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Bridging Crop Improvement for Feeding Children 6-23 months: Cowpea and Soybean Varieties in Zambia-A laboratory analysis

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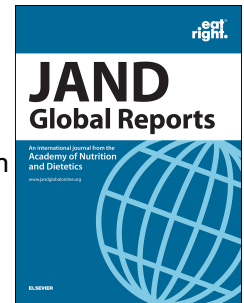
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1 **Bridging Crop Improvement for Feeding Children 6-23 months: Cowpea and Soybean**

2 **Varieties in Zambia-A laboratory analysis**

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RESEARCH SNAPSHOT**Research Question:**

What is the genotype-specific proximate and mineral composition of released and pre-released cowpea and soybean varieties in Zambia, and how do mineral synergies and antagonisms influence their potential contribution to feeding children aged 6–23 months?

Key Findings:

In this laboratory-based cross-sectional analysis of ten cowpea and seven soybean genotypes, soybean varieties showed significantly higher energy, protein, fat, calcium, zinc, copper, and selenium, while cowpea genotypes were richer in carbohydrates, fiber, potassium and magnesium with variable iron content. Principal Component Analysis (PCA) identified significant mineral synergies and antagonisms.

Bridging Crop Improvement and Feeding Children aged 6-23 months: Nutritional Potential of Released and Pre-Released Cowpea and Soybean Varieties in Zambia

BACKGROUND

Improving complementary feeding for children aged 6–23 months is a public health priority, and cowpea and soybean may offer affordable, protein- and mineral-rich options.

METHODS

Study subjects: 10 cowpea & 7 soybean genotypes

Nutrient quantification: Association Of Analytical Chemists (AOACs) & Inductively Coupled Plasma Optical Emission Spectroscopy (ICP-OES)

Data analysis: Principal Component Analysis (PCA)

CONCLUSION

Selected cowpea & soybean genotypes could strengthen complementary feeding by improving nutrient adequacy for children.

SOYBEANS



Nutrients

Rich in energy, protein, fat, calcium (Ca), phosphorous (P), copper (Cu), zinc (Zn) & selenium (Se)

Mineral-Positive associations

Mg-P

Both of them explained PC1 variation

Mineral-Weak Negative associations

K-(Ca-Fe)

PC2 variation was explained by K and Ca

COWPEAS



Nutrients

Rich in carbohydrates, protein, fiber, potassium, magnesium & Iron

Mineral-Positive associations

Cu-Zn

Ca-(K-Mg-Na-P)

K-(Mg-Na-P)

PC1 variation was explained by Ca, K, Mg, & Zn.

Mineral-Weak Negative associations

Cu-Fe

Both of them explained PC2 variation

ABSTRACT

Background: Improving complementary feeding for children aged 6–23 months is a public health priority. Cowpea and soybean can address protein and micronutrient gaps, yet genotype-specific nutrient profiles remain poorly characterized in Zambia.

Objective: To assess genotype-specific nutrient composition and mineral interactions relevant to feeding of children aged 6-23 months in Zambia.

Design: A laboratory-based cross-sectional study assessed nutrient composition using Association of Official Analytical Chemists (AOAC) and Inductively Coupled Plasma Optical Emission Spectroscopy (ICP-OES) methods.

Participants/Setting: Ten cowpea genotypes (four released varieties and six breeding lines) and seven soybean genotypes (five released varieties and two breeding lines) from the University of Zambia were analyzed at the Zambia Bureau of Standards (ZABS).

Methods: Mineral variability and associations were assessed using medians with ranges, Spearman's correlations, principal component analysis (PCA) and cluster analysis.

Results: Soybean genotypes were higher in energy (473 kcal/100 g), protein (43%), fat (21%), Ca, P, Cu, Zn, and Se; cowpea genotypes were higher in carbohydrates (47%), fiber (7%), K, Mg, and Fe. Cowpea genotypes showed significant positive correlations among Cu–Zn, Ca–K/Mg/Na/P and K–Mg/Na/P ($r=0.65–0.94$; $p<0.05$), while soybean genotypes showed a similar pattern between Mg–P ($r=0.96$; $p<0.05$); negative associations were weak. Cowpea genotypes PCA explained 76.52% of variation (PC1=60.30%; PC2=16.22%), while soybean genotypes PC1 explained 41.47%. Cluster analysis corroborated PCA-based genotype differentiation.

Conclusions: Nutrition-sensitive selection of cowpea and soybean genotypes may have potential to support dietary adequacy among children and other population groups at risk of poor nutritional status

Key words: Cowpea–soybean genotypes; Mineral associations; Complementary feeding; Principal components analysis; Child nutrition.

INTRODUCTION

Cowpea (*Vigna unguiculata*) and soybean (*Glycine max*), key crops within the family Leguminosae, occupy a strategic position at the nexus of crop improvement, food systems, and child nutrition.¹ In Zambia and comparable low- and middle-income countries, cowpea and soybean are affordable, locally available sources of essential nutrients that can improve dietary quality and strengthen complementary feeding practices among children aged 6–23 months, particularly where access to animal-source foods is constrained by cost and availability.^{2–4} Despite their recognized nutritional potential and relevance for addressing malnutrition and food insecurity, legumes such as cowpea and soybean remain underutilized in many developing regions, including within child feeding and nutrition-sensitive programmes.^{5,6}

