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Research Article

Genetic Diversity based on Cluster and Principal Component Analyses in Potato (*Solanum Tuberosum L.)* for Yield and Processing Attributes

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ABSTRACT

Potato (*Solanum tuberosum L.*) is a versatile food crop and a source of cheap human diet in many countries. It can be used as fresh products and commercially processed foods such as French fries and chips. Research efforts carried out in Ethiopia related to processing quality were limited in their scope of quality parameters considered. Therefore, this experiment was conducted during the main rainy season of 2017 at Holetta with the objectives of the nature and magnitude to know genetic diversity and the characters contributing in potato genotypes and also to screen out genetically diverse parents for developing high yielding and quality potato by using cluster and principal component analysis. A total of 24 potato genotypes were evaluated for 23 quantitative and six qualitative traits in randomized complete block design with three replications. The first eight principal components accounted 90.26% for the observed variations among 24 potato genotypes. Of these, the first, the second and the third principal components constituted 28.69%, 18.74% and 13.00% of the variation, respectively. The genetic distances among the 24 potato genotypes ranged from 3.40 to 11.80 and the genotypes were grouped into eight clusters based on quantitative and qualitative traits. Cluster II consisted of 25%, Cluster IV, I, III contained 20.83%, 16.67% and 12.5% of genotypes, respectively, while Cluster VI, VII and VIII each consisted of one genotype. In conclusion, genotypes grouped under Cluster II and VIII worth further evaluation to obtain genotypes with highest total tuber yield, specific gravity of tuber, dry matter content, total starch content, acceptable tuber physical and frying quality with other desirable traits.

Keywords: Genetic diversity; Clustering; Principal component analysis; Tuber quality

INTRODUCTION

The potato plant is a versatile food crop and a cheap source of food in many countries. It is the third most important food crop in terms of consumption in the world after rice and wheat. The genetic diversity of potatoes Solanum Section Petota (Solanaceae) may be grouped in wild and cultivated potatoes. The cultivated potatoes *Solanum tuberosum* are tetraploid (2n=4x= 48), while the native are highly diverse, diploids (2n=2x=24), triploids (2n=3x=36), tetraploids (2n=4x=48), pentaploids (2n=5x=60) and hexaploids (2n=6x=72). For a successful breeding program, the presence of genetic diversity and variability play a vital role [1]. Information on genetic diversity in elite germplasm is essential for identifying promising lines for trait of interest and estimating genetic distinctness among parents. Selection of genetically diverse parents is mandatory for exploitation of trangressive segregation. Vast genetic distance among parents is prerequisite for securing useful heterosis in progeny. Diversity in plant genetic resources provides opportunity for plant breeders to develop new and improved cultivars with desirable characteristics, which include both farmer-preferred traits (high yield potential, large seed, etc.) and breeder-preferred traits (pest and disease resistance and photosensitivity, etc.). Genetic diversity facilitates breeders to develop varieties for specific traits like quality improvement and tolerance to biotic and abiotic stresses [2].

Cluster analysis and principal component analysis (PCA) are most frequent genetic diversity assessing methods while securing relative basic differences between them. Cluster analysis is a

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classification method, which is used to arrange a set of cases into clusters. The aim of set cases within a cluster is more similar to each other and helps to researchers to give summary information on data [3]. Cluster analysis is commonly used in social, medical and agricultural sciences. In addition, cluster analysis is being used to exposing of similarity and diversity. In Ethiopia, a number of improved potato varieties have been released by different research centres and institutions. However, the released varieties have not satisfied the consumer for process making. Therefore, the present study was conducted to find out the nature and magnitude of genetic diversity and the characters contributing in potato genotypes for tuber quality, yield and yield related traits and also to screen out genetically diverse parents for developing high yielding and quality potato by using cluster and principal component analysis [4].

