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Heritability Estimates for Micronutrient Composition of Sweetpotato Storage Roots

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Abstract. **Iron and zinc are micronutrients essential to the human diet but are in deficient supply to many in the tropics. Fortifying the micronutrient content of staple crops like sweetpotato [*Ipomoea batatas* (L.) Lam.] would go far in alleviating this intractable problem. This article presents estimates of broad-sense heritability for iron and zinc content in sweetpotato roots using a technique based on full-sibling families. Among individual genotypes, iron and zinc concentration varied by a fourfold and sixfold difference, respectively, whereas dry matter concentration showed a threefold variation. Family mean estimates varied significantly for the three traits. High broad-sense heritability for iron (H = 0.74), zinc (H = 0.82), and dry matter concentration (H = 0.93) were obtained among full-sibling families. These results suggest that traditional breeding strategies like mass selection could improve the micronutritional value of sweetpotato and that true sweetpotato seed, which has no international phytosanitary restrictions on transfer, can be used to quickly estimate heritability.**

Sweetpotato is vital to small-scale farmers with limited land, labor, and capital. It performs well in poor soils lacking sufficient nutrients and water and thrives in fertile environments where yields exceed those of cereal crops (Woolfe, 1992). Although sweetpotato is thus invaluable in combating chronic food shortages, the crop could contribute further to dietary micronutrition in regions of the tropics and subtropics where deficiencies are pronounced (Reddy et al., 2005).

Although micronutrients iron, zinc, and vitamin A are essential to the human diet, large segments of the global population experience health consequences from inadequate intake (Cichy et al., 2005; Long et al., 2004; Mason et al., 2001). Strategies to overcome these shortages abound, from fortified, processed foods to vitamin supplements; a complement to these approaches is to fortify existing staple crops (HarvestPlus, 2007). Notable among these efforts is the development of carotene-enriched rice (golden rice) through genetic transformation (Datta et al., 2007) and widely available β-carotene-rich sweetpotato germplasm. The use of this approach has been demonstrated with sweetpotato through the improved vitamin A status of children (van Jaarsveld et al., 2005).

Our present interest is to complement the known caloric and carotenoid contributions of sweetpotato to the diet by improving iron and zinc concentration. Recent evaluation of sweetpotato germplasm indicated an up to threefold difference in iron and zinc concentrations with iron up to ≈7 ppm fresh weight (fw) and zinc ≈4 ppm fw (Courtney, 2007). These innate mineral levels are meaningful from a dietary standpoint so the objective of this study was to determine the effectiveness of breeding for increased iron and zinc content as ascertained by broad-sense heritability estimates using a novel technique.

Materials and Methods

Plant material and experimental design. Field research was done at the Sweetpotato Research Center at Chase, LA. The soil was a Gilbert silt loam (fine-silty, mixed, thermic) with a pH of 5.4 to 5.6. Sixteen random soil samples were collected from the plot and average soil nutrients levels were as follows (2005 data): calcium = 795 ppm; copper = 0.73; magnesium = 140 ppm; phosphorus = 106 ppm; potassium (K) = 132 ppm; sodium (Na) = 98 ppm; zinc (Zn) = 0.87 ppm; and iron (Fe) = 74 ppm. Results were similar in 2004 except for Na = 49 ppm and K = 64 ppm.

Fifteen full-sibling families with a common male parent (PC03_1) were obtained as true botanical seed from the International Potato Center (CIP), Lima, Peru. The 15 female parents were 103001 (HY3.4), 103003 (YARDA), 103004 (192096.3), 103005 (193067.3), 103006 (195306.8), 103009 (195605.54), 103014 (199009.7), 103015 (199014.2), 103018 (199020.2), 103024 (100027.3), 103031 (199035.7), 103032 (199047.10), 103033 (199062.1), 103036 (100056.14), and 103072 (199076.1). Pedigrees of these CIP parents include parents from South America, the United States, and China. In the spring of 2004, seedlings derived from the true seed were transplanted in a completely randomized design with four replications. Seven full-sibling genotypes (each genotypically different progeny) were transplanted per single-row plot at 0.3 m between plants, a row width of 1 m, and 1.5 m between plots. The five middle plants of each plot were considered for analysis and represented a replicate. This experiment was repeated in the spring of 2005. Where possible, a single marketable root [U.S. #1 grade (5.1 to 8.9 cm diameter and 7.6 to 22.9 cm long)] was harvested for analysis from each of the five middle plants per plot. Preliminary studies have shown that genotype interroot variation for Fe and Zn concentration is not significant at either plant or interplant levels (Courtney, 2007). Roots were harvested on 21 Sept. 2004 and 30 Sept. 2005, 120 and 115 d after planting, respectively.