Cowpea (*Vigna unguiculata*), a legume native to Africa, is a climate-resilient and versatile crop widely cultivated across tropical and subtropical regions, including Southern Africa.^{7,8} It plays a multifaceted role within food systems as a staple food for human consumption, a feed resource for livestock, and an important income-generating commodity for smallholder farmers.⁹ Cowpea is consumed in diverse forms, including snacks, soups and stews, enhancing its adaptability for incorporation into complementary feeding (CF) and nutrition-sensitive food products.¹⁰ Cowpea is also recognized for its health-promoting properties, including potential protective effects against

non-communicable diseases and obesity, largely attributable to its phytochemical profile, dietary fiber content, and favorable fatty acid composition.¹¹ However, cowpea also contains anti-nutritional factors such as phenolic compounds, phytic acid, and protease inhibitors, which can compromise protein digestibility and reduce the bioavailability of critical micronutrients, notably iron and zinc, nutrients of public health concern among young children.^{12,13} Phenolic compounds are known to bind dietary proteins and chelate divalent metal ions, while phytic acid forms complexes with phosphorus and other essential minerals, limiting their absorption and utilization in the human body.^{14–16}

Soybean (*Glycine max*) is a nutrient-dense legume of growing relevance to public health, particularly for improving the quality of complementary foods. Its high protein and lipid content makes soybean a valuable plant-based source of energy, essential amino acids, and fat-soluble nutrients needed during the critical window of growth and development.¹⁷ Originating from East Asia and cultivated for millennia,¹⁸ soybean is now produced across diverse agroecological zones worldwide, including sub-Saharan Africa, where Zambia is an emerging producer.^{19,20}

Soybean utilization in Zambia has historically focused on commercial value chains, such as cooking oil extraction and animal feed, with limited integration into household diets.^{21,22} For human consumption, soybean is mainly processed into soy flour, instant soy products, and textured

vegetable protein (TVP), which can be incorporated into porridges, relishes, and composite complementary foods suitable for young children.^{21,23}

Similar to cowpea, soybean also contains relatively high levels of anti-nutritional factors, including protease inhibitors and lectins, which may impair protein digestibility and nutrient absorption in young children if inadequately treated.^{24,25}

Household- and community-level processing methods such as soaking, boiling, germination, and fermentation are critical for improving the suitability of cowpea- and soybean-based foods for infant and young child feeding (IYCF). These processing techniques have been shown to substantially reduce anti-nutritional factors, thereby enhancing protein digestibility, mineral bioavailability, and overall nutrient quality of legume-based complementary foods.^{13,26}

Emerging evidence further demonstrates significant genetic and nutritional diversity among cowpea and soybean genotypes, offering opportunities for nutrition-sensitive crop selection and breeding to enhance the nutrient density of complementary foods.²⁷ Leveraging genotype-specific nutrient profiles can support the selection of soybean and cowpea varieties that are better aligned with the nutrient requirements and developmental milestones of children during the complementary feeding (CF) period. However, in Zambia, data on the nutrient composition of locally available cowpea and soybean varieties remain limited. In response, this study aims to characterize the nutrient profiles of selected cowpea and soybean genotypes. The findings will generate evidence to inform the integration of nutrient-dense, appropriately processed legumes

into child nutrition programmes, complementary feeding (CF) guidelines, and food-based strategies to improve dietary quality in early childhood.

MATERIALS AND METHODS

Study Setting and subjects

This laboratory based cross sectional study was designed to generate evidence relevant food-based strategies targeting nutrition milestones among children aged 6-23 months in resource-limited settings. It utilized ten cowpea genotypes (four released varieties and six advanced lines) and seven soybean genotypes (five varieties and two lines) obtained from the University of Zambia (UNZA). All selected genotypes were derived from indigenous cowpea and soybean germplasm that had demonstrated favorable agronomic traits, including high yield potential and insect resistance characteristics that are important for ensuring stable, affordable, and sustainable food sources for household-level child feeding programmes. It was hypothesized that selected indigenous cowpea and soybean genotypes with favorable agronomic traits would differ significantly in nutrient composition, with some genotypes exhibiting superior protein, energy, and micronutrient profiles suitable for improving nutrient adequacy during complementary feeding.

Laboratory analyses were conducted at the Zambia Bureau of Standards (ZABS), ensuring analytical rigor and reliability of nutrient data. The laboratory is accredited by the Southern African Development Community Accreditation Services (SADCAS) as a testing laboratory (Accreditation Number: TEST-5 0018).

Nutrient Data Base Information

Nutrient composition of cowpea and soybean data were collected using Microsoft excel software versions 2013. Final calculations were completed using STATA software version 2015 (StataCorp, College Station, Texas). The dataset supporting this study is publicly available in the Open Science Framework (OSF) repository (<https://doi.org/10.17605/OSF.IO/BZAXH>).

Macronutrient Composition

This is the quantitative assessment of key macronutrient components; moisture, total protein, fat, ash, and dietary fiber.²⁸ These components are critical for supporting optimal growth and development during early childhood, as protein and fat contribute to tissue growth, brain development, and energy provision, while fat also enhances the absorption of fat-soluble vitamins.²⁹ Moisture and dietary fiber influence food bulk, viscosity, and digestibility,^{30,31} and ash content provides an indirect measure of total mineral content needed to meet micronutrient requirements.³²

Determination of macronutrient composition

Macronutrient composition was determined using Laboratory Testing Methods (LTM) 002IL in accordance with AOAC method 968.08.³³ Analyses were performed in the General Chemistry Laboratory using a Perten Grain Analyzer called PerkinElmer DA 7250™ NIR Analyzer (PerkinElmer, Inc., Waltham, Massachusetts, USA), which applies near-infrared reflectance (NIR) spectroscopy to rapidly and reliably quantify key macronutrients and fiber.^{34,35} Finely ground cowpea and soybean samples were analyzed in duplicate, with mean values for moisture, protein, fat, fiber, and ash expressed as percentages, providing evidence to inform the suitability of these legumes for improving diet quality for children aged 6-23 months.