MATERIALS AND METHODS

Description of the study area

Matter The study was conducted at Holleta Agricultural Research Center (HARC), Ethiopia. Holetta Agricultural Research Center is located at 090 00'N, 380 30'E at an altitude of 2400 m.a.s.l. The annual rainfall of 1041.4 mm, mean relative humidity of 58.70%, and mean maximum and minimum temperature of 21.70 0C and 6.70 0C, respectively. The main rainy season is from June to September, which account for 70% of the rainfall while the remaining thirty percent is from February to April. The soil of the center is red Nitosol, which is characterized with an average organic content of 1.8%, Nitrogen 0.17%, pH 5.24, and phosphorus 4.55 ppm [5].

Experimental materials and design

A total of 24 potato genotypes were used for the experiment. These included 21 genotypes selected from the germplasm introduced from International Potato Center (CIP) and three released varieties (Table 1). The 24 genotypes were planted at Holleta Agricultural Research Center experiment station during the main cropping season of 2017. The experiment was laid out in randomized complete block design (RCBD) with three replications and each plot was 3.6 m (length) x 4.5 m (width) (16.2 m2 gross plot size) consisted six rows each containing 12 plants and thus 72 plants per plot. The spacing between rows and plants was 0.75 m and 0.30 m, respectively. The spacing between plots and adjacent replications was 1 m and 1.5 m, respectively. Planting was done at June 26, 2017 during the main growing season after the rain commenced and when the soil was moist enough to support emergence and Harvesting was carried out in November 10, 2017 [6].

No.	Accession code	No.	Accession code
1	CIP-396034.268	13	CIP-394611.112
2	CIP-393220.54	14	CIP-392617.54
3	CIP-395017.229	15	CIP-381381.20
4	CIP-392797.27	16	CIP-398180.289
5	CIP-395112.19	17	CIP-.398190.89
6	CIP-399075.7	18	CIP-398190.404
7	CIP-393280.64	19	CIP-391058.175
8	CIP-398098.65	20	CIP-396034.103
9	CIP-393385.39	21	CIP-391046.14
10	CIP-396027.205	22	Belete
11	CIP-393077.159	23	Gudanie
12	CIP-399002.52	24	Dagim

Table 1: List of potato genotypes used for this study.

Data collection

Phenology and growth parameters: Data was recorded for phenology and growth parameters; days to 50% flowering, days to maturity, plant height (cm), average stems number and leaf area index (cm-3).

Yield and yield components: Data was recorded for yield parameters; shoot dry mass weight (g), tubers dry mass weight (g), total biomass weight (g), average tuber number per hill, average tuber weight (g/tuber), tuber size distribution:- small (< 35 mm), medium (35 to 50 mm), and large (>50 mm) size tubers (%), total tuber yield (t ha-1), marketable tuber yield (t ha-1) and unmarketable tuber yield (t ha-1) [7].

Tuber physical and internal quality traits

Geometric mean diameter (Dg) (mm): The sizes of ten randomly selected tubers from each plot were measured as length, width and thickness using digital caliper with an accuracy of 0.01 mm. The geometric mean diameter (Dg) was calculated by using the following equation:

Dg = (LWT) 0.333, Where: L is the length; W is the width and T is thickness of the tuber.

Length to width ratio: Recorded as the ratio of tubers length to width and then expressed in terms ratio.

Sphericity of the tuber (Φ) (%): Tuber sphericity was determined by the following formula as described by Φ= (Dg/ L) ×100 [8].

Where, Φ is sphercity of the tuber, Dg is geometric mean diameter and L is length

Surface area (S) (mm^2): Tubers surface area was determined according to the following formula: $S = \pi Dg 2$

Where, S is surface area and Dg is geometric mean diameter

Tuber shape: This was described eight types of tuber shape, which was transformed into numerical scores from 1 to 8, where

1 = compressed, 2= round, 3=ovate, 4=obovate, 5= elliptic, 6=oblong, 7=long-oblong and 8=elongate.

Eye depth: Described five types of tuber eye depth, which was transformed into numerical scores from 1 to 5, where 1= Protruding, 2 = Shallow, 3 = Medium, 4 = Deep, and 5 = very deep.

Tuber skin color: This was assessed visually according to a color card from 1–9 scale, where 1= white-cream, 2= yellow, 3=orange, 4=brown, 5=pink, 6=red, 7=red-purple, 8=purple and 9= blackish. Tuber skin color i.e., white or red and others were noted by visual observation immediately after harvesting.