Micronutrient analysis. Zinc and Fe analysis was based on the methods of Norbotten et al. (2000). Harvested roots were cured at 85 °C and 85% relative humidity for 5 d and stored at 60 °C for up to 2 weeks. Each root was washed in tap water and allowed to air-dry. They were then rinsed in double-distilled water, peeled with a stainless steel knife, and rinsed in double-distilled water again. The roots were then sectioned, weighed, and dried at 80 °C for 48 h, after which they were weighed again. Dry matter was based on the differential between the results of the two weights, then the dry samples were pulverized using an IKA A10 Basic Analytical Mill (IKA Works, Wilmington, NC), and bottled in Corning Snap-Seal tubes (product no. 1730; Corning, New York, NY).

Analysis for various minerals was based on the methods of Havlin and Soltanpour (1980) and Huang and Schulte (1985). In short, 1-g samples were digested in 5 mL of nitric acid. The samples were placed on a Magnum 120 Plant/Soil Digester (Ivesdale, IL). After 45 min, a 3-mL aliquot of H_2O_2 was added to each sample, before the block reached 90 °C. The samples were heated until the volume was reduced to 0.5 mL, then diluted to 12.5 mL, and filtered using Whatman #2 paper (Whatman Intl., Maidstone, UK). The samples were then quantified for the various minerals through inductively coupled

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plasma mass spectrometry using a Spectro Ciros Charged Coupled Device (Kleve, Germany). For every 20 samples, a National Institute for Standards and Technology (Gaithersburg, MD) 1547 peach sample was used for repeatability measurements to ensure consistent results between runs.

Aluminum is an indicator of soil Fe contamination. In all cases, samples that showed aluminum levels above 3 ppm dry weight basis (dwb) were considered to be contaminated and were discarded. A generic threshold of greater than 5 to 6 ppm dwb was suggested by Pfeiffer and McClafferty (2007), but recent perspectives suggest greater than 3 ppm is more appropriate (Wolfgang Pfeiffer, personal communication, 2006) Additional research is needed in the area of contamination thresholds.

Assay data were analyzed using a technique adapted from Cisse and Ejeta (2003) for sorghum heritability estimates. Broad-sense heritability based on family means was calculated using the formula: $H = [MS_{among\ families} - MS_{year*family}]/MS_{among\ families}$, where MS is the mean square. Analyses of variance were used to examine differences in Fe and Zn concentration on fw basis and percent root dry matter concentration. Presented data represent a combined set for the 2 years given no significant year effect at $P = 0.05$.

Results and Discussion

Family mean estimates for Fe concentration varied significantly from 3.99 ppm for 103014 to 5.35 ppm for 103015 on a fw basis (Table 1). Iron concentration for individual genotypes across all families ranged from 2.22 ppm for a genotype of 103033 to 9.97 ppm for a genotype of 103072. The extent of variability observed among individual genotypes in the present study is greater than the 2.5-fold difference previously found among a diverse collection of 61 genotypes (Courtney, 2007) and indicates a broad range of potential iron uptake in this present population.

The Fe concentrations observed in the present population would make a meaningful contribution to the human diet. A root weighing 300 g from the highest ranking genotypes would provide 25% to 35% of the daily allowance of Fe based on an average daily requirement of 8 mg·d^{-1} (Anonymous, 2007). Furthermore, the broad-sense heritability (H) estimate for iron concentration was 0.74, suggesting there is potential in breeding for increased Fe concentration.

Data documenting Fe concentration in plants is based on estimates with low aluminum concentration. Aluminum is considered an artifact of soil contamination and residual soil Fe is included in the Fe concentration measurement (Gabriela Burgos, personal communication, 2004). Mean aluminum estimates by family ranged from 1.61 ppm for 103032 to 2.22 ppm for 103009; the family with the highest mean Fe estimate, 103015, had a mean aluminum concentration of 1.30 ppm. The two highest ranking genotypes (greater than 9 ppm Fe concentration) had aluminum concentrations of less than 2.3 ppm. The genotype with the highest aluminum concentration (≈3 ppm) ranked in the middle of the population (4.9 ppm Fe). Collectively, these data suggest that soil contamination was not a confounding factor in our heritability estimates.