Carbohydrate content was estimated using the difference method, a standard approach in food composition analysis that is relevant for assessing the energy contribution of carbohydrates to foods. Specifically, carbohydrate content (%) was calculated as 100 minus the sum of moisture (%), ash(%), protein(%), fat(%), and crude fiber(%), as applied in a recent study conducted in an East African context³⁶ as follows.

$$\text{Carbohydrate content (\%)} = 100 - [\text{Moisture (\%)} + \text{Ash (\%)} + \text{Protein (\%)} + \text{Fat (\%)} + \text{Crude fiber (\%)}]$$

The total energy value (TEV) of cowpea and soybean was estimated using the Atwater general conversion factors, which quantify the energy contribution of protein, fat, and carbohydrates in foods. This approach provides a standardized method for assessing the energy density of foods and is particularly relevant to inform the suitability of these legumes for improving diet quality for children aged 6-23 months.^{37,38} The Atwater system applies standardized energy conversion factors for the major macronutrients; protein, fat, and carbohydrates, irrespective of the food source. This approach enables consistent estimation of the energy density of complementary foods and supports assessment of whether legume-based diets, such as those derived from cowpea and soybean, can adequately contribute to the energy requirements.³⁷ The energy values applied represent the average metabolizable energy of macronutrients, accounting for physiological losses during digestion, absorption, and urinary nitrogen excretion. Standard Atwater conversion factors of 4.0 kcal/g for protein, 9.0 kcal/g for fat, and 4.0 kcal/g for carbohydrates³⁹ were used to estimate the energy levels of cowpea and soybean genotypes. This also helped to verify the energy values reported by the laboratory.

Determination of minerals

The determination of mineral content was conducted using Laboratory Test Method (LTM) 001 in accordance with Association of Analytical Chemists (AOAC) Official Method 968.08.³³ Mineral analysis was performed using Inductively Coupled Plasma Optical Emission Spectroscopy (ICP-OES; PerkinElmer Inc., Waltham, Massachusetts, USA), a technique widely used for the sensitive and reliable quantification of macro- and trace minerals in foods.⁴⁰ ICP-OES analysis, atoms and ions are excited within an intense electromagnetic field, causing them to emit light at element-specific wavelengths, with emission intensities directly proportional to their concentrations. ICP-OES enables precise, simultaneous detection and quantification of multiple essential macro and trace minerals in food, provided their characteristic emission wavelengths do not significantly overlap.^{41,42}

Samples were dry-ashed using a muffle furnace, followed by acid digestion with hydrochloric acid to solubilize the mineral content. The digested samples were diluted with distilled water and analyzed in duplicate using ICP-OES after instrument calibration with appropriate standards. Samples were introduced into the instrument as aerosols, allowing element-specific emission at characteristic wavelengths to be detected and quantified. Mineral concentrations were expressed as mg/kg (parts per million, ppm), providing standardized estimates relevant for evaluating micronutrient levels in cowpea and soybean genotypes with potential in feeding children aged 6-23 months.

Institutional Review Board

This study was deemed exempt under the National Health Research Authority (NHRA) Act No. 2 of 2013 of the Laws of Zambia. Reference: <https://www.nhra.org.zm/>

Data analysis

Nutrient composition of cowpea and soybean genotypes were summarized using median with ranges to capture central tendency and variability. Kruskal–Wallis test was used to check for differences within genotypes. Similarly, a Mann Whitney U test was used to check for differences between the two legumes. Spearman’s rank correlation analysis examined associations between mineral pairs, describing patterns of nutrient co-occurrence and potential relationships relevant to mineral profiles in legume-based complementary foods. This non-parametric approach was appropriate given the limited number of genotypes analyzed and the absence of assumptions of normality and homogeneity of variance, ensuring robust interpretation of nutrient profiles in cowpea and soybean genotypes.⁴³

Principal Component Analysis (PCA) was applied to the grain mineral composition of cowpea and soybean genotypes as an exploratory technique to reduce data dimensionality and identify patterns of mineral variability and association.³⁴ Principal components (PCs) were derived from eigenvalues representing the variation explained by each component, while eigenvector loadings indicated the relative contribution of individual minerals following linear transformation of the original variables.⁴⁴ Loadings of $\geq \pm 0.30$ were considered meaningful, and components with eigenvalues >1 were retained, consistent with comparable nutrition profiling studies.⁴⁴ This approach identified dominant mineral patterns and mineral-dense genotype groupings potentially relevant for selecting nutrient-rich complementary foods to support the micronutrient requirements of children aged 6–23 months, without implying causal relationships among minerals.⁴⁵

Cronbach’s alpha coefficient was calculated using mineral variables with meaningful loadings on the retained PCs to assess the internal consistency of the identified nutrient patterns rather than the

PC loadings themselves.⁴⁶ This provided additional evidence of the reliability of the multivariate structure and strengthened confidence in identifying nutritionally relevant cowpea and soybean genotypes for complementary feeding applications.

In addition, cluster analysis was conducted to group cowpea and soybean genotypes with similar mineral profiles, facilitating identification of nutritionally comparable and mineral-dense genotype clusters with potential application in the development of complementary foods.

Validity of nutrient analysis

The study used standardized and validated laboratory methods to ensure reliable nutritional profiling assessments. Macronutrient composition was analyzed using AOAC 968.08 with a calibrated NIR Grain Analyzer, while mineral content was determined using ICP-OES following AOAC protocols, with duplicate analyses and instrument calibration to enhance accuracy. Internal validity was strengthened through quality-controlled procedures at a SADCAS-accredited ZABS laboratory and appropriate statistical analyses. However, no external validation through dietary intake data or feeding trials was conducted.

RESULTS

Cowpea and soybean genotypes profile

The study assessed indigenous cowpea and soybean genotypes sourced from the University of Zambia (UNZA). The sample comprised ten cowpea genotypes (four released varieties and six breeding lines) and seven soybean genotypes (five released varieties and two breeding lines) as shown in **Table 1**.

