	39	*DOR500*	65	36	*DOR500*	61	37
Minimum	57	32		53.7	33.5		57	33		61	27		61	31
Maximum	96	43		89.3	48.3		94	48		83	44		91	48
Mean	72	36		73.0	40.2		78	42		73	36		75	39
SD	7	4		7	4		10	4		8	4		8	5
CV (%)	10%	11%		10%	10%		10%	10%		11%	10%		11%	13%
SEM	2.8	1.5		4.3	2.4		4.1	1.7		3.3	1.5		3.2	2.3
LSD(0.05)	8	4		12	7		11	5		9	4		9	6
H^2_ac	0.74	0.44		0.58	0.36		0.63	0.51		0.37	0.46		0.54	0.43

SD Standard deviation of the mean, *CV (%)* Coefficient of variation, *SEM* Standard error of the mean, *LSD* Least significant different, H^2_*ac* Broad sense heritability for mean across environments

Table 6: ANOVA for yield stability using Finlay and Wilkinson's joint regression

Source	d.f.	Set1	d.f.	Set3	d.f.	Set4	d.f.	Set5
Genotypes	232	1071669***	142	327096*	111	255977	36	267385*
Environments	3	425862981***	3	73816425***	4	9340273***	3	11549147***
Sensitivities	232	1147870***	142	352624**	111	239804	36	227165
Residual	449	695830	281	248873	321	234833	68	145499
Total	916	2297980	568	682928	547	306717	143	435980

d.f. Degree of freedom, *,**,*** Significant at P≤0.05, P≤0.01, P≤0.001

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Table 7: Sorted sensitivity estimates for 16 least sensitive (most stable) genotypes in set1, set2, set4 and set5

Set1 genotypes	Sensitivity	Mean	Mean square deviation	Set3 genotypes	Sensitivity	Mean	Mean square deviation
CMKN1572	-0.1807	2017	5319550	SMB15	-0.1691	1367	366610
CMKN606	-0.0891	2074	360071	DAN10	-0.0335	1869	9109
CMKN793	-0.0509	2452	3466055	SMN13	0.0644	1402	163909
CMKN641	0.0932	2211	795161	SMR88	0.1347	1462	48418
CMKN1765	0.1156	1516	2620077	SMC101	0.2025	1260	80511
CMKN1250	0.1456	2150	272297	SMR125	0.2796	2128	276966
CMKN1320	0.1904	2126	597253	SMC146	0.4348	1773	655679
CMKN208	0.1971	1661	3216657	SMN57	0.4511	1576	21834
CMKN1606	0.2045	2152	2029734	SMB19	0.5119	1549	202306
Gitanga	0.2761	1376	1177784	SMC41	0.526	1204	53318
CMKN1609	0.2793	1726	7214	SMR101	0.5317	1702	368846
CMKN1139	0.2833	2918	410101	ACC714	0.5394	2237	48976
RWV3006	0.2921	2187	617486	SMC20	0.54	1520	171133
CMKN1560	0.3003	1857	560104	SMR115	0.5557	1601	14234
CMKN1945	0.3138	3014	1445095	SMN41	0.5573	1659	362346
CMKN960	0.3513	1593	4186	SMC152	0.5592	1250	17293
s.e.	0.3524	417			0.3985	249	
Mean		2497				1749	
Set4				Set5			
NUAK424	-1.496	1202	103744	NUS33	-0.1095	1357	159020
NUAK532	-0.8897	1099	4864	NUS11	0.0256	1378	60345
NUA678	-0.8315	1247	633905	NUS20	0.1888	1676	279473
NUAK512	-0.5707	1119		NUS4	0.3723	1733	189930
NUA661	-0.533	1167	152942	NUS2	0.3957	1696	105764
NUA645	-0.5294	1228	72913	NUS19	0.4649	1525	442754
NUA622	-0.4878	935	253884	NUS26	0.6157	2050	314252
NUAK480	-0.3427	983	336635	NUS1	0.7251	1419	200728
NUA720	-0.3254	1282	380227	NUS31	0.7364	1394	47377
NUA642	-0.3116	995	183870	NUS15	0.7497	1668	95054
NUA652	-0.2581	1129	351452	NUS18	0.8327	1673	31
NUA672	-0.2209	1364	99758	NUS25	0.8556	1551	101401
NUA647	-0.1633	1461	85108	NUS24	0.8627	1680	110688
NUAK145	-0.1511	1176	261682	NUS12	0.8641	1442	66677
NUA680	-0.0651	1227	293661	NUS6	0.8762	1724	65085
NUA702	-0.0557	1373	805185	RWR2245	0.9015	2324	132822
s.e.	0.8048	217			0.3834	191	
Mean		1381				1756	