Family mean estimates for zinc varied significantly from 2.27 ppm for 103031 to 3.12 ppm for 103001 on a fw basis (Table 1). Zinc concentration for individual genotypes ranged from 1.04 ppm for a genotype of 103009 to 6.40 ppm for a genotype of 103009. The extent of variability among individual genotypes in the present study is greater than the threefold difference found in a germplasm screen (Courtney, 2007) and indicates a broad range of potential Zn uptake in this present population.

Zinc concentrations, like those for Fe, in the current study are meaningful to meet human nutritional needs. A root weighing 300 g from the highest ranking genotypes would provide 25% of the daily allowance of Zn based on an average daily requirement of 8 mg·d^{-1}. The highest ranking genotypes had Zn levels nearly double the concentration of any genotype previously assayed (Courtney, 2007) and suggest that genetic engineering is not necessary to improve Zn concentration. The broad-sense H estimate for Zn concentration was 0.82. This estimate suggests that breeding for increased Zn concentration is possible.

Family mean estimates for dry matter varied significantly from 25.56% for 103009 to 33.28% for 103072. Dry matter concentration for individual genotypes ranged from 16.35% for a genotype of 103009 to 45.39% for a genotype of 103024, similar to a previous germplasm screen (Courtney, 2007). The broad-sense H estimate for dry matter concentration was 0.92. Our results were higher but consistent with previous narrow-sense heritability values (0.48 and 0.65) estimated by Jones (1977) and Liang (1982), respectively. Jones (1986) showed that broad-sense H estimates tended to be higher than narrow-sense estimates in sweetpotato for the same trait estimates.

Taken in concert, these results suggest that improvements in Fe and Zn concentration in sweetpotato are possible using traditional mass selection breeding techniques. The high levels present in this population alone underscore the use of including Fe and Zn as selectable breeding traits. The challenge is to combine high concentrations of Fe and Zn and high dry matter with other agronomic traits adaptable to varied tropical environments.

One caveat to our work is that H estimates may differ for other sweetpotato populations. Another is that a different soil environment will most likely affect micronutrient uptake and thus alter H estimates.

Broad-sense H estimates such as those derived from this study are not as informative as narrow-sense estimates because total genetic variance is not partitioned; however, in the face of international restrictions on the transfer of sweetpotato plant material, the method used here with true seed allows populations to be quickly evaluated in different environments without the transfer of plant material.

Table 1. Half-sibling family mean estimates for iron, zinc, and dry matter in sweetpotato storage roots.

Family	Iron[z, y]	Zinc[z, y]	Dry matter (%)
103001	5.13 ± 0.23 a	3.12 ± 0.14 a	32.05 ± 0.81 ab
103003	4.78 ± 0.25 ab	2.74 ± 0.15 abc	30.62 ± 0.88 abc
103004	4.76 ± 0.31 ab	2.47 ± 0.18 abc	31.43 ± 1.10 abc
103005	4.80 ± 0.26 ab	2.52 ± 0.16 abc	29.27 ± 0.93 abcd
103006	5.33 ± 0.27 a	3.03 ± 0.16 ab	30.24 ± 0.96 abc
103009	5.15 ± 0.21 a	2.40 ± 0.13 bc	25.56 ± 0.76 d
103014	3.99 ± 0.23 b	2.34 ± 0.14 bc	28.28 ± 0.81 bcd
103015	5.35 ± 0.27 a	2.80 ± 0.16 abc	31.99 ± 0.98 ab
103018	4.20 ± 0.26 ab	2.34 ± 0.15 bc	30.03 ± 0.91 abc
103024	4.93 ± 0.21 ab	2.72 ± 0.12 abc	28.84 ± 0.74 bcd
103031	4.66 ± 0.21ab	2.27 ± 0.12 c	27.01 ± 0.74 cd
103032	5.05 ± 0.25 ab	2.79 ± 0.15 abc	29.17 ± 0.89 abcd
103033	4.59 ± 0.23 ab	2.43 ± 0.14 bc	29.23 ± 0.82 abcd
103036	5.12 ± 0.22 a	2.75 ± 0.13 abc	29.78 ± 0.77 abc
103072	4.75 ± 0.28 ab	2.36 ± 0.17 bc	33.28 ± 1.01 a

[z]Iron and zinc concentrations in ppm on a fresh weight basis.
[y]Mean separation within columns by LSD, $P < 0.05$.

