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VARIATIONS IN THE MINERAL CONCENTRATION AMONG DIFFERENT POTATO (*SOLANUM TUBEROSUM* L.) ACCESSIONS

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ABSTRACT: A collection of one hundred tetraploid potato accessions was evaluated in relation to the concentrations of nine minerals (Fe, Zn, Cu, Mn, Ca, Mg, S, P and K) required for human nutrition. Large amount of variability for selected minerals was observed in the germplasm. Significant positive correlations ($P < 0.001$) were found between Fe-Cu (r^2=0.41), Zn-Cu (r^2=0.46), Mn-K (r^2=0.51) and Ca-Mg (r^2=0.40), whereas significant negative correlation ($P < 0.001$) was observed between P-K (r^2= -0.36). The first two principle components accounted 42.72% of the total observed variations. Unweighted pair group method with arithmetic mean (UPGMA) based clustering distinguished the potato accessions into different groups based on Gower distances. These results will be useful for developing biofortified potato varieties using traditional and modern breeding methods. Moreover, variations in the mineral contents within the potato germplasm could also be used for the identification of molecular markers associated with mineral concentrations through genome wide association studies (GWAS).

KEYWORDS: Potato, germplasm diversity, minerals, biofortification, iron, zinc

INTRODUCTION

To maintain optimal health, human body requires a diverse as well as balanced diet containing both micronutrients and macronutrients. The micronutrients are vitamins and minerals, which are required in minute quantities but are essential for human health. Minerals are categorised into two sub-groups; 1) macro-minerals and 2) micro-minerals (trace minerals). The macro-minerals involves calcium (Ca), magnesium (Mg), sulphur (S), phosphorous (P), potassium, (K) sodium (Na) and (Cl), while the micro-minerals involves iron (Fe), zinc (Zn), copper (Cu), manganease (Mn), cobalt (Co), boron (B), molybdenum (Mo), chromium (Cr) and fluoride (F) (Mohammad *et al.,* 2017). These minerals play important roles in human body to maintain a good health and there deficiencies may lead to serious health issues (reviewed by Mohammad *et al.,* 2017; Sun *et al.,* 2021). The micronutrient malnutrition is a global issue, which is highly common in developing countries. Globally about 2 billion people are suffering from it and around one third out of these 2 billion people live in India (Kotecha 2008). Among all the minerals, the deficiency of iron and zinc is widespread. Iron deficiency cause anemia. Worldwide, anemia affects 1.62 billion individuals and highly prevalent in children and adolescent girls (McLean *et al.,* 2009). The largest portion of global malnourished population live in India. The Indians take a daily diet that is far from satisfactory and less than 50% of recommended dietary allowance (RDA) is taken by over 70% of Indian population (Kotecha 2008). In India, about 69% children below age 5 years and are affected by anemia (Kotecha 2008).

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During the last decade, several scientific and technological attempts were made to reduce the micronutrient malnutrition. The governments has developed several new policies with an aim to reduce hidden huger throughout the world (Tulchinsky 2015). Most of these policies are based upon the use of diversified diet, food supplements, food fortification and crop biofortification. All these have their own merits and demerits. However, among all crop biofortification has emerged as a sustainable solution for this serious issue. In the recent years, a large number of studies were conducted to develop biofortified crops by using different approaches (Garg *et al.,* 2018; Singh *et al.,* 2021a). Potato is world's most consumed non-grain staple food crop. It can produce more food per unit time, water and area compared to other major food crops (Azimuddin *et al.,* 2009). It is a modest source of nutrients and previous studies showed that the high bioavailability of Fe and Zn from potato tubers compared to cereals (Andre *et al.,* 2015; Jongstra *et al.,* 2020). The presence of genetic variations for mineral content in the potato germplasm suggest that nutrient rich potato cultivars have potential supply a much higher portion of RDA for various minerals than the present potato cultivars (Sharma *et al.,* 2017; Subramanian *et al.,* 2017; Singh *et al.,* 2021b). Potato appears as an ideal crop for biofortification as it is a versatile crop with an enormous industrial demand and high consumer's acceptability. The biofortified potatoes can be developed by using agronomic approaches, conventional and modern breeding. The generation of biofortified crops by breeding is considered as the most suitable way as crops developed by this method produce self-biofortified seeds. Therefore, once a biofortified variety developed it can be used for years. However, the knowledge of nutrient rich accessions in the crop germplasm is the most basic need to develop biofortified crops. In this study, one hundred highly diverse potato accessions were studied for nine mineral concentrations.