Tuber flesh color: It was evaluated visually using the color card from 1–8, where 1=white, 2=cream, 3=yellow (bright), 4=yellow, 5=yellow (intense), 6=red, 7=purple, 8=violet.

Chips and French Fries Color: Sample preparation uniform-sized (100–150 g) tubers were peeled and collected in tap water. Sliced using potato slices and collected in tap water. The slices were blotted on paper towels to remove the free water. Before frying, the frying oil was heated for about 10 to 15 min until the required temperature of 176°C reached and it was measured using thermometer. Slices with a total weight of 700 g were fried in liquid sunflower oil at a temperature of 176–180 °C using electronic deep fat fryers until bubbling ceased (3 - 4 min). It was measured using a standard color chart having scale ranging from grade 1 to 5 (1 = the lightest color (white to cream), 2 = light tan, 3 = dark tan, 4 = brown and 5 = dark brown, chip and French fries color between grade 1 and 2 is commercially acceptable [9].

Specific gravity of tubers (Sg) (gcm-3): It was determined using the weight in air/weight in water method. Five kilogram tubers of all shapes and sizes were randomly taken from each plot. The selected tubers were washed with water. First it was weighed in air and then re-weighed suspended in water and the specific gravity determined according to the following formula.

$$\text{Specific gravity} = \frac{\text{Weight in air}}{\text{Weight in air} - \text{Weight in water}}$$

Dry matter content (%): The total dry matter content (DMC) was calculated according. Five tubers of each treatment were chopped (about 500 g total) into small 1-2 cm cubes. They were mixed thoroughly and two sub-samples of 200 g each were taken. The exact weight of each sub-sample was recorded as fresh weight. Subsequently, each sub-sample was placed in an oven set at 80°C for 48 hours and dried until constant weight. Each subsample were weighed immediately and recorded as dry weight. The dry matter content for each sub-sample was then computed with the following formula.

$$\text{Dry matter content (\%)} = \frac{\text{dry weight}}{\text{fresh weight}} * 100$$

Total starch content (g/100g): This was estimated from dry matter. Starch content (%) = 17.55 + 0.891 * (tuber dry weight% – 24.182). where dry matter was determined as indicated above it was measured from tubers of the five randomly selected plants to be used for tuber dry mass estimation was sliced and kept in oven at 80°C for 48 hours and weighted after cooling in room temperature

Data analysis

Genetic distance and clustering

Genetic distance of 24 potato genotypes was estimated using Euclidean distance (ED) calculated from quantitative and qualitative traits after standardization (subtracting the mean value and dividing it by the standard deviation) as established as follows:

$$EDjk = \sqrt{\sum_{i=1}^{n}(Xij - Xik)}^{2}$$

Where;

EDjk = distance between genotypes j and k; xij and xik = phenotype traits values of the ith character for genotypes j and k, respectively; and n = number of phenotype traits used to calculate the distance. The distance matrix from phenotype traits was used to construct dendrogram based on the Unweighted Pair-group Method with Arithmetic Means (UPGMA). The results of cluster analysis were presented in the form of dendrogram. In addition, mean ED was calculated for each genotype by averaging of a particular genotype to the other 23 genotypes. The calculated average distance (ED) was used to estimate which genotype(s) is closest or distant to others [10].

Principal Component Analysis

Principal component analysis (PCA) was computed to find out the characters, which accounted more to the total variation. The data were standardized to mean zero and variance of one before computing principal component analysis. The principal component based on correlation matrix was calculated using SAS software. According to Gutten's lower bound principle that eigenvalues <1 should be ignored.

RESULTS AND DISCUSSION

Cluster analysis

The genetic distances result of 276 pair of potato genotypes are presented in Table 2. The highest genetic distances (Euclidean distance) was between CIP-396027.205 and CIP-392617.54 (11.78) followed by between CIP-396027.205 and CIP-394611.112 (11.72), between CIP-398098.65 and CIP-396027.205 (11.60) and between CIP-396027.205 and Belete (11.60), while the lowest genetic distances (Euclidean distance) was between CIP-395017.229 and CIP-392797.27 (3.39) followed by between CIP-391058.175 and CIP-391046.14 (3.45), between CIP-393220.54 and CIP-391058.175 (3.62) and between CIP-398098.65 and CIP-394611.112 (3.70) (Table 2). Further, genetic distances among introduce genotypes were higher than those among the released varieties. This indicated that there is a higher chance of improving tuber yield, physical and internal quality traits through selection and hybridization of potato genotypes for yield and processing quality.

