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Assessment of Genetic Diversity of Some Indigenous Collections of Upland Taro [Colocassia esculenta var. antiquorium (L.) Schott] for Selection of Genotypes Aiming at Improvement in Breeding Programme

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Abstract: *Upland taro [Colocassia esculenta var. antiquorium (L). Schott] is a crop of tremendous economic and social importance for its nutritive values, high energy yield per unit area, wide adaptability and high level genetic diversity. Yet received little attention for its genetic improvement and limited studies have been conducted on molecular evaluation of the crop. The objective of present study was to study the genetic divergence among twenty genotypes of upland taro using D^2 and principal component analysis. As per cluster analysis, the genotypes were grouped into six clusters consisting of 4, 9, 3, 1, 2 and 1 genotypes which revealed that there exist considerable diversity among the genotypes. Some of the local collections viz., BCC 15, BCC 18 and BCC 25 were very close to each other and may be a duplication which was evident in the D^2 analysis. As per dendrogram, Muktakesi was also closely related to Topi. Highest genetic dissimilarity was noticed between the genotypes BCC 21 and BCC 30, BCC 18 and BCC 30 and between BCC 30 and BCC 46. But, considering the magnitude of genetic distance, contribution of different characters towards the total divergence and magnitude of cluster means for different characters, the genotypes FC-4, FC-11, PKS-1, BCC-32, BCC-30 and Muktakeshi might be selected as suitable genotypes for future breeding program.*

Keywords: *Dendrogram, genetic diversity, parent selection, RAPD profiling, Upland taro.*

I. Introduction

Taro *[Colocasia esculenta (L.) Schott]* is one of the oldest cultivated crops grown for its edible corms and leaves ([1], [2], [3]). It is an important tropical tuber crop, used as a staple food or subsistence food by millions of people in the developing countries in Asia, Africa and Central America. World-wide it is the fifth most consumed root vegetable (FAOSTAT, 2005) with over 25% produced in Oceania and South-East Asia. The corms, leaves and petioles are used as a vegetable and considered as a rich source of carbohydrates, proteins, minerals and vitamins like, iron, phosphorus, zinc, potassium, copper, manganese and thiamin, riboflavin, pyridoxin, ascorbic acid and niacin respectively. Taro corms are very high in starch, and are a good source of dietary fiber and are also credited with having medicinal values. Production potentiality of taro does not only depend on cultural practices and management, but also on the suitability of taro varieties. Despite its importance as a popular edible tuber crop, very little attention has been devoted to the genetic improvement of taro. Information on the sexual potentialities of the crop has been very fragmentary, and the improvement programme have been largely dependent on the exploitation of the existing genetic variability among the cultivars ([2], [4], [5], [6], [7], [8]). For a long time it was believed that taro plants do not flower, and therefore fail to produce seeds ([9], [10]). However, later reports from different countries indicate that many clones flower and produce viable seeds ([11], [9], [10], [12], [13], [14]).

Genetic diversity is a useful tool in quantifying the degree of divergence in a biological population at genotypic level and to assess relative contribution of different components to the total divergence both at intra and inter-cluster levels ([15]). Cluster analysis is also carried out to detect divergent parents for hybridization purposes and to attain meaningful group constellations of a collection of genotypes. The progress of breeding is conditioned by the magnitude and nature of inter-relationship among the characters and variation of different characteristics. Knowledge about genetic control of the characters is essential in formulating an efficient breeding scheme as it provides not only a basis for selection but also some valuable indication relating selection of parents to be hybridized.

Among various PCR based assays, Randomly Amplified Polymorphic DNA primers (RAPD) are more important because they do not need any sequence information ([16]). The RAPD technique has received a great deal of attention from population geneticists ([17]) because of its simplicity and rapidity in revealing DNA-level genetic variation, and therefore has been praised as the DNA equivalent of allozyme electrophoresis ([18]).

RAPD analysis gives more accurate estimates between closely related populations and less accurate estimates for distantly related populations. RAPD data has been used for phyogenetic studies and generally supported existing taxonomies based on morphology. Among several efficient methods for revealing genetic variability within and among plant populations, the most widely applied methods is random amplified DNA polymorphism (RAPD) ([19], [20]). The research was done to analyze and identify some promising local collections and genotypes of taro, aiming at the genetic diversity using D^2 statistics based on agro-morphological parameters and RAPD array. Considering the availability of genetic variability, its scope of yield improvement and export potential, the present investigation was undertaken to search suitable diverse germplasm as suitable donor parents for the utilization in future breeding program.