MATERIALS AND METHODS

One hundred highly diverse tetraploid potato accessions used in the current study were obtained from Indian Council of Agricultural Research - Central Potato Research Institute (ICAR-CPRI), Shimla, India. Seed tubers were sown in November 2019 at ICAR-Central Potato Regional Station (CPRS), Modipurum, India. The plot size was 3.6m^2 and the row to row and plant to plant spacing was 60 X 20 cm. The crop was raised under uniform agronomic practices. The tubers were harvested after 90 days from the date of planting. The soil of the experimental field has pH 6.69 in reaction and the available mineral nutrients were in sufficient amounts. The standard protocols were followed for field sampling of potato tubers and for preparation of oven-dried and milled potato tuber samples (Porras *et al.,* 2014). Seven even sized unblemished tubers of each accession were selected for this study. As the tubers of some accessions were small and irregular in shape. Therefore, whole tubers were used for the mineral analysis. The tubers were thoroughly washed with tap water to remove the surface soil, then by 0.1% nitric acid and then with double distilled water. Later the air-dried tubers were chopped with high quality stainless steel knives and were kept in hot air oven at 70°C and then powdered samples were used for the estimation of mineral concentrations after acid digestion. Acid digestion was performed with nitric acid in analytical grade nitric acid in microwave digester (CEM, MARS6™) and mineral analysis was performed using inductively coupled-plasma mass spectrometry (iCAP 7000 Series ICP-OES, Thermo Scientific™). The concentration of phosphorus was estimated by using vanadate-molybdate (yellow) method using spectrophotometer (UV-1700 PharmaSpec Shimadzu™).

Data of the mineral concentrations was used to calculate Pearson's coefficient of correlation among the minerals. The correlation analysis was performed in R software using the package 'psych'. Box plots were used to illustrate the extent of diversity of tuber mineral content among the 100 potato accessions. The principal component of analysis was computed after data transformation and a biplot was obtained using correlation matrix to illustrate the relationships among the traits. A cluster analysis of the mineral concentrations was performed using Gower distances and a dendrogram was constructed.

RESULTS AND DISCUSSION

Variation in the mineral concentrations

There were larger variations in the mineral concentrations in the potato germplasm studied. The descriptive statistics (range, mean, standard deviation (SD) and the coefficient of variance) for the mineral content are presented in the table-1. The concentrations Fe varied from (23.60-76.90 mg kg^{-1}), Zn from (6.80-36.00 mg kg^{-1}), Cu from (3.80-23.10 mg kg^{-1}), Mn from (7.20-28.40 mg kg^{-1}), Ca from (0.02-0.10%), Mg from (0.04-0.29%), S from (0.11-0.49%), P from (0.13-0.39%) and K from (0.48-2.61%) on dry weight basis. The substantial differences among the accessions for different mineral concentrations were observed and are presented as boxplots in figure 1. Each boxplot represents five statistical bars from bottom to top; 1) the lowest observed value, 2) lower quartile, 3) median, 4) upper quartile, 5) highest observed value. The distributions of concentration of these minerals in the germplasm are depicted in figure 2. There were significant differences in the mineral content within the whole set of 100 potato accessions tested in this study, suggesting that for these minerals there is some genetic potential, which can be used to enhance mineral content in the potato tubers.

The accessions with highest Fe content (76.90 mg kg^{-1} in JEX/A-274, 76.40 mg kg^{-1} in JEX/A-413, and 74.90 mg kg^{-1} in JEX/A-199 and JEX/A-920) were belongs to *Solanum tuberosum* ssp. *andigena*. The mean Fe content (54.19 mg kg^{-1}) in potato germplasm was higher than the mean Fe content (43.87 mg kg^{-1}) observed in the cultivated Indian potato varieties. Sharma *et al*. (2017) reported a mean Fe concentration of 43.94 mg kg^{-1} in the whole tubers of popular Indian potato varieties, while Andre *et al*. (2007) reported an average Fe concentration of 54.95 mg kg^{-1} in Andean potato cultivars. In this study, the mean Zn concentration (22.56 mg kg^{-1}) observed was slightly higher than previously reported (20.40 mg kg^{-1}) by Haynes *et al*. (2012) and there were 40 accessions with Zn concentration more than mean. Dalamu