Generally, 36 (13.04%) pair of genotypes had genetic distances between 3.38 to 5.48, 124 (44.93%) pair of genotypes had genetic distances between 5.49 to7.58, 86 (31.161%) pairs of genotypes had genetic distance between 7.59 to 9.67 while 30 (10.87%) pair of genotypes had genetic distances between 9.68 to 11.78 (Figure 1 and Table 2).

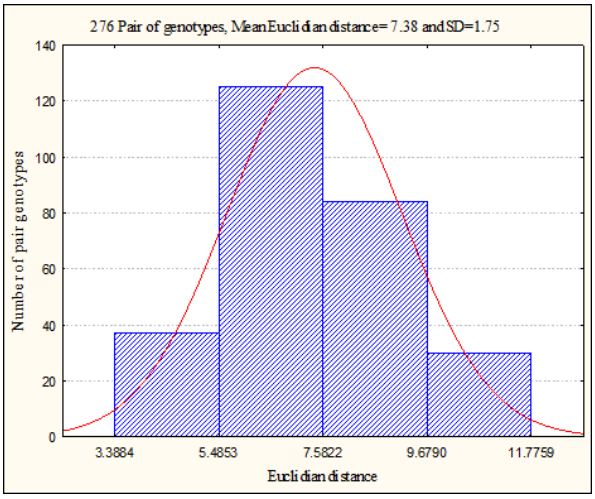

Figure 1: Distribution of 276 pair of potato genotypes into different categories of Euclidean distances.

In this study, the mean genetic distance (Euclidean distance) of each potato genotype to other 23 genotypes was calculated to generate information about the most distant and closest genotypes (Table 2). Genotypes CIP-399002.52 (9.59) followed by CIP-396027.205 (9.08) and CIP-399075.7 (8.77) had the highest Euclidean distance; whereas CIP-391058.175 (6.03) followed CIP-396034.268 (6.25) and CIP-381381.20 (6.33) had the lowest Euclidean distance. Generally, including the two released varieties (Belete and Dagim), 11 (45.83%) potato genotypes had mean genetic distance of above 7.38 and 13 (54.17%) potato genotypes showed mean genetic distance of below 7.38. The result indicated the presence of considerable distances or dissimilarities among the studied genotypes which could be used as parental genotypes in the crossing breeding program. Similar findings were also reported among potato genotypes [11].

Genotypes	Minimum	Maximum	Mean	SD	CV (%)
CIP-396034.268	4.59	8.53	6.25	1.26	20.15
CIP-393220.54	3.62	9.38	6.74	1.55	23.01
CIP-395017.229	3.4	11	7.2	1.59	22.1
CIP-392797.27	3.4	10.7	6.49	1.58	24.31
CIP-395112.19	4.6	10.7	7.29	1.48	20.26
CIP-399075.7	6.7	10.5	8.77	1.15	13.17
CIP-393280.64	4.73	9.32	6.77	1.17	17.36
CIP-398098.65	3.7	11.6	7.71	1.72	22.36
CIP-393385.39	4.7	10.4	8.28	1.44	17.4
CIP-396027.205	4.8	11.8	9.08	1.88	20.71
CIP-393077.159	4.39	8.89	6.74	1.32	19.63
CIP-399002.52	7.2	11	9.59	1.05	10.92
CIP-394611.112	3.7	11.7	7.86	1.87	23.85
CIP-392617.54	4.7	11.8	7.56	2.09	27.69
CIP-381381.20	4.46	8.87	6.33	1.35	21.35
CIP-398180.289	4.55	9.52	6.8	1.37	20.2
CIP-398190.89	4.5	10.7	7.01	1.78	25.36
CIP-398190.404	4.7	10.6	6.96	1.7	24.47
CIP-391058.175	3.45	8.57	6.03	1.36	22.55
CIP-396034.103	5.9	10.5	7.86	1.2	15.27
CIP-391046.14	3.5	10.1	7.39	1.7	23.04
Belete	4.7	11.6	7.63	1.96	25.74
Gudaine	4.09	9.14	6.82	1.11	16.21
Dagim	5.3	11	8.06	1.62	20.07
Overall	3.4	11.8	7.38	1.75	23.69

Table 2: Range and mean Euclidean distance of 24 potato genotypes estimated from 23 quantitative and six qualitative traits evaluated at Holetta in 2017.

The descriptive numeric data on the qualitative traits standardized or converted into a binary matrix using a Euclidian distance analysis procedure. The Euclidean distance matrix of

276 pair of genotypes estimated from tuber quality, yield and yield related traits was used to construct dendrograms based on the Unweighted Pair-group methods with Arithmetic Means (UPGMA). In the present study, all the 24 potato genotypes were grouped into eight clusters, in terms of quantitative and qualitative traits. Cluster II contained six (25%) potato genotypes, cluster IV had five genotypes (20.83%). cluster I had four (16.67%) genotypes, cluster III and V contained each three (12.5%) genotypes and cluster VI, VII and VIII had each one genotype. The three commercial varieties fall in cluster II and cluster IV. Several workers indicated wide genetic diversity and phylogenetic association among potato genotypes evaluated by them (Figure 2) [12].

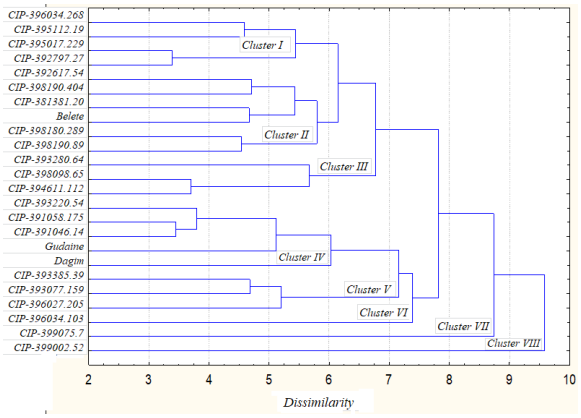

Figure 2: Dendrogram depicting dissimilarity of 24 potato genotypes by Unweighted Pair group Method with Arithmetic Means (UPGMA) clustering method from Euclidean distances matrix estimated from 23 quantitative and six qualitative traits.

Cluster II had showed days to 50% flowering (55.50 days), leaf area index (2.94cm-3), tubers dry mass weight (922.28 g/plant), total biomass weight (1169.15g/plant), average tuber weight (79.14 g/tuber), total tuber yield (36.04 t ha-1), marketable tuber yield (32.50 t ha-1), oblong tuber shape (50%), shallow and medium eye depth (50%), white- cream tuber skin color (66.67), cream tuber flesh color (66.67), brown chips color (50%) and light tan French fries color (50). Cluster III had early maturity (89.47 days), obovate tuber shape (66.67%), deep eye depth (66.67%), pink, red and red-purple tuber skin color (33.33%), white, cream and yellow (bright) tuber flesh color (33.33%), light tan chips color (66.67%) and white to cream French fries color (100). Cluster IV showed early maturity (89.47 days), elliptic tuber shape (60%), shallow eye depth (80%), yellow tuber skin color (80%), cream and yellow tuber flesh color (40%), dark tan chips and French fries color (60%). In cluster V genotypes showed sphericity of the tuber (92.17%), equally (66.67%) of round tuber shape, very deep eye depth, red tuber skin color, white tuber flesh color, dark tan chips and French fries color. Cluster VI, VII and VIII had contained each one genotype. Cluster VI showed Average stems number (5.53), medium size tubers (51.59%), specific gravity of tubers (1.10gcm-3), round tuber shape, very deep eye depth, red tuber skin color, cream tuber flesh color, white to cream chips and French fries color [13]. Cluster VII showed medium maturity (106.00 days), length width ratio (1.87), elliptic tuber shape, shallow eye depth, pink tuber skin color, Yellow tuber flesh color, white to cream chips and French fries color. Cluster VIII showed medium maturity (106.00 days), plant height (122.70 cm), shoot dry mass weight (439.00g/plant), average tuber number per hill (15.06), unmarketable tuber yield (3.69 t ha-1), small size tubers (56.67%), specific gravity of tubers (1.10gcm-3), dry matter content (25.75%), total starch content (18.95g/100g), ovate tuber shape, shallow eye depth, pink tuber skin, white flesh color, light tan chips and French fries color.

According to the cluster mean analysis for characters, developing varieties for processing purpose and tuber yield through selection further evaluation of genotypes from Cluster II and VIII is possible to obtain genotypes with highest total tuber yield, specific gravity of tuber, dry mater content, total starch content, acceptable tuber physical and frying quality with other desirable traits. Arslanoglu, et al. reported the grouping of 146 local potato genotypes collected from the Eastern Black Sea region of Turkey and into 27 clusters that had higher mean values for desirable morphological traits including tuber shape, skin color, eye color, flesh color, eye depth, skin texture, light sprout color, growth habit, flower color. Rangare and Rangare also reported that potato genotype clusters constructed and that had higher mean values for desirable traits including tuber yield and quality traits [14].

Principal component analysis

In this study, principal component (PC) analysis showed that the first eight principal components accounted for 90.26% of the total variation among 24 potato genotypes for the 23 quantitative and six qualitative traits. Of these, the first, the second and the third principal components constituted 28.69%, 18.74% and 13.00% of the variation, respectivel. The first eight components were retained in analysis because eigen values are >1. The others factors having eigenvalue < 1 were ignored due to Gutten's lower bound principle.

The results of the principal component analysis showed that more than two traits with small contribution accounted for each principal component load and the total contribution of the PC to the variation observed among genotypes. The total contribution of the first three principal component axes was 60.43%. The cumulative contribution of PC1 was due to the contribution (>0.25) of leaf area index, average tuber weight, total tuber yield, marketable tuber yield, geometric mean diameter and surface area of tubers. Shoot dry mass weight, average tuber number per hill, specific gravity of tubers, dry matter content, total starch content, plant height and tuber skin color contributed (>0.25) more to PC2, while average stems number, large size tubers, sphericity of the tuber and tubers eye depth contributed more to PC3. This indicated that these traits had higher contributions to the total differentiation of the genotypes into clusters. Thus selection efforts based on these traits including physical and frying quality may be more effective [15].

A similar trend in principal component analysis among potato genotypes has also been suggested by Mondal et al., in potato genotypes. Latifeh and Davoud reported greater eigenvector values for tuber yield and tuber uniformity traits in the first and/or second principal components. Rabeai et al. also

identified seven traits with three first major components in normal and drought stress condition.

CONCLUSION

The principal component analysis showed that the first eight principal components accounted for 90.26% among 24 potato genotypes for the twenty- nine traits. The genetic distances of the 24 potato genotypes ranged from 3.40 to 11.80 with the mean, standard deviation and coefficient of variation having the values of 7.38, 1.75 and 23.69%, respectively. Dendrograms constructed based on the Unweighted Pair-group Methods with Arithmetic Means (UPGMA) from Euclidean distance matrix of 276 pair of genotypes was capable to group the 24 potatogenotypes into eight clusters based on quantitative and qualitative characters. Cluster II, IV and I contained six (25 %), five (20.83 %), four (16.67 %) potato genotypes, respectively, cluster III and V each contained three (12.5 %) genotypes and cluster VI, VII and VIII each consisted one genotype. The three commercial released varieties were belonging to cluster II and cluster IV. Analysis of the cluster mean for characters revealed the possibility of obtaining or developing varieties with highest total tuber yield, specific gravity of tuber, dry mater content, total starch content, acceptable tuber physical and frying quality with other desirable traits for processing purpose and tuber yield through selection of genotypes in Cluster II and VIII.

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CONFLICT OF INTEREST

The Authors declare that there is no conflict of interest.

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