II. Materials And Methods

2.1 Plant Material

The experiment was carried out with 20 different genotypes collected by AICRP on Tuber crops, BCKV from different parts of West Bengal and outside. The experiment was laid out in RCBD with three replications at the Central Horticultural Research Station, Mondouri, Bidhan Chandra Krishi Viswavidyalaya, Nadia, West Bengal. The experimental site was at 9.75 MSL, at approximately $22^0$57` N latitude and $88^0$20`E longitude (TABLE 1).

2.2 Topography and soil

Topographical situation of the experimental site is under gangetic new alluvial plains of West Bengal. The soil is sandy loam and slightly acidic. The relevant chemical properties of soil are given in TABLE 2.

2.3 Agro-climatic condition

The experimental site is under subtropical humid region. Meteorological observations were taken during the Dry and cool season (November- February) and Dry and hot season (March-May), (TABLE 3).

2.4 Agromorphological Characterization

Data was taken from ten randomly selected plants from each plot on both qualitative and quantitative traits. Observations were taken on to qualitative traits like leaf colour and leaf shape (Table 4) and following quantitative traits such as length of main sucker (cm), girth of main sucker (cm), no. of side suckers/plants, no. of petioles/clump, length of leaf lamina (cm), breadth of leaf lamina (cm), number of side tuber/plants, weight of side tuber/plants, estimated corm yield (t/ha).

2.5 Statistical Analysis

2.5.1 Analysis of variance in Randomized Complete Block Design (RCBD)

Difference between genotypes for different characters were tested for significance using analysis of variance on the basis of the following method complied by ([21]).

2.5.2 Analysis variance (ANOVA)

Test of significance of differences between individual treatments were done if calculated F value were significant. This was again done by computing critical difference (CD) at 5% level of significance.

Data were subjected to both univariate and multivariate analysis. Among different components of variation, Phenotypic coefficient of variation (PCV), Genotypic coefficient of variation (GCV) and Heritability were done.

2.6 DNA Extraction

Genomic DNA was extracted from the tender leaves of the plant, using standard protocol followed by ([22]). 100 mg leaf sample was taken and crushed in pre-chilled pestle mortar in liquid nitrogen. The DNA pellet was re-suspended in 100 µl of TE buffer (Tris-HCl 10mM and EDTA 1mM pH 8.0). The quality and quantity of extraction was verified by running each extracted DNA samples in 1% agarose gels stained with ethidium bromide. Samples were kept in -20°C.

2.7 PCR Amplification Through RAPD Markers

RAPD detects polymorphism at random. Here short single (~10nt) primer was used for PCR amplification. RAPD being a dominant marker produces large number of polymorphic bands so it can differentiate between very closely related organisms too.

PCR amplification was performed using the protocol outlined by ([23]) with slight modifications. The DNA was amplified with ten decamer primers from Operon (TABLE 5). Ingredients of each reaction included template DNA 25-30 ng, 100 mM each dNTPs, Taq DNA polymerase 0.5 units, $MgCl_2$ 600 mM, 10x PCR

reaction buffer (Bangalore Genei Ltd.) and 15ng of each decamer primer in a total volume of 25µl. Amplification was performed in a thermocycler (PCR system, eppendorf Mastercycler, U.S.). Complete reaction consisted of 36 cycles, each cycle consisting of three steps; denaturation at 92^0C for 30 seconds, annealing at 38^0C for 30 seconds, extension at 72^0C for 1 minutes, with an initial denaturation at 94^0C for 2 minutes and final extension at 72^0C for 7 minutes, followed by cooling at 4^0C temperature. 15µl of amplified PCR products were loaded on 1% agarose (Sisco Research Laboratory) gel containing ethidium bromide (0.5µg ml^{-1} of agarose) at 100 Volt for five hours in TAE buffer (Tris 1.6M, Acetic acid 0.8M, EDTA 40mM).

The gel was photographed and analyzed. A 100bp ladder (Biolab, England) was included in each gel as a molecular weight standard.

2.8 Data Analysis

Amplified fragments from all the primers, which were reproducible over two amplifications, were scored by the Total Lab gel documentation software (Biotech R. & D. Laboratories, Yercaud, India). The size of the fragments was estimated by using 1kb ladder marker, which was run along with amplified products in both sides. Bands were manually scored '1' for presence and 0 for absence and the binary data were used for statistical analysis. A genetic dissimilarity matrix was calculated according to Squared Euclidean Distance that estimated all pair-wise differences in the amplification product ([24]) and the cluster analysis was performed based on Ward's method using a minimum variance algorithm ([25]).

III. Results And Discussion

3.1 Morphological Characterization

Diversity in leaf colour and shape is considered to be most important as it is the primary criteria frequently used to characterize each of the diverse germplasm and to distinguish them visually. A wide degree of variation in leaf colour and shape existed among different selected genotypes, which were summarized in TABLE 4. It is quite evident that based on leaf color and shape though some of these genotypes could be identified, but the genotypes belonging to same color group might not always be easily identifiable as there was no clear color variation amongst them.

Hence, attempts were made to find out variability in respect of other physical parameters like length of main sucker, girth of main sucker, no. of side suckers per plants, no of petioles per plants, length of leaf lamina, breadth of leaf lamina, number of side tuber per plants, wt. of side tuber per plants, corm yield.

3.2 Analysis of genetic variability

Mean value of the genotypes for different character and estimates of different genetic variability parameters are shown in TABLE 9. Length of main sucker was highest in Muktakeshi (91.7 cm.) and lowest in BCC-26 (46.3cm) with mean value 72.4cm. The character had moderately high GCV and PCV indicating higher variability among the genotype. The character had high heritability (97.36%) with moderately high GA % over mean indicating additive nature and possibility of improvement through selection.

Girth of main sucker ranged from 10.57 to 16.31cm. with mean value 13.43 cm. BCC-46 had the highest girth. The character had moderate GCV and PCV with high heritability (89.72%) and moderate GA % over mean indicating less influence of the environment and possibility of improvement of the character through selection. No. of side sucker per plant was in Muktakeshi and lowest in PKS-1 with mean value of 3.31. The character had moderately low heritability (68.4%) with moderate GA% (38.20) over mean indicating less possibility of improvement of the character.

No. of petioles/clump, no. of side tuber/plants, wt. of side tubers/plants and corm yield had high GCV and PCV (>30%) with high heritability (>80%) and high GA % over mean indicating greater variability of the characters and greater possibility of improvement through selection. length of leaf and breath of leaf lamina had moderately low GCV and PCV indicating less variability for the character but had high heritability with moderate GA% over mean indicating less possibility of improvement.

Genetic diversity assesses the variability existing within the population and plays an important role in crop improvement program. The objective of present study was to identify closely or distantly related genotypes. In the present study D^2 statistics was used for assessing the underline genetic diversity within the population of 20 genotypes of taro collected from different districts of West Bengal and other parts of India.

3.3 Grouping of Varieties into Cluster

Distribution of 20 genotypes of taro in different clusters based on D^2 value is presented in TABLE 6. From the clustering pattern of the genotypes it was revealed that the 20 genotypes were grouped under six clusters. Cluster no. II bears maximum no. of genotypes (9). Among these 8 genotypes viz. BCC-1, BCC-2, BCC-10, BCC15, BCC-18, BCC21, BCC24 and BCC25 were collected from different district of West Bengal (TABLE 1). The genotypes seem to be very close. Another genotype i.e., Topi was collected from CTCRI,

Kerala and was grouped in Cluster II. The Cluster I included 4 genotypes out of which BCC-38, BCC-46 were collected from the Hooghly and GK-1 was collected from Murshidabad district. Another variety Telia, collected from Kerala was also grouped in Cluster I indicating its closeness with these Bengal genotypes. Cluster III included 3 genotypes (FC-4, FC-11 and PKS-1) from Assam. Cluster IV and Cluster VI included one genotype each. BCC-32 included in Cluster IV, was a collection from Nadia, West Bengal as well as Sonajuly, local genotype of Orissa, was included in Cluster VI. The Cluster V had 2 genotypes viz., BCC-30 and Muktakeshi from 24 Pgs., West Bengal and Kerala respectively.

3.4 Average Intra and Inter Cluster Distance

Average intra and inter cluster distance value of 20 taro genotypes (TABLE 7) revealed that Cluster V had highest intra cluster distance (16.23) indicating the dissimilarity between the two genotypes viz., BCC-30 and Muktakeshi. The lowest intra cluster D^2 value was noticed in cluster IV and cluster VI (0.0) indicating that these clusters bear only 1 genotype BCC-32 and Sonajuly respectively, followed by Cluster III (8.70) indicating closeness of the 3 genotypes (FC-4, FC-11 and PKS-1) in this group. All these 3 genotypes were collected from Jorhat, Assam. Intra cluster D^2 value of Cluster II was 13.39 and that of Cluster I was 12.77. These 2 clusters included 9 and 4 genotypes respectively. In the former cluster out of 9 genotypes, 8 where collected from different districts of West Bengal which indicated their closeness though variation in leaf shape and colour was observed in few cases. In the Cluster I also BCC-38, BCC-46 and GK-1 were collected from nearby districts of Bengal but morphologically all of them were not similar.

The inter cluster D^2 value ranged from 16.73 between Cluster II and V to 50.89 between Cluster III and IV. All the genotypes of Cluster III were collected from Assam where as the one genotypes of Cluster IV (BCC-32) collected from southern part of Bengal which showed their real dissimilarity. Inter cluster distances between Cluster III and V (37.67), Cluster I and IV (35.01), Cluster II and III (32.24) and Cluster IV and VI (32.69) were also comparatively larger in magnitude. The distance between Cluster I and VI, Cluster II and VI were comparatively small in magnitude which indicated there closeness. The inter cluster distances in all the clusters were higher than the intra cluster distances suggesting wider genetic diversity among the genotype of different groups. The results were in agreement with the previous reports ([26], [21]).

3.5 Selection of characters for future improvement

The parents for hybridization program should be selected based on the magnitude of genetic distance, contribution of different characters towards the total divergence and magnitude of cluster means for different characters performance having maximum heterosis. A higher heterosis could be produced from the crosses between genetically distant parents ([27]). There was existence of variation for almost all of the characters among different clusters (TABLE 8). Genotypes of distantly located clusters were suggested to use in hybridization programs for obtaining a wide spectrum of variation among the segregates ([28], [29], [30]). Cluster IV was the most important cluster for its highest contribution towards length of main sucker (84.72 cm.), no. of side tuber (20.47), weight of side tuber (378.97 g.) and corm yield (24.20 t/ha) and ranks second for no. of petioles/clump (13.62). These are considered as improvable characters through selection in future hybridization programme. The genotypes of cluster V were more important for girth of main sucker (16.92 cm.), no. of side sucker (4.48) and no. of petioles per clump (14.60). Cluster Ibearing 9 genotypes contributed significantly for weight of side tuber (270.61 g.), corm yield (16.29 t/ha) and no. of petioles per clump (9.23).

3.6 Genotypes selection for improved breeding programme

Higher inter and intra-cluster distances indicated higher genetic variability among genotypes between and within clusters, respectively. The minimum inter and intra-cluster distance indicated closeness among the genotypes of two clusters and within the cluster also. Genotypes belonging to the distant clusters could be used in hybridization program for obtaining a wide spectrum of variation among the segregants ([31]). Crossing might be carried out between genotypes belonging to different clusters having genetic distances (D2) greater than 12.5 ([32]). Thus genotypes selected from the distant clusters II, III, IV, V and VI may manifest the highest level of heterotic response in crosses.

Maximum heterosis observed in those crosses, which were involved moderately diverged parents ([33]). Researchers also reported the heterobeltiosis from the cross between intermediate distant classes ([34]). Previous findings say that genetic diversity is very much important factor for any hybridization program aiming at genetic improvement of yield especially in self-pollinated crops like rice ([35], [36]). Mahalanobis's D2 statistic is a powerful tool for choosing diverse parents for hybridization ([37]). Therefore crosses between the genotypes of clusters III and IV, III and V, II and III, IV and VI would give high manifestation of heterosis as well as wide spectrum of genetic variation in F2 generation. The inter cluster distance was highest between cluster III and cluster IV (50.89) followed by cluster III and cluster V (37.67) suggesting wider genetic diversity

among the genotypes of these groups and also maximum contribution towards characters considered most important for improvement in hybridization and yield of the crop.

3.7 Genetic Diversity Using RAPD Markers

Amplified fragments from 10 selected decamer primers were used for the present analysis. Amplified fragments which were reproducible over two amplifications were scored by the total lab gel documentation software (Biotech R & D Laboratories, India). The size of the fragments (molecular weight in base pair) was estimated by using ladder marker with the range of 50bp to 1.5kb, which was run along with the amplified products in both sides. Bands were manually scored '1' for presence and '0' for absence and the binary data were used for statistical analysis. A genetic similarity matrix was calculated according to NTSYSpc software that estimated all pair-wise difference in the amplifications product and the cluster analysis was performed based on Jaccard's method using minimum variance logarithm.

Analysis revealed that 9 primers out of 10 amplified total 41 bands, out of them 5 bands were monomorphic. Band sizes ranged from 60bp to 1500bp and the number of scorable bands per primer ranged from 3 to 8 with an average of 4.55 bands per primer. 9 primers were highly informative because they either amplified more than 4 polymorphic or monomorphic bands, which could differentiate between specific Colocasia accessions. The amplifications products by the primers 1, 2, 3, 4, 5, 7, 8, 9 and 10 are illustrated in Fig. 1, Fig. 2 and Fig. 3.

Based on estimated proximity matrix scale generated from the data, lowest genetic similarity (0.27) i.e., highest genetic dissimilarity, was noticed between the genotype of BCC 21 and BCC 30 and followed by (0.39) between BCC 18 and BCC 30 and similar distance was noticed between BCC 30 and BCC 46. The closest proximity (0.95) was noticed between BCC 15 and BCC 18 and followed by (0.90) between BCC 18 and BCC 25. Thus BCC-15, BCC-18 and BCC-25 and were very close to each other and were placed in the same group. Muktakesi was closely related to Topi with the similarity index of 0.88 units followed by 0.82 units between Telia and FC-4. Lowest similarity index (0.56) was found between Sonajuly and FC-11 followed by 0.57 between Muktakesi and FC-11. The dendrogram made from these data showed that the 20 cultivars formed five distinct clusters (I-V) (Fig. 4) based on 50% dissimilarity. Six genotypes viz., BCC-1, BCC15, BCC-18, BCC-25, BCC-32 and BCC-2 could be grouped into Cluster I. Muktakeshi, Topi, BCC-38, FC-4, Telia, BCC-46, PKS-1, FC-11, GK-1 and Sonajuly (ten genotypes) could be grouped into Cluster II. BCC-21 could be placed into cluster III whereas BCC-10 and BCC-24 could be grouped together in Cluster IV. BCC-30 could be placed into a separate Cluster V.

IV. Conclusion

These diverse genotypes may be employed in future crop improvement programmes wherever possible. These findings indicated that genotypes of cluster III (FC-4, FC-11, PKS-1) vs. genotype belonging to cluster IV (BCC-32) and genotypes of cluster III vs. genotypes of cluster V (BCC-30 and Muktakeshi) could be used in plant breeding program for development of upland taro variety *[Colocassia esculenta var. antiquorium (L.) Schott]*. RAPD markers provide a fast and efficient tool for genetic variability assessment and is in current use in plant genetic resources management. Also, it was accepted that for the assessment of genetic diversity, molecular markers were superior to morphological, biochemical and other method like pedigree and heterosis ([38]).

Low genetic dissimilarity of parents yielded weak heterosis effects and future studies need to be conducted by using a broader genetic base ([39]). This was the first study assessing the relationship of hybrid vigour with the genetic distances between parents, conducted on a tropical root crop. The taro breeding in India is limited to the National level research programme. Structuring the genetic diversity is necessary to optimize the use of germplasm by breeders for which molecular level screening is highly warranted. Large scale production and evaluation of the crop revealed the tremendous scope for the genetic improvement of the crop through hybridization and selection. Indian agriculture has to be accompanied by intense genetic improvement of the crop for which it is essential to have International co-operation among taro breeders and establishing a procedure for germplasm exchange.

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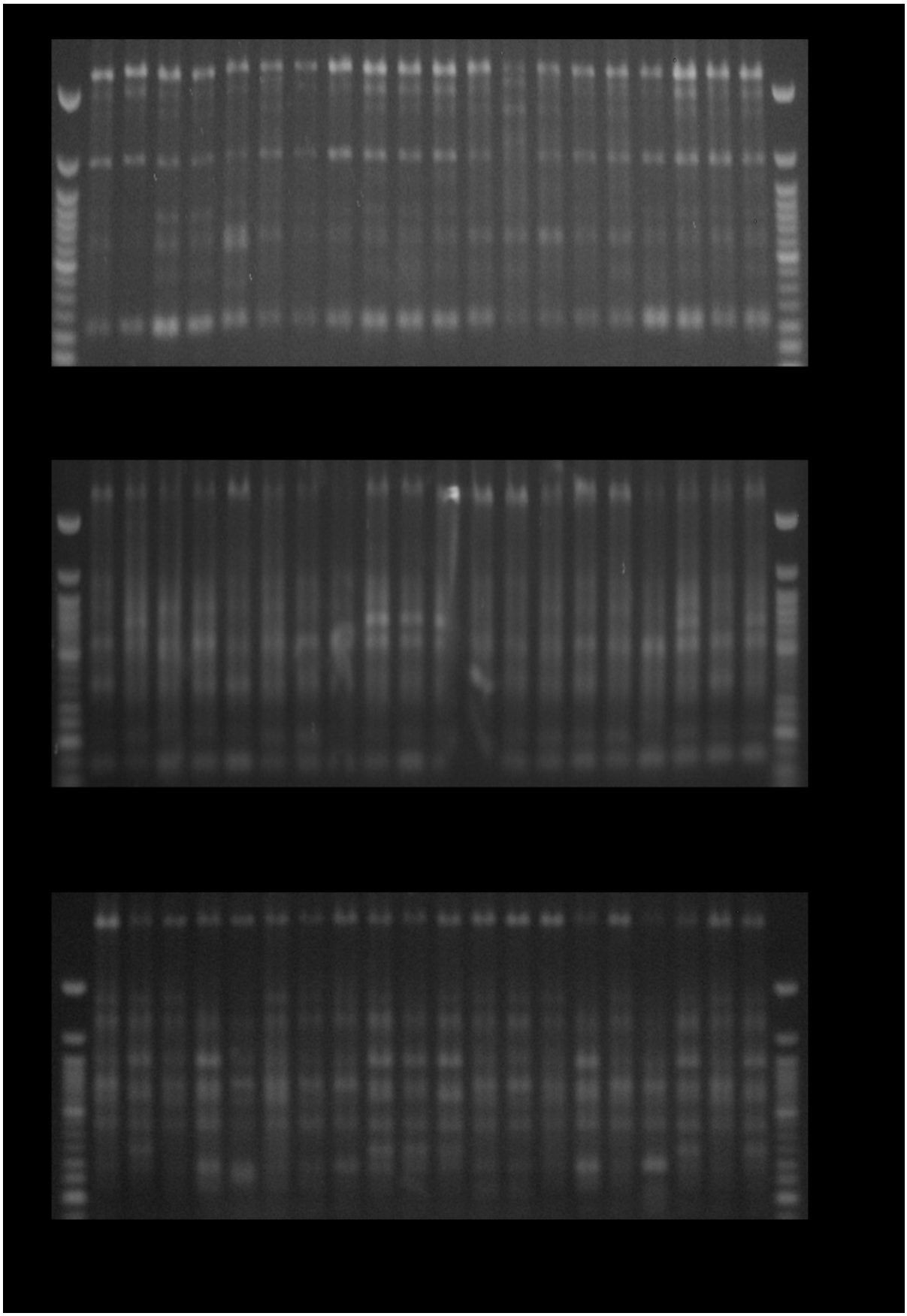
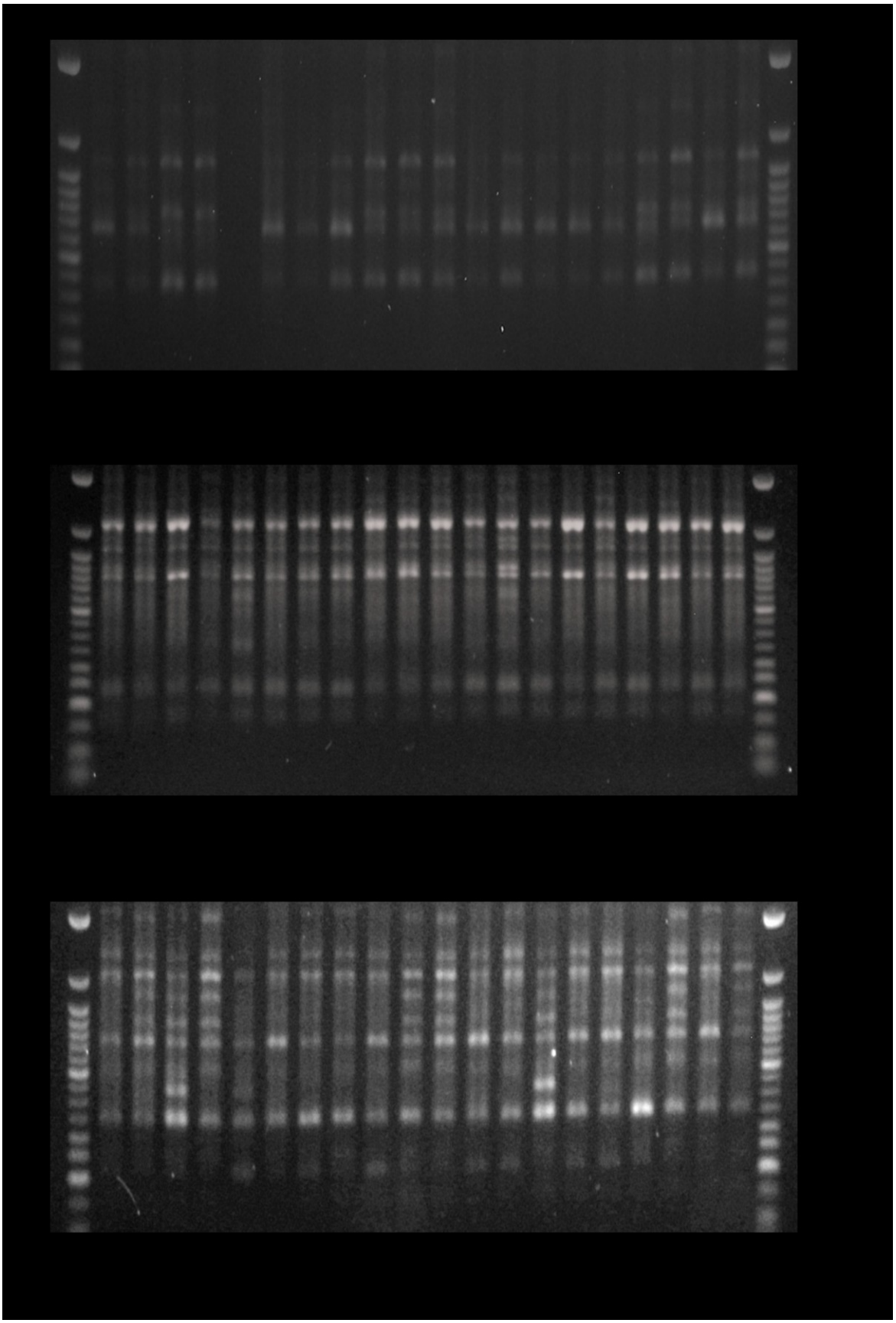
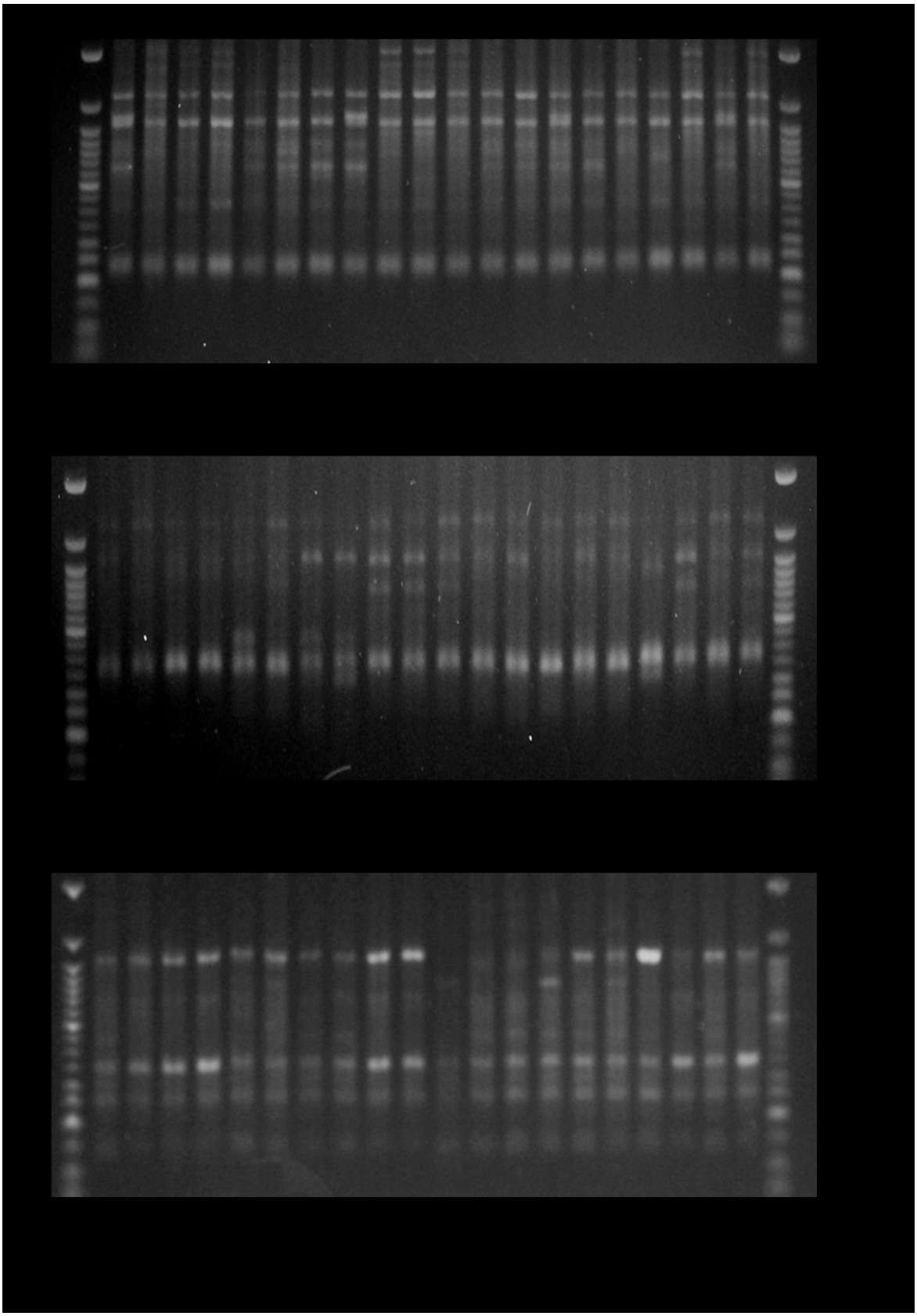


The electronic patterns of RAPD of selected accessions of Taro using primers OPA01,OPA02 and OPA03. LANE 1-22: Ladder, BCC-1, BCC-2, BCC-10, BCC-15, BCC-18, BCC-21, BCC-24, BCC-25, BCC-30, BCC-32, BCC-38, BCC-46, Sonajuly, Muktakeshi, Telia, Topi, FC-4, FC-11, GK-1, PKS-1, Ladder



Gel image of selected accessions of Taro using primers OPA04,OPA05 and OPA07. LANE 1-22:Ladder, BCC-1, BCC-2, BCC-10, BCC-15, BCC-18, BCC-21, BCC-24, BCC-25, BCC-30, BCC-32, BCC-38, BCC-46, Sonajuly, Muktakeshi, Telia, Topi, FC-4, FC-11, GK-1, PKS-1, Ladder



Banding patterns of selected accessions of Taro using primers OPA08,OPA09 and OPA10. LANE 1-22:Ladder, BCC-1, BCC-2, BCC-10, BCC-15, BCC-18, BCC-21, BCC-24, BCC-25, BCC-30, BCC-32, BCC-38, BCC-46, Sonajuly, Muktakeshi, Telia, Topi, FC-4, FC-11, GK-1, PKS-1, Ladder