Table 1. Summary of variations in terms of mean, standard deviations (SD), and ranges in the whole tuber mineral content in 100 potato accessions

	Fe (ppm)	Zn (ppm)	Cu (ppm)	Mn (ppm)	Ca (%)	Mg (%)	S (%)	P (%)	K (%)
Min	23.60	6.80	3.80	7.20	0.02	0.04	0.11	0.13	0.48
Max	76.90	36.00	23.10	28.40	0.10	0.29	0.49	0.39	2.61
Mean	51.71	22.56	10.27	14.93	0.04	0.12	0.24	0.23	1.37
SD	14.22	6.02	4.03	6.16	0.02	0.05	0.08	0.06	0.69
CV	27.50	26.67	39.26	41.25	44.91	45.15	34.52	25.23	49.98

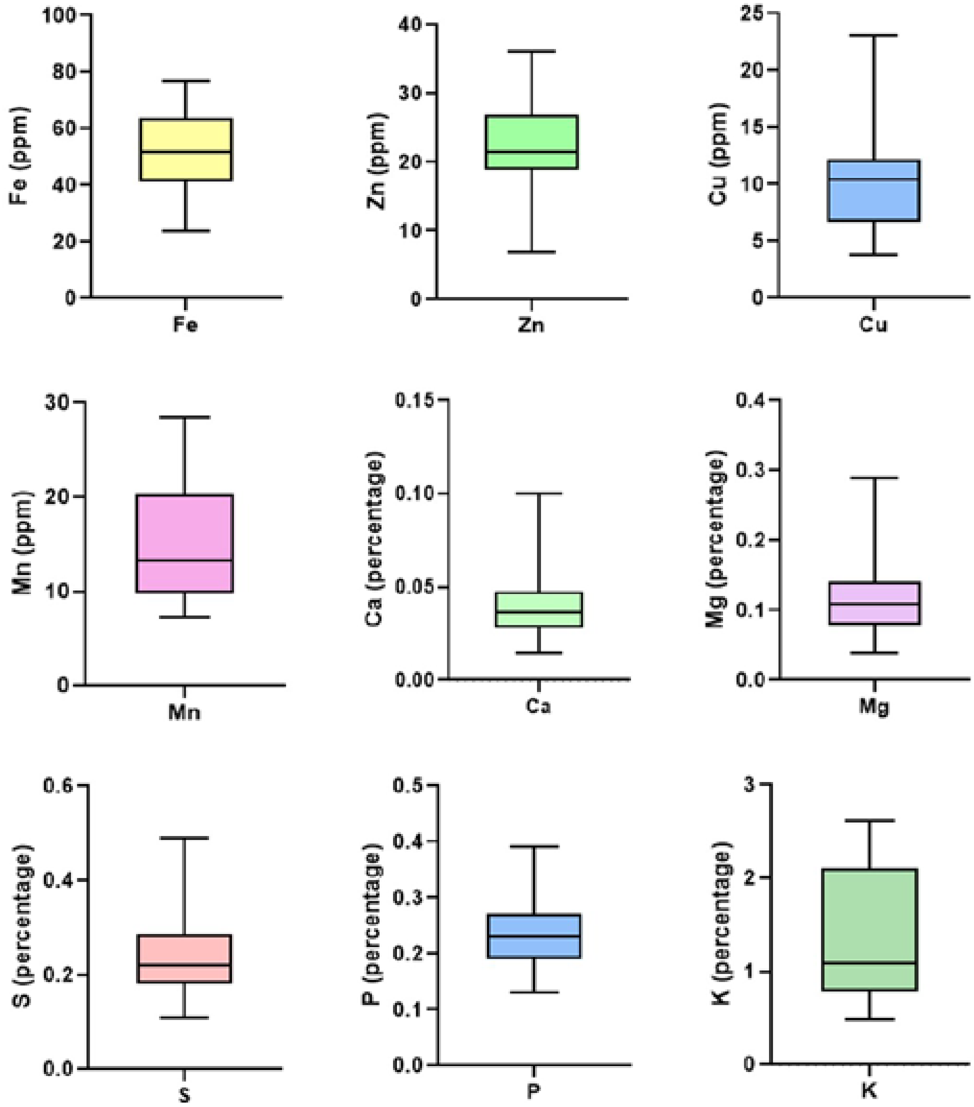

Figure 1. Box plots representing variations in potato tuber mineral (Fe, Zn, Cu, Mn, Ca, Mg, S, P and K) content