Macronutrient compositions of cowpea and soybean genotypes

According to Table 2, cowpea genotypes had a median energy content of 372 kcal/100 g (371–375), with median moisture (12%; 11–13), carbohydrates (47%; 46–49), protein (37%; 36–38), fat (4%; 3–5), crude fiber (7%; 7–8), and ash (4%; 4–4). Among the genotypes, Bubebe had the highest carbohydrate content, LT 4-2-4-1 the highest protein, while BB 10-4-2-3 recorded the highest moisture, fat, crude fiber, and ash values.

In soybean, the median energy content was 473 kcal/100 g (466–481), with median moisture (8%; 7–8), carbohydrate (29%; 27–32), protein (43%; 40–44), fat (21%; 20–22), crude fiber (6%; 5–6), and ash (6%; 5–6). Among the varieties, SG Sentinel had the highest energy and fat, Dina the highest moisture, crude fiber, and ash, Lukanga the highest carbohydrate, and GX 2033-16 GZ the highest protein.

Major minerals

Table 3 summarizes the mineral composition of cowpea and soybean genotypes as median concentrations (range, mg/kg). In cowpea, median concentrations were calcium (1312.7; 802.0–1913.8), potassium (20788.0; 15,090.1–24,023.9), magnesium (3154.6; 2351.0–3673.8), sodium (437.9; 61.9–830.7), and phosphorus (3596.2; 2358.5–4773.9). Among the genotypes, LT 11-3-3-12 had the highest calcium, potassium, and magnesium concentrations, while Lukusuzi had the highest sodium and phosphorus.

For soybean, median mineral concentrations (mg/kg) were calcium (6519.1; 5637.5–7411.5), potassium (16536.4; 15,301.0–17,260.0), magnesium (2223.6; 2055.5–2415.0), sodium (184.4; 95.9–264.3), and phosphorus (5667.9; 4709.5–6075.5). Among the varieties, SC Saga had the highest calcium, magnesium, and phosphorus concentrations, Dina the highest potassium, and GX 2033-16 GZ the highest sodium.

Trace Elements

According to Table 3, cowpea trace element concentrations (mg/kg) were copper (6.1; 4.5–7.5), iron (119.1; 58.4–518.9), and zinc (30.2; 21.5–34.4). Among the genotypes, Lunkwankwa had the highest copper concentration, BB 10-4-2-3 the highest iron, and LT 3-8-4-6 the highest zinc. **For soybean**, trace element concentrations (mg/kg) were copper (8.5; 7.4–10.0), iron (87.6; 70.7–107.0), zinc (50.1; 47.4–56.3), and selenium (3.1; 1.3–6.3). Among the varieties, SC Saga had the highest copper, zinc, and selenium concentrations, while TG 2033-93GZ had the highest iron.

Comparison of Macronutrient Composition and Mineral Concentrations Within and Between Cowpea and Soybean Genotypes

No statistically significant differences in macronutrient composition, major mineral concentrations, or trace element concentrations were observed within cowpea genotypes or within soybean genotypes. However, significant differences between cowpea and soybean ($p < 0.05$) were observed for all macronutrient components assessed, including moisture, carbohydrate, crude fiber, energy, protein, fat, and ash; major minerals including Ca, K, Mg, and P; and trace elements including Cu, Zn, and Se. Fe was the only trace element that did not differ significantly between the two crops.

Cowpea grain mineral correlations

Figure 1 presents pairwise associations among cowpea grain minerals relevant to feeding children aged 6–23 months. Significant positive associations ($p < 0.05$) were observed between Cu and Zn ($r = 0.7$); Ca and K ($r = 0.9$), Mg ($r = 0.8$), Na ($r = 0.65$), and P ($r = 0.7$); K and Mg ($r = 0.9$), Na ($r = 0.8$), and P ($r = 0.7$); and Mg and Na ($r = 0.8$). Weak negative mineral associations included Cu vs (Fe $r = -0.09$).

Soybean grain mineral correlations

Pairwise associations among soybean grain minerals relevant to feeding children aged 6–23 months revealed a significant positive association between Mg and P ($r = 1.0$, $p < 0.05$). Weak negative mineral associations included K vs Ca ($r = -0.29$) and Fe ($r = -0.25$) (**Fig. 1**).

Cowpea grain mineral principal component analysis

The eight measured mineral characteristics of cowpea were reduced to two PCs. Both components had eigenvalues ≥ 1.00 and together explained 77% of the total variation in mineral composition relevant to feeding children aged 6–23 months. Cronbach's alpha coefficients indicated high internal consistency for the two PCs, with $\alpha = 0.93$ for PC1, classified as excellent, and $\alpha = 0.80$ for PC2, classified as acceptable⁴⁶.

PC1 explained 60.30% of the total variance (eigenvalue = 4.82) and was characterized by positive loadings for K, Mg, Zn, P, Ca (0.4) and Na (0.3). PC2 explained 16% of the variance (eigenvalue = 1.30) and had loadings as follows: positive for Fe and Ca (0.5), being negative for Cu (−0.6) and Zn (−0.3).

The cowpea varieties Lukusuzi and Lutembwe, together with breeding lines LT 11-3-3-12, LT 4-2-4-1, and LT 3-8-4-6, contributed to PC1. Contribution to PC2 was observed for cowpea varieties Lukusuzi, Bubebe, and Lunkwankwa, as well as breeding lines LT 11-5-2-2 and BB 10-4-2-3 (**Table 4**).

Cowpea grain mineral principal component analysis biplot

PCA biplots were used to visualize the distribution and mineral associations of cowpea genotypes relevant to complementary feeding (CF) (**Fig. 2**). The direction of the positive and negative loadings in the biplots reflects the nature of the associations between the PCs and mineral variables.⁴⁷ PCA of the ten cowpea genotypes selected based on copper content showed that the variety *Lunkwankwa* and the lines LT 3-8-4-6, LT 4-2-4-1, and BB 10-4-2-3 were positively

associated with Cu, Zn, P, and Fe, respectively. In contrast, the variety *Lukusuzi* and line LT 11-5-2-2 were negatively associated with Na, while line LT 14-3-3-12 showed a negative association with Ca.