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Table 8: Correlation coefficients of yield, iron, cooking time and hydration coeffients to other traits in each of the genotype sets

	YDHA	Iron	YDHA	Iron	YDHA	Iron	YDHA	HC	YDHA	Iron	Zinc	COOKT	HC	YDHA	Iron
	Set1		Set2		Set3		Set3_Sel		Set4					Set5	
YDHA	-		-		-		-		-					-	
Iron	0.04	-	-0.03	-	-0.17	-			-0.24*	-				-0.12	-
		0.54	0.07	0.46	-0.06	0.66			-0.14	0.62				-0.02	0.66
Zinc	0.01	***		***		***				***	-				***
COOKT									0.07	-0.12	-0.11	-			
HC							-0.19	-	-0.18	0.08	0.16	-0.21*	-		
Can score							-0.28 *	0.25 *	-0.19	0.18	0.09	-0.08	0.28 **		

YDHA Yield in kg ha^{-1}, *COOKT* Cooking time, *HC* Hydration coeficient, *Can score* Visual quality score, *, *** Significance at P≤0.05 and P≤0.001 for a two-sided test of correlations different from zero

REFERENCES

1. **Development Initiatives.** Global Nutrition Report: Shining a light to spur action on nutrition. Bristol, UK: 2018 Development Initiatives. https://globalnutritionreport.org/reports/global-nutrition-report-2018/executive-summary/ *Accessed 7th/02/2020*.

2. **Black MM, Quigg AM, Hurley KM and MR Pepper** Iron deficiency and iron-deficiency anemia in the first two years of life: Strategies to prevent loss of developmental potential. *Nutr Rev*. 2011; **69 Suppl 1**:S64-S70. https://doi.org/10.1111/j.1753-4887.2011.00435.x

3. **WHO (World Health Organization).** Nutritional Anemias: Tools for Effective Prevention and Control. Geneva: World Health Organization, 2017. [ISBN 978-92-4-151306-7; http://apps.who.int/iris/bitstream/handle/10665/259425/9789241513067-eng.pdf?sequence=1 *Accessed 18th/06/2020.*

4. **WHO (World Health Organization).** The global prevalence of anemia in 2011. Geneva: 2015. https://apps.who.int/iris/bitstream/handle/10665/177094/9789241564960_eng.pdf?sequence=1 *Accessed 11th/06/2020.*

5. **Harper JL, Marcel EC and CB Emmanuel** Iron deficiency anemia: Practice essentials, Pathophysiology and Etiology. Medscape. 2015. Retrieved from http://www.health24.com/Medical/Anaemia/Anaemia-20130216-2 *on 29th/07/2020*.

6. **Ruel MT and H Alderman** Maternal and Child Nutrition Study Group. Nutrition-sensitive interventions and programmes: how can they help to accelerate progress in improving maternal and child nutrition? *Lancet* 2013; **382(9891)**:536-51. https://doi.org/10.1016/S0140-6736(13)60843-0

7. **Welch RM** Breeding strategies for biofortified staple plant foods to reduce micronutrient malnutrition globally. *Journal of Nutrition* 2002; **132:**495S-499S.

8. **Girard AW and O Olude** Nutrition education and counselling provided during pregnancy: effects on maternal, neonatal and child health outcomes. *Paediatric and Perinatal Epidemiology* 2012; **26 Suppl 1**:191-204. https://doi.org/10.1111/j.1365-3016.2012.01278.x

9. **Bouis HE and A Saltzman** Improving nutrition through biofortification: a review of evidence from HarvestPlus, 2003 through 2016. *Global Food Security* 2017; **12**:49–58. https://doi.org/10.1016/j.gfs.2017.01.009

10. **Petry N, Boy E, Wirth J and R Hurrell** The Potential of the Common Bean (*Phaseolus vulgaris*) as a Vehicle for Iron Biofortification. *Nutrients* 2015; **7(2):**1144-1173.