et al. (2017, 2019) reported similar ranges for Zn content in potato tubers. Highest Zn concentration was observed in CP 3191. There were significant differences in the Cu content within the germplasm studied. The Cu content observed in the current study ranged from (3.80-23.10 mg kg^{-1}), while in the previous studies values reported for Cu content in potato ranged from 2.6 - 10.8 mg kg^{-1} (Subramanian *et al*., 2017), 4.43 - 22.47 mg kg^{-1} (Singh *et al*., 2020) and 5.13 - 21.06 mg kg^{-1} (Sharma *et al*., 2017). The concentration of Mn varied from 7.20 mg kg^{-1} in CP 3144 to 28.40 mg kg^{-1} in CP 3245 and CP 1679. The Mn concentration above 20 mg kg^{-1} were observed in 25% accessions. We observed a mean Mn concentration of 14.93 mg kg^{-1} was higher than previously reported by Subramanian *et al*. (2017), but less than the mean Mn concentration reported by Sharma *et al*. (2017) in whole tubers. There were significant variations among the accessions for secondary as well as macro elements (Ca, Mg, S, P and K) (Table 1).

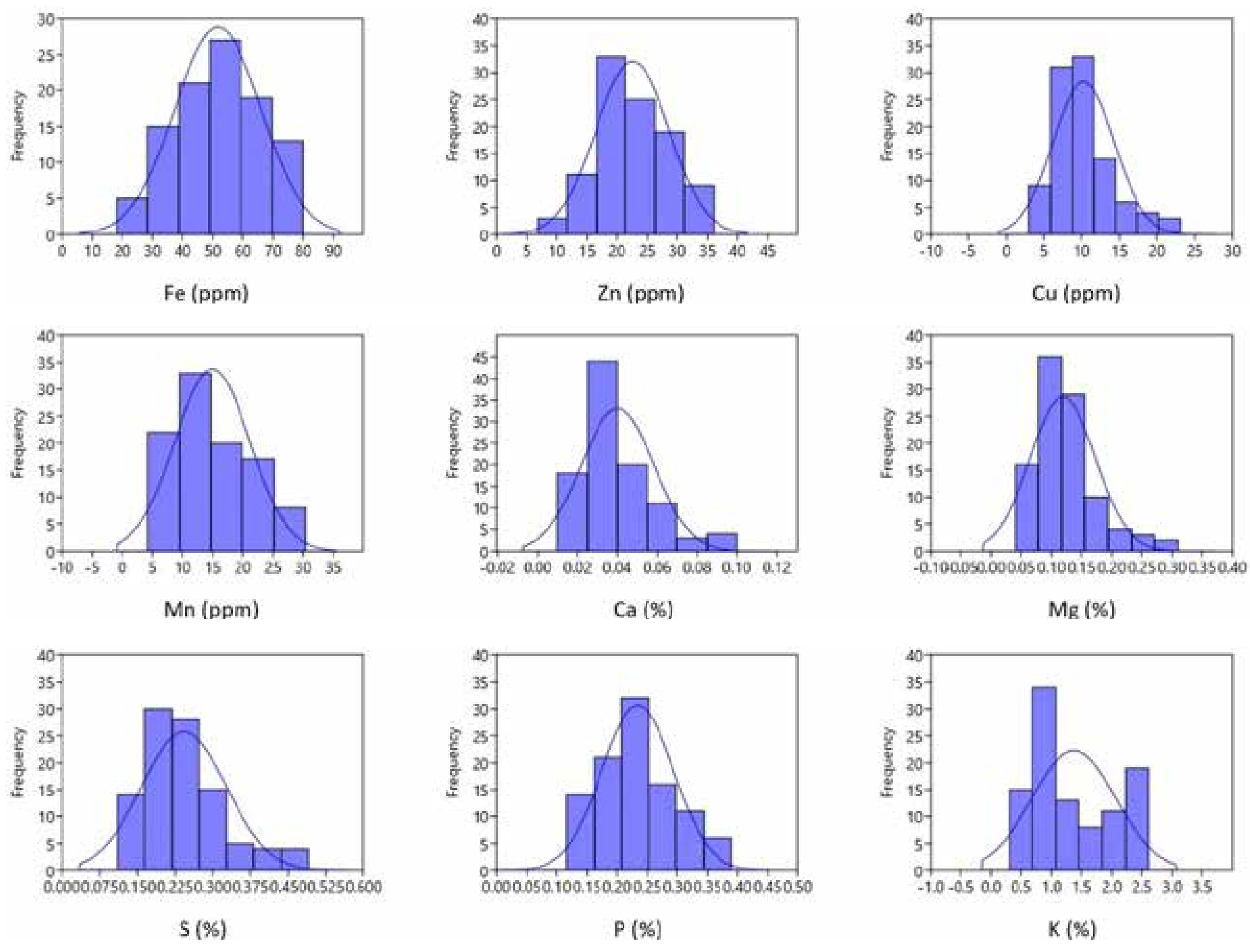

Figure 2. Distribution of mineral concentrations in the tubers of 100 tetraploid potato accessions

Relationships among the traits

In this study, correlations between different minerals were studied and it was observed that Fe, Zn and Cu were positively correlated (Figure 3). Positive and significant correlations were found between Fe-Zn (r^2 = 0.27, p = 0.01), Fe-Cu (r^2 = 0.41, p = 0.001), Zn-Cu (r^2 = 0.46, p = 0.001), Mn-K (r^2 = 0.51, p = 0.001) and Ca-Mg (r^2 = 0.40, p = 0.001). Some other weak to moderately positive correlations were observed between the traits studied. Previous studies also reported positive correlations between these micronutrients in potato (Dalamu *et al.*, 2019; Singh *et al.*, 2020). Significant positive correlations were also observed between P-Fe (r^2 = 0.31, p = 0.01) and P-Zn (r^2 = 0.27, p = 0.01). Earlier, Abebe et al. (2012) also reported positive correlations between P-Fe and P-Zn concentrations in the potato tubers. A negative and significant correlation was observed between K and P (r^2 = –0.36, p = 0.001). The UPGMA based clustering by utilizing the Gower dissimilarity matrix confirms the existence of wide diversity among the potato germplasm studied (Figure 4). The potato germplasm was grouped into three larger clusters, which would be helpful to select and breed the accessions from different groups to unite the desirable genes.