Soybean grain mineral principal component analysis

PCA of soybean mineral composition identified three PCs with eigenvalues ≥ 1.00 , explaining 82% of total variation and indicating patterns of mineral association among soybean genotypes. PC1 (eigenvalue=3.73) explained 41% of total variance, PC2 (eigenvalue=2.27) explained 25%, and PC3 (eigenvalue=1.37) explained 15%. Collectively, these components accounted for 82% of cumulative variance, indicating major patterns of mineral association potentially relevant to feeding children aged 6–23 months in Zambia (**Table 4**).

The internal consistency of the mineral groupings within each PC, as assessed using Cronbach's alpha (α), was high for PC1 ($\alpha = 0.83$), indicating strong coherence among mineral loadings, moderate for PC2 ($\alpha = 0.65$), indicating acceptable internal consistency, and low for PC3 ($\alpha = 0.53$), indicating weaker internal consistency among contributing minerals. PC1 therefore had high internal consistency. Soybean genotypes contributing to PC1 for major minerals (Ca, K, Mg, Na, and P), characterized by higher positive loadings of Mg and P, included the varieties *Lukanga*, *Safari*, *Dina*, *SC Saga*, and *SG Sentinel*, as well as the lines GX 2033-16 GZ and TG 2033-93 GZ.

Soybean grain mineral principal component analysis biplot

The PCA biplot of the seven soybean genotypes selected based on copper (Cu) content illustrated the distribution of mineral associations relevant to feeding of children aged 6-23 months (**Fig.2**). The biplot showed that the varieties *Dina* and *Safari* were positively associated with K, while the line GX 2033-16 GZ was negatively associated with Ca, Fe, and Na. In addition, the lines TG 2033-93 GZ and TG 2033-93 GZ displayed negative associations with Fe.

Cowpea and Soybean Grain Mineral Composition Cluster Analysis

Cluster analysis grouped the ten cowpea genotypes into four mineral-based clusters and the seven soybean genotypes into three clusters. For both legumes, the cluster groupings corresponded closely with the patterns identified by PCA. Consequently, detailed cluster-specific results were not presented, while PCA and Cronbach's alpha results were retained for reporting.

DISCUSSION

This study demonstrates that specific locally adapted cowpea and soybean genotypes exhibit nutrient profiles of direct relevance to feeding children aged 6-23 months, a period marked by high nutrient requirements and persistent risks of micronutrient deficiencies.⁴⁸ Macronutrient composition results showed that soybean genotypes, particularly *SG Sentinel* and *SC Saga* contained higher levels of energy and fat, while *SG Sentenel*, *Dina*, *Safari* and *GX2033-16GZ* were higher in protein, compared with cowpea genotypes. Similar to the present results, studies from sub-Saharan Africa and South Asia consistently report that soybean genotypes are characterized by higher fat and protein⁴⁹ content than cowpea and other pulses, making them particularly suitable for complementary foods. For example, recent evidence shows that soybean-based blends has the potential for use as an effective complementary diet capable of meeting the daily dietary requirements for children with protein energy malnutrition (PEM).⁵⁰ The higher energy and fat levels observed in genotypes such as *SG Sentinel* and *SC Saga*, alongside elevated protein in *SG Sentinel*, *Dina*, *Safari*, and *GX2033-16GZ*, are therefore consistent with broader findings highlighting soybean's value as a nutrient-dense legume for child feeding.⁵¹ Conversely, the observed higher carbohydrate, dietary fiber and moderate protein content in cowpea genotypes such as *Bubebe*, *Lutembwe*, *Lukusuzi*, *LT 11-5-2-2*, *LT 11-3-3-12*, and *LT 4-2-4-1* aligns with recent literature emphasizing cowpea's role as an inexpensive food crop particularly in food-

insecure households.^{52,53} Cowpea could be used to support a food-based approach to improving dietary adequacy among children aged 6-23 months in resource-constrained settings.

Mineral profiling revealed clear crop- and genotype-specific patterns in nutrients of public health importance. Soybean genotypes *SC Saga*, *Lukanga*, and *GX 2033-16 GZ* consistently exhibited higher calcium (Ca) concentrations, while *SC Saga*, *Dina*, and *Safari* recorded higher phosphorus (P) levels. In contrast, cowpea genotypes *Lukusuzi*, *LT 11-3-3-12*, and *LT 4-2-4-1* showed higher potassium (K) and magnesium (Mg) concentrations, and wide variation in iron (Fe) content was observed across cowpea genotypes, with *BB 10-4-2-3* recording notably high Fe levels. These genotype-specific differences are highly relevant in public health contexts where Fe deficiency anemia, inadequate Ca intake, and low dietary diversity remain prevalent among young children.^{54,55}

Correlation and PCA highlighted important positive mineral associations that may enhance the nutritional value of legume-based complementary foods. In cowpea, strong positive associations among Ca, K, Mg, and P were driven largely by genotypes such as *Lukusuzi*, *Lutembwe*, *LT 11-3-3-12*, *LT 4-2-4-1*, and *LT 3-8-4-6*, indicating that selection of these genotypes may simultaneously improve multiple interrelated minerals essential for bone development, neuromuscular function,⁵⁶ and energy metabolism⁵⁷ during early childhood. Positive copper (Cu) –zinc (Zn) associations, particularly evident in *Lunkwankwa* and *LT 3-8-4-6*, further suggesting potential contributions to immune function and enzymatic processes critical for growth and infection resistance among children aged 6–23 months.^{58,59}

At the same time, multivariate analyses indicated potential mineral associations relevant to public health interpretation. In soybean, contrasting Ca and K loadings occurred in components involving *Dina*, *Safari*, and *GX 2033-16 GZ*, while inverse patterns involving Cu, Fe, and selenium (Se)

were observed among cowpea genotypes, particularly BB 10-4-2-3. These relationships reflect known mineral interactions where high levels of one mineral may inhibit absorption or utilization of another if diets are poorly balanced.⁶⁰ These mineral associations may influence Fe and Zn absorption in young children, supporting balanced formulations and appropriate processing rather than reliance on single-genotype nutrient density.