11. **CIAT (Centro Internacional de Agricultura Tropical).** Beans: What we do-breeding better crops. (2018). https://ciat.cgiar.org/what-we-do/breeding-better-crops/beans/ *Accessed on 17th/07/2020.*

12. **Ganesan K and B Xu** Polyphenol-Rich Dry Common Beans (*Phaseolus vulgaris* L.) and Their Health Benefits. *International Journal of Molecular Science* 2017; **18(11)**:2331. https://doi.org/10.3390/ijms18112331

13. **Campos-Vega R, Oomah BD, Loarca-Piña G and HA Vergara-Castañeda** Common beans and their non-digestible fraction: Cancer inhibitory activity-an overview. *Foods* 2013; **2(3)**:374-392. https://doi.org/10.3390/foods2030374

14. **Boy E, Haas J, Petry N, Cercamondi CI, Gahutu JB, Mehta S, Finkelstein JL and RF Hurrell** Efficacy of iron-biofortified crops. *African Journal of Food, Agriculture, Nutrition and Development* 2017; **17:**11879-11892.

15. **Haas JD, Luna SV, Lung'aho MG, Wenger MJ, Murray-Kolb LE, Beebe S, Gahutu JB and IM Egli** Consuming iron-biofortified beans increases iron status in Rwandan women after 128 days in a randomized controlled feeding trial. *Journal of Nutrition* 2016; **146**:1586-1592. 25. https://doi.org/10.3945/jn.115.224741

16. **Luna SV** *The effects of iron-biofortified beans on iron status, physical performance and physical activity in Rwandan women*. Doctoral thesis, Cornell University, Ithaca NY, 2015.

17. **Murray-Kolb LE, Wenger MJ, Scott SP, Rhoten SE, Lung'aho MG and JD Haas** Consumption of iron-biofortified beans positively affects cognitive performance in 18- to 27-year-old Rwandan female college students in an 18-week randomized controlled efficacy trial. *Journal of Nutrition* 2017; **147(11)**:2109-2117. https://doi.org/10.3945/jn.117.255356

18. **DOST-FNRI (Department of Science and Technology-Food and Nutrition Research Institute).** Philippine Dietary Reference Intake. 2015; FNRI Bldg., DOST Compound, Gen. Santos Avenue, Bicutan, Taguig: Philippines.

19. **EURRECA (European micronutrient Recommendations Aligned: Nutri-RecQuest).** Eurreca Micronutrient Database, 2012.

20. **Mulambu J, Andersson MS, Palenberg M, Pfeiffer W, Saltzman A, Birol E, Oparinde A, Boy E, Asare-Marfo D, Lubobo A, Mukankusi C and S Nkalubo** Iron beans in Rwanda: crop development and delivery experience. Chapter 10. *African Journal of Food, Agriculture, Nutrition and Development*. Rural Outreach Program, 2017; **17(2)**: 12026-12050.

21. **CIAT (Centro Internacional de Agricultura Tropical)** Improved beans for the developing World. Executive summary. annual report 2008. Outcome Line. SBA-1. https://cgspace.cgiar.org/bitstream/handle/10568/69042/executive_summary_2008_beans.pdf?sequence=45&isAllowed=y *Accessed on 18th/10/2017.*

22. **Amongi W, Mukankusi C, Sebuliba S and F Mukamuhirwa** Iron and zinc grain concentrations diversity and agronomic performance of common bean germplasm collected from East Africa. *African Journal Food, Agriculture, Nutrition and Development* 2018; **18(3):** 13717-13742. https://doi.org/10.18697/ajfand.83.17070

23. **Kimani PM and A Warsame** Breeding second-generation biofortified bean varieties for Africa. *Food Energy Security* 2019; **8:e00173**.1-17. https://doi.org/10.1002/fes3.173

24. **Connorton JM and J Balk** Iron Biofortification of Staple Crops: Lessons and Challenges in *Plant Genetics, Plant and Cell Physiology* 2019; **60(7)**: 1447-1456. https://doi.org/10.1093/pcp/pcz079

25. **Izquierdo P, Astudillo C, Blair MW, Iqbal AM, Raatz B and KA Cichy** Meta-QTL analysis of seed iron and zinc concentration and content in common bean (*Phaseolus vulgaris* L.). *Theoretical and Applied Genetics* 2018; **131**:1645-1658. https://doi.org/10.1007/s00122-018-3104-8

26. **Ribeiro ND, Jost E, Maziero SM, Storck L and DP Rosa** Selection of common bean lines with high grain yield and high grain calcium and iron concentrations. *Revista Ceres, Viçosa* 2014; **61(1)**: 077-083.