Literature Cited

Anonymous. 2007. Dietary reference intake tables. Institute of Medicine of the National Academy of Sciences. 20 July 2007. <http://www.iom.edu/Object.File/Master/7/294/Webtableminerals.pdf>.

Cichy, K.A., S. Forster, K.L. Grafton, and G.L. Hosfield. 2005. Inheritance of seed zinc accumulation in navy bean. Crop Sci. 45:864–870.

Cisse, N. and G. Ejeta. 2003. Genetic variation and relationships among seedling vigor traits in sorghum. Crop Sci. 43:824–828.

Courtney, M. 2007. Genotypic variability and inheritance of iron and zinc in sweetpotato. M.S. thesis. School of Plant, Environmental, and Soil Sciences, Louisiana State University, Baton Rouge. 19 Sept. 2007. <http://etd.lsu.edu/docs/available/etd-02252007-121902/unrestricted/thesis.pdf>.

Datta, S.K., K. Datta, V. Parkhi, M. Rai, N. Baisakh, G. Sahoo, S. Rehana, A. Bandyopadhyay, Md. Alamgir, Md.S. Ali, E. Abrigo, N. Oliva, and L. Torrizo. 2007. Golden rice: Introgression, breeding, and field evaluation. Euphytica 154:271–278.

HarvestPlus. 2007. 19 Sept. 2007. <http://www.harvestplus.org/micronut.html>.
Havlin, J.L. and P.N. Soltanpour. 1980. A nitric acid plant tissue digest method for use with inductively coupled plasma spectrometry. Commun. Soil Sci. Plant Anal. 11:969–980.
Huang, C.-Y. and E.E. Schulte. 1985. Digestion of plant tissue for analysis by ICP emission spectroscopy. Commun. Soil Sci. Plant Anal. 16:943–958.
Jones, A. 1977. Heritabilities of seven sweet potato root traits. J. Amer. Soc. Hort. Sci. 102:440–442.
Jones, A. 1986. Sweetpotato heritability estimates and their use in breeding. Hort. Sci. 21:14–17.
Liang, L. 1982. The inheritance of crude starch percentage and its correlation with root yield and other traits in sweet potatoes. Chiayi Agr. Expt. Sta. Taiwan Agr. Res. Inst. Natl. Sci. Council Monthly 10:765–772.
Long, J.K., M. Bänziger, and M.E. Smith. 2004. Diallel analysis of grain iron and zinc density in southern-African adapted maize hybrids. Crop Sci. 44:2019–2026.
Mason, J.B., M. Lotfi, N. Dalmiya, K. Sethuraman, and M. Dietchler. 2001. The micronutrient report: Current progress and trends in the control of vitamin A, iodine, and iron deficiencies. International Development Research Centre, Ottawa, 19 Sept. 2007. <http://www.micronutrient.org/resources/publications/mn_report.pdf>.
Norbotten, A., E.B. Loken, and A.H. Rimestad. 2000. Sampling of potatoes to determine representative values for nutrient content in a national food composition table. J. Food. Compos. Anal. 13:369–377.
Pfeiffer, W.H. and B. McClafferty. 2007. Biofortification: Breeding micronutrient-dense crops, p. 61–91. In: M. Kang and P.M. Priyadarshan (eds.). Breeding major food staples. Blackwell Publishing, Oxford, UK.
Reddy, B.V.S., S. Ramesh, and T. Longvah. 2005. Prospects of breeding for micronutrients and β-carotene-dense sorghums. ISMN 46:10–14.
van Jaarsveld, P.J., M. Faber, S.A. Tanumihardjo, P. Nestel, C.J. Lombard, and A.J. Spinnler Benade'. 2005. β-carotene-rich orange-fleshed sweet potato improves the vitamin A status of primary school children assessed with the modified-relative-dose-response test. Amer. J. Clin. Nutr. 81:1080–1087.
Woolfe, J.A. 1992. Sweetpotato: An untapped food resource. Cambridge University Press, New York, NY.