Cluster analysis grouped the ten cowpea genotypes into four mineral-based clusters and the seven soybean genotypes into three, demonstrating heterogeneity in mineral composition across genotypes. The clusters reflected variation in major and trace minerals relevant to bone development, electrolyte and energy metabolism, immune function, growth, and haemoglobin synthesis in children aged 6–23 months.^{61–66} For both legumes, the cluster patterns closely corresponded with the mineral variability captured by PCA, whose retained PCs demonstrated high internal consistency. As cluster analysis provided limited additional information, dendrograms and detailed cluster-specific characteristics were not presented. PCA and Cronbach's alpha were therefore retained as the primary multivariate approaches for characterizing mineral variability and identifying nutritionally relevant cowpea and soybean genotypes for complementary feeding.

Within this exploratory analytical framework, despite the limited number of genotypes included in the PCA, the laboratory findings indicate patterns relevant to broader nutrition-sensitive agriculture and food-based strategies,^{67,68} by identifying soybean and cowpea genotypes showing comparatively favourable mineral profiles and associations. These findings may inform, rather than confirm, the selection of ingredients for formulating legume-based complementary foods with improved micronutrient composition. Given the potential constraints imposed by anti-nutritional factors such as phytates,⁶⁹ further formulation studies and appropriate household- and community-

level processing methods are needed to enhance mineral bioavailability and maximize nutritional benefit.⁷⁰

Conclusion: Overall, the findings indicate that specific cowpea genotypes (Lukusuzi, Lutembwe, Lunkwankwa, LT 11-3-3-12, LT 4-2-4-1, LT 3-8-4-6, BB 10-4-2-3) and soybean genotypes (SC Saga, Lukanga, Safari, Dina, SG Sentinel, GX 2033-16 GZ, TG 2033-93 GZ) possess favourable mineral profiles with potential to inform the development of nutrient-dense complementary food ingredients for children aged 6–23 months. Subject to further formulation, processing, bioavailability, and efficacy studies, these genotypes may contribute to food-based strategies addressing multiple nutrient gaps. Future research should integrate genotype-by-environment analyses, processing trials, and bioavailability assessments to strengthen the evidence base for nutrition-sensitive legume interventions in Zambia and similar settings. Promoting affordable, accessible, safe, and culturally acceptable utilization of these legumes may have potential to support dietary adequacy among children and other population groups at risk of poor nutritional status

Strengths and Limitations: The study provides robust genotype-specific nutrient evidence from accredited laboratory analyses, forming a strong foundation for subsequent bioavailability assessments, processing studies, complementary food formulation, and feeding trials. However, because the study was laboratory-based, included relatively few genotypes, and relied on calculated energy values without sensory evaluation, bioavailability, or feeding trial data, the

findings should be interpreted cautiously and cannot be directly generalized to complementary food performance, real-world intake, or growth outcomes among children aged 6–23 months.

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Table 1: Cowpea and soybean genotypes analyzed for nutrient composition in relation to complementary feeding applications in Zambia.

S/N	Cowpea	Genotype	S/N	Soybean	Genotype
	genotypes			genotypes	
1.	Lutembwe	Variety	1.	Lukanga	Variety
2.	Lukusuzi	Variety	2.	Safari	Variety
3.	LT 4-2-4-1	Line	3.	Dina	Variety
4.	LT 3-8-4-6	Line	4.	SC Saga	Variety
5.	LT 11-3-3-12	Line	5.	GX 2033-16 GZ	Line
6.	LT 11-5-2-2	Line	6.	TG 2033-93GZ	Line
7.	Bubebe	Variety	7.	SG Sentinel	Variety
8.	BB 14-16-2-2	Line			
9.	BB 10-4-2-3	Line			
10	Lunkwankwa	Variety			

Table 1: Dry-weight proximate composition of cowpea and soybean genotypes, detailing macronutrient and energy profiles with implications for dietary adequacy in complementary feeding of children aged 6–23 months in Zambia.

	Energy	Moisture	Carbohydrates	Proteins	Fats	Crude fiber	Ash
	(Kcal100g ⁻¹)	%	%	%	%	%	%
Cowpea genotypes ^a							
Lutembwe	371	12	48	37	3	7	4
Lukusuzi	375	11	47	38	4	7	4
LT 4-2-4-1	372	12	46	38	4	7	4
LT 3-8-4-6	372	12	47	37	4	7	4
LT 11-3-3-12	371	12	47	38	4	7	4
LT 11-5-2-2	373	12	48	37	4	7	4
Bubebe	368	11	49	37	3	8	4
BB 14-16-2-2	373	11	47	37	4	7	4
BB 10-4-2-3	372	13	45	37	5	8	4
Lunkwankwa	373	12	47	36	4	8	4
Median	372	12	47	37	4	7	4
Range	(371-375)	(11-13)	(46-49)	(36-38)	(3-5)	(7-8)	(4-4)
Soybean genotypes ^a							
Lukanga	473	7	32	40	21	6	5
Safari	472	8	28	43	21	6	6
Dina	473	9	28	43	21	6	6

SC Saga	477	8	29	41	22	6	6
GX 2033-16 GZ	466	8	29	44	19	6	6
TG 2033-93GZ	473	8	29	42	21	6	6
SG Sentenel	481	8	27	43	22	5	5
Median	473	8	29	43	21	6	6
Range	(466-481)	(7-8)	(27-32)	(40-44)	(19-22)	(5-6)	(5-6)

^aP-Value for genotypes > 0.05

Table 1: Mineral composition of cowpea and soybean genotypes (ppm; mg kg⁻¹) relevant to feeding of children aged 6–23 months in Zambia.

	Major minerals					Selected trace elements			
	Ca	K	Mg	Na	P	Cu	Fe	Zn	Se
Cowpea genotypes ^a									
Lutembwe	1436.1	19505.3	3235.7	384.6	4173.7	6.2	62.1	29.5	<0.1
Lukusuzi	1475.1	23549.0	3403.8	830.7	4773.9	6.2	72.6	31.4	<0.1
LT 4-2-4-1	1501.8	22762.3	3279.1	685.2	4500.0	6.9	61.1	33.1	<0.1
LT 3-8-4-6	1173.7	21688.8	3282.9	502.5	3068.9	6.8	58.4	34.4	<0.1
LT 11-3-3-12	1913.8	24023.9	3673.8	636.8	4630.6	5.5	60.4	32.5	<0.1
LT 11-5-2-2	1287.7	22154.4	3265.7	400.0	3228.0	5.9	81.2	32.4	<0.1
Bubebe	1448.7	20370.6	3139.1	334.4	2824.5	6.6	152.9	30.6	<0.1
BB 14-16-2-2	802.0	15090.1	2351.0	288.7	2443.2	4.5	58.9	21.5	<0.1
BB 10-4-2-3	1145.7	19498.6	2924.4	61.9	2358.5	5.3	518.9	23.6	<0.1
Lunkwankwa	942.5	19237.1	2990.1	253.7	3960.2	7.5	64.7	32.9	<0.1
Median	1312.7	20788.0	3154.6	437.9	3596.2	6.1	119.1	30.2	-

Range	802.0- 1913.8	15090.1- 24023.9	2351.0- 3673.8	61.9- 830.7	2358.5- 4773.9	4.5- 7.5	58.4- 518.9	21.5- 34.4	-
Soybean Genotypes ^a									
Lukanga	6640.0	17075.0	2210.0	95.9	5269.5	9.3	88.7	47.4	2.6
Safari	5637.5	17034.0	2250.0	117.4	5988.0	8.4	70.7	48.1	3.1
Dina	5739.0	17260.0	2250.0	171.9	6036.0	8.4	85.6	54.8	2.4
SC Saga	7411.5	16353.0	2415.0	263.2	6075.5	10.0	91.3	56.3	6.3
GX 2033-16 GZ	7105.5	16982.0	2220.0	264.3	5832.0	7.6	90.0	52.3	1.3
TG 2033-93GZ	6595.0	15750.0	2165.0	198.4	5765.0	8.5	107.0	49.5	2.6
SG Sentenel	6505.5	15301.0	2055.5	179.8	4709.5	7.4	79.8	48.6	3.5
Median	6519.1	16536.4	2223.6	184.4	5667.9	8.5	87.6	50.1	3.1
Range	5637.5- 7411.5	15301.0- 17260.0	2055.5- 2415.0	95.9- 264.3	4709.5- 6075.5	7.4- 10.0	70.7- 107.0	47.4- 56.3	1.3- 6.3

Ca: Calcium, K: Potassium, Mg: Magnesium, Na: Sodium, P: Potassium, Cu-Copper, Fe-Iron, Zn-Zinc, Se-Selenium, ^aP-value for genotypes >0.05

Table 4: Principal Component Analysis of mineral loading patterns in cowpea & soybean genotypes, highlighting mineral synergies and antagonistic interactions with implications for feeding of children aged 6–23 months in Zambia.

Mineral	Principal Components													
	Cowpea								Soybean					
	1	2	3	4	5	6	7	8	1	2	3	4	5	6
Cu	0.2	-0.6	0.5	0.1	0.3	0.4	0.1	-0.3	0.4	-0.1	-0.5	0.4	0.0	0.2
Fe	-0.2	0.5	0.6	0.3	0.4	-0.0	0.0	0.3	0.2	0.4	0.3	0.7	-0.3	0.2
Zn	0.4	-0.3	0.2	0.1	-0.3	-0.2	-0.2	0.7	0.4	0.0	0.2	-0.3	-0.2	0.7
Se	-	-	-	-	-	-	-	-	0.3	0.1	-0.6	-0.3	-0.2	-0.1
Ca	0.4	0.5	-0.0	-0.4	-0.2	0.7	-0.2	0.1	0.3	0.5	-0.0	0.1	0.7	-0.2
K	0.4	0.3	0.1	0.3	-0.1	-0.3	-0.6	-0.5	0.1	-0.6	0.3	0.2	0.5	0.2
Mg	0.4	0.2	0.2	-0.1	-0.2	-0.3	0.7	-0.2	0.5	-0.2	-0.1	-0.0	0.1	-0.2
Na	0.3	0.1	-0.5	0.7	0.2	0.2	0.2	0.1	0.3	0.4	0.4	-0.3	0.1	-0.2
P	0.4	-0.0	-0.2	-0.4	0.7	-0.2	-0.1	0.2	0.4	-0.3	0.3	0.0	-0.4	-0.6
Eigenvalue (λ)	4.8	1.3	1.0	0.5	0.3	0.1	0.0	0.0	3.7	2.3	1.4	0.9	0.5	0.3
Total Variance%	60.3	16.2	12.1	6.2	3.7	1.2	0.1	0.1	41.5	25.3	15.2	10.1	5.5	2.4

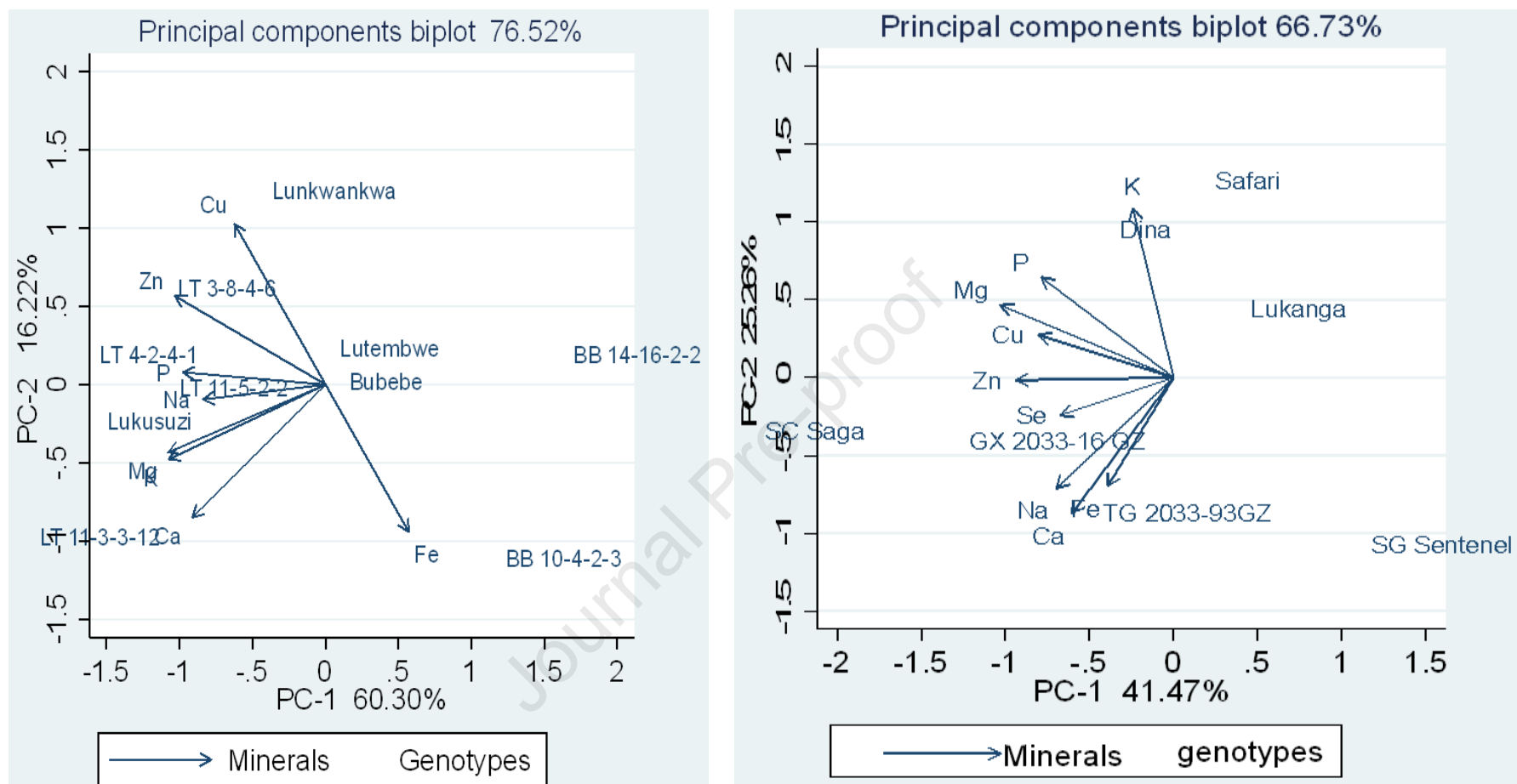
Cumulative Variance%	60.3	76.5	88.7	94.9	98.6	99.8	99.9	100	41.5	66.7	82.0	92.1	97.6	100
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Cu: Copper, Fe: Iron, Zn: Zinc, Se: Selenium, Ca: Calcium, K: Potassium, Mg: Magnesium, Na: Sodium, P: Phosphorus

								1.0	Cu	1.0									
							1.0	0.6	Fe	-0.1	1.0								
						1.0	0.5	0.2	Zn	0.7*	-0.4	1.0							
				1.0	-0.1	-0.2	0.3		Se	0.0	0.0	0.0	1.0						
			1.0	0.1	0.4	0.8	0.5		Ca	0.2	0.0	0.3	0.0	1.0					
		1.0	-0.3	-0.4	-0.1	-0.3	0.3		K	0.1	-0.1	0.5	0.0	0.9	1.0				
	1.0	0.5	0.1	0.1	0.5	0.0	0.5		Mg	0.2	-0.3	0.6	0.0	0.8*	0.9	1.0			
	1.0	0.1	-0.5	0.6	-0.1	0.7	0.6	-0.1	Na	0.1	-0.3	0.6	0.0	0.6*	0.8	0.8*	1.0		
1.0	0.3	1.0*	0.4	0.1	0.0	0.7	0.1	0.4	P	0.3	-0.2	0.5	0.0	0.7*	0.7*	0.8*	0.6*	1.0	
P	Na	Mg	K	Ca	Se	Zn	Fe	Cu		Cu	Fe	Zn	Se	Ca	K	Mg	Na	P	
Soybean									Cowpea										

P: Phosphorus, Na: Sodium, Mg: Magnesium, K: Potassium, Ca: Calcium, Se: Selenium, Zn: Zinc, Fe: Iron, Cu: Copper *p< 0.05

Fig. 1: Spearman's correlations showing mineral interaction patterns among cowpea & soybean genotypes with implications for nutrient bioavailability in complementary feeding of children aged 6–23 months.



Key: Cu: Copper, Fe: Iron, Zn: Zinc, Ca: Calcium, Mg: Magnesium, Na: Sodium, P: Phosphorus, K: Potassium, Se: Selenium

Fig. 2: Multivariate spatial distribution of cowpea & soybean genotypes highlighting mineral composition patterns with implications for complementary feeding of children aged 6–23 months in Zambia.