27. **Ribeiro ND, Mambrin RB, Storck L, Prigol M and CW Nogueira** Combined selection for grain yield, cooking quality and minerals in the common bean. *Revista Ciencia Agronomica* 2013; **44**(**4**): 869-877.

28. **Gregorio GB** Progress in breeding for trace minerals in staple crops. *Journal of Nutrition* 2002; **132**: 500S-502S.

29. **Smith MR, Veneklaas E, Polania J, Rao IM, Beebe SE and A Merchant** Field drought conditions impact yield but not nutritional quality of the seed in common bean (*Phaseolus vulgaris* L.). *PLoS ONE* 2019; **14(6)**: e0217099. https://doi.org/10.1371/journal.pone.0217099

30. **Martins SM, Melo PGS, Faria LC, Souza TLPO, Melo LC and HS Pereira** Genetic parameters and breeding strategies for high levels of iron and zinc in *Phaseolus vulgaris* L. *Genetics and Molecular Research* 2016; **15**:01-14. https://doi.org/10.4238/gmr.15028011

31. **Pereira HS, Del Peloso MJ, Bassinello PZ, Guimarães CM, Melo LC and LC Faria** Genetic variability for iron and zinc content in common bean lines and interaction with water availability. *Genetics and Molecular Research* 2014; **13(3**): 6773-6785. https://doi.org/10.4238/2014.August.28.21

32. **Ugen MA, Karanja D, Birachi E, Katabalwa C, Ouma J and R Mutuku** *Pre-cooked beans for improving food and nutrition security and income generation in Kenya and Uganda*. Final Technical Report (October 2014 to June 2017). PP. 1-26. Retrieved from https://idl-bnc-idrc.dspacedirect.org/bitstream/handle/10625/56860/IDL-56860.pdf *on 24th/12/2019.*

33. **Wiesinger JA, Cichy KA, Tako E and RP Glahn** The fast cooking and enhanced iron bioavailability properties of the manteca yellow bean (*Phaseolus vulgaris* L.). *Nutrients* 2018; **10(11)**: 1609. https://doi.org/10.3390/nu10111609

34. **Cichy KA, Wiesinger JA and FA Mendoza** Genetic diversity and genome-wide association analysis of cooking time in dry bean (*Phaseolus vulgaris* L.). *Theoretical and Applied Genetics* 2015; **128**:1555-1567. https://doi.org/10.1007/s00122-015-2531-z

35. **Katuuramu DN, Luyima GB, Nkalubo ST, Wiesinger JA, Kelly JD and KA Cichy** On-farm multi-location evaluation of genotype by environment interactions for seed yield and cooking time in common bean. *Scientific Reports* 2020; **10**:3628. https://doi.org/10.1038/s41598-020-60087-2

36. **Mughi IM** *Inheritance and characterization of cooking time, seed iron and zinc content in selected African common bean germplasm* 2014 Thesis (Master of Science in plant breeding and seed systems), Makerere University, Kampala. UG.

37. **Garcia RAV, Rangel PN, Bassinello PZ, Bassinello PZ, Brondani C, Melo LC, Sibov ST and RP Vianello-Brondani** QTL mapping for the cooking time of common beans. *Euphytica: Netherlands Journal of Plant Breeding* 2012; **186(3):** 779-792. https://doi.org/10.1007/s10681-011-0587-7

38. **Jacinto-Hernandez C, Azpiroz-Rivero S, Acosta-Gallegos JA, Hernandez-Sanchez H and I Bernal-Lugo** Genetic analysis and random amplified polymorphic DNA markers associated with cooking time in common bean. *Crop Science* **43:**329–332. (2003). https://doi.org/10.2135/cropsci2003.0329

39. **Siddiq M and MA Uebersax** Dry Beans and Pulses Production and Consumption-an Overview. In: Dry Beans and Pulses: Production, Processing and Nutrition, Siddiq, M.A. and M.A. Uebersax (Eds.). John Wiley and Sons, USA., 2012, pp. 3-22.

40. **Kelly JD and KA Cichy** Dry bean breeding and production technologies, in *Dry beans and pulses: production, processing and nutrition*, ed. by Siddiq M and Uebersax MA. John Wiley, Oxford, 2013; pp. 23–54.