Principal component analysis

The vector view of genotype trait biplot provided a brief summary of the

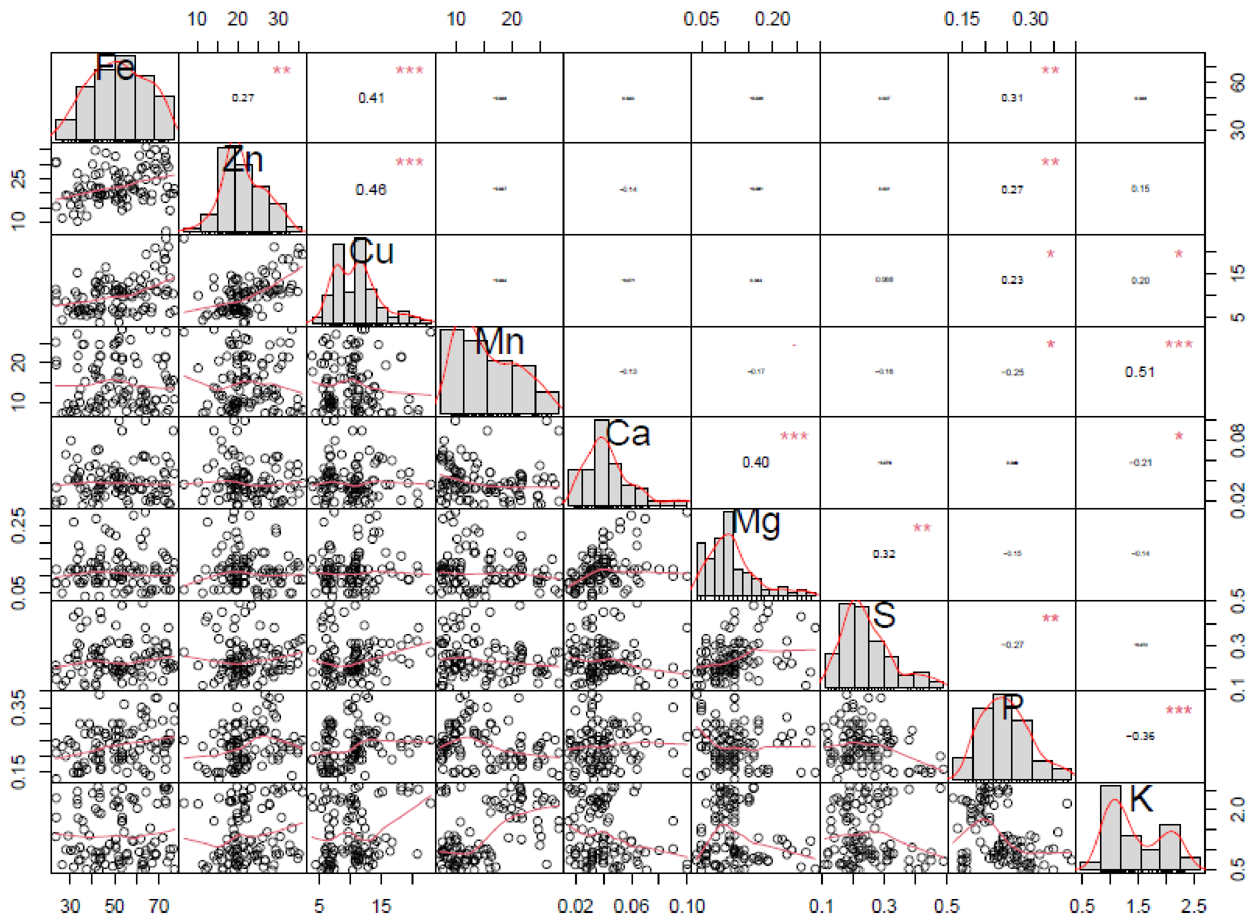

Figure 3. Pearson's correlation matrix for tuber mineral (Fe, Zn, Cu, Mn, Ca, Mg, S, P and K) content of potato accessions.
Note: ***, significant at P < 0.001; **, significant at P < 0.05; *, significant at P < 0.05

inter relationships among the mineral concentrations (Figure 5). The cosine of the angle between different vectors of any traits approximates the correlation coefficient between them (Akçura 2011; Akcura and Kokten 2017). The angles between the different mineral vectors suggest that Fe, Zn, Cu and P concentrations were positive and significantly correlated. Likewise, significant positive correlations were found between Mg–Ca and Mn–K, respectively. Other associations among the traits were not highly significant. The PC1 and PC2 explained 22.23% and 20.49% of the total variations. PC1 was positively loaded with Fe, Zn, Cu and P and negatively loaded by Mn, Mg and K. In PC2, K and Mn were dominating variables on the positive side, whereas Ca and Mg were the negatively dominating variables. The first three principal components showed 59.32% of the total variation. The details of the eigenvectors, eigenvalues and percent variation explained by first five principal components are provided in table 2.

CONCLUSION

Potato is a staple crop of many countries. India is the second largest potato producing country, after China. The information about the diversity present in potato germplasm will help in the screening of germplasm with desirable traits. The present study provides