41. **Nyawira NS and KP Macharia** Canning quality of new grain runner bean lines developed in Kenya. *The Experiment* 2017; **43(1)**: 2463-2479.

42. **Cichy K, Fernandez A, Kilian A, Kelly J, Galeano C, Shaw S, Brick M, Hodkinson D and E Troxtell** QTL analysis of canning quality and colour retention in black beans. *Molecular Breeding* 2014; **33**:139-154. https://doi.org/10.1007/s11032-013-9940-y

43. **Qureshi AMI and R Sadohara** Breeding Dry Beans (*Phaseolus vulqaris* L.) with Improved Cooking and Canning Quality Traits. In: Qureshi A., Dar Z., Wani S. (eds) Quality Breeding in Field Crops. Springer, Cham (2019).

44. **Sandhu KS, You FM, RLConner, PM Balasubramanian and A Hou** Genetic analysis and QTL mapping of the seed hardness trait in a black common bean (*Phaseolus vulgaris*) recombinant inbred line (RIL) population. *Molecular Breeding* 2018; **38**:34. https://doi.org/10.1007/s11032-018-0789-y

45. **CIAT (Centro Internacional de Agricultura Tropical)** Standard system for the evaluation of bean germplasm. Van Schoonhoven, A. and Pastor-Corrales, M.A (complilers). Cali, Colombia. 1987; 54.

46. **Mukamuhirwa F, Tusiime G, Richard E, Mukankusi MC, Gibson P and R Edema** Characterization and identification of potential sources of high seed iron and zinc content among Uganda common bean germplasm. *Ambit Journal of Agriculture* 2015; **1(1)**: 01-16.

47. **Wang N and JK Daun** Determination of cooking times of pulses using an automated Mattson cooker apparatus. *Journal of Science Food and Agriculture* 2005; **85**:1631-1635. https://doi.org/10.1002/jsfa.2134

48. **VSN International** Genstat *for Windows* 20th Edition. VSN International, Hemel Hempstead, UK. Web page: Genstat.co.uk (2019).

49. **Holland JB, Nyquist WE and CT Cervantes-Martínez** Estimating and interpreting heritability for plant breeding: An update. *Plant Breeding* 2003; Rev. **22**:9-112.

50. **Finlay KW and GN Wilkinson** The analysis of adaptation in a plant-breeding programme. *Australian Journal of Agricultural Research* 1963; **14**:742-754.

51. **Ramalho MAP, Santos JB, Pinto CABP, Souza EA, Goncalves FMA and JC Souza** *Genética na agropecuária* (5a ed.). 2012. Lavras, MG: UFLA.

52. **Robinson HF, Comstock RE and PH Harvey** Estimates of heritability and the degree of dominance in corn. *Agronomy Journal* 1949; **41**:353-359.

53. **Mukamuhirwa F, Tusiime G and MC Mukankusi** Genotype x Environment interactions for higher Iron and Zinc in selected bean varieties. *Ambit Journal of Agriculture* 2015; **2**:16-39.

54. **Binagwa PH, He G, Bonsi E, Traore SM, Jaynes J and CK Bonsi** Determining grain seed micronutrient contents (iron and zinc) and cooking time for selected dry bean cultivars. *African Journal of Food, Agriculture, Nutrition and Development* 2020; **20(1**): 15415-15430 https://doi.org/10.18697/ajfand.89.18900

55. **Balasubramanian P, Slinkard A, Tyler R and A Vandenberg** A modified laboratory canning protocol for quality evaluation of dry bean (*Phaseolus vulgaris* L.). *Journal of Science, Food and Agriculture* 2000; **80(6):** 732-738.

56. **Hosfield GL, Uebersax MA and LG Occena** Technological and genetic improvements in dry bean quality and utilization. Proceedings of the Idaho Bean Workshop, August 3-4, 2000, University of Idaho, Moscow, pp: 135-152.

57. **Binagwa PH, Magdalena W, Michael K, Zakayo E, Mbiu J, Msaky J, Mdachi M, Kasubiri F, Kisamo A, Nestory SM and JC Rubyogo** Selian Agricultural Research Institute (SARI) released seven (7) improved common bean varieties January 2018. Fact Sheet 1.

58. **Nkalubo S, Kesiime E, Olaboro S, Aseete P, Nakibuuule J, Nantongo A, Ssekandi W and F Nakkazi** Legume news. Quarterly brief, 2016; **1(2)**: 1-2.