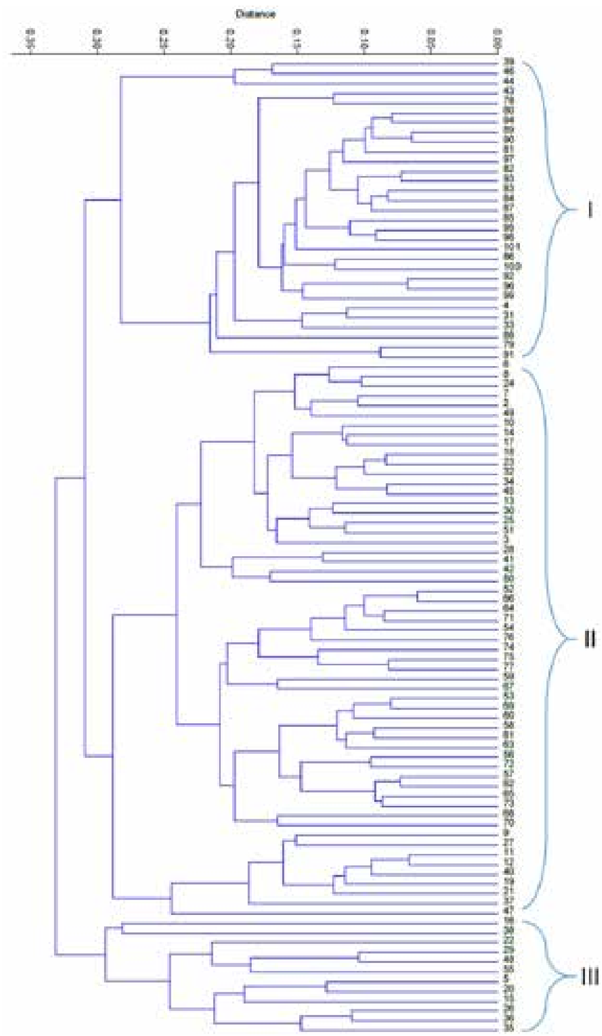

Figure 4. UPGMA based clustering for mineral content in 100 tetraploid potato accessions

Table 2. The details of loadings, eigenvalues, individual and cumulative percentages of variation explained by the first five principal components after accessing the nine mineral traits in 100 tetraploid potato accessions.

Variables	PC 1	PC 2	PC 3	PC 4	PC 5
Fe (ppm)	0.63852	0.14812	0.18301	0.14271	0.64711
Zn (ppm)	0.61292	0.33011	0.22477	-0.06395	-0.46048
Cu (ppm)	0.66902	0.27593	0.34559	0.08096	-0.11766
Mn (ppm)	-0.38949	0.64073	0.11123	0.32583	0.23925
Ca (ppm)	0.06453	-0.63886	0.13361	0.65362	0.00395
Mg (ppm)	-0.05661	-0.52914	0.66887	0.25450	-0.13103
S (ppm)	0.02371	-0.19570	0.67522	-0.59601	0.20086
P (ppm)	0.74270	-0.06140	-0.45054	0.10717	-0.00434
K (ppm)	-0.24185	0.70475	0.39139	0.29945	-0.13117
Eigenvalue	2.00069	1.84446	1.49316	1.08555	0.77664
Variance (%)	22.23	20.49	16.59	12.06	8.63
Cumulative variance	22.23	42.72	59.32	71.38	80.01

the information about the diversity for mineral concentrations in the potato tubers. Our results suggest that a broad range of variations exist in the concentration of different minerals critical for human health in the potato germplasm. These findings would be a useful resource for developing nutrient-rich potato varieties. Development of biofortified potato cultivars can play a crucial role to overcome the mineral deficiencies.

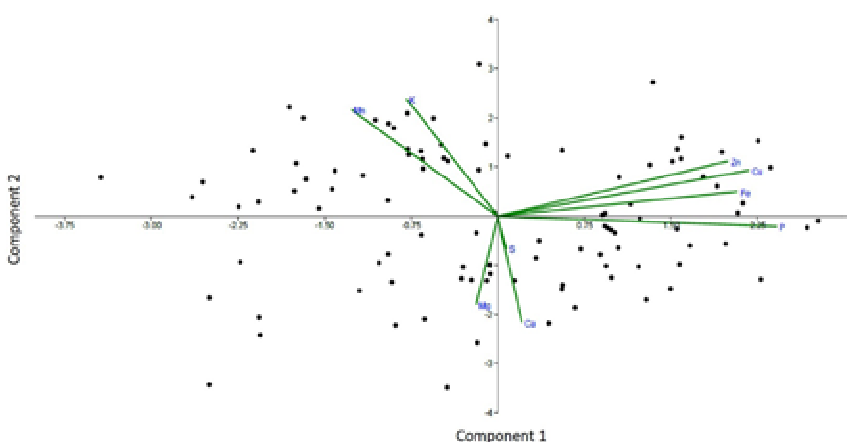

Figure 5. Biplot of Principal component analysis (PCA) of mineral (Fe, Zn, Cu, Mn, Ca, Mg, S, P and K) concentrations present in the potato tubers of 100 potato accessions.

The variations observed in this study can be used to identify the molecular markers associated with mineral concentrations. This study provides the basic insights in relation to the presence of variations in the mineral content. However, there is a need to perform more such experiments under different geographical locations. The findings of this study would be very advantageous for potato breeders, who are breeding to develop nutrient rich potato cultivars.

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CONFLICTS OF INTEREST

The authors confirm that this manuscript has no conflicts of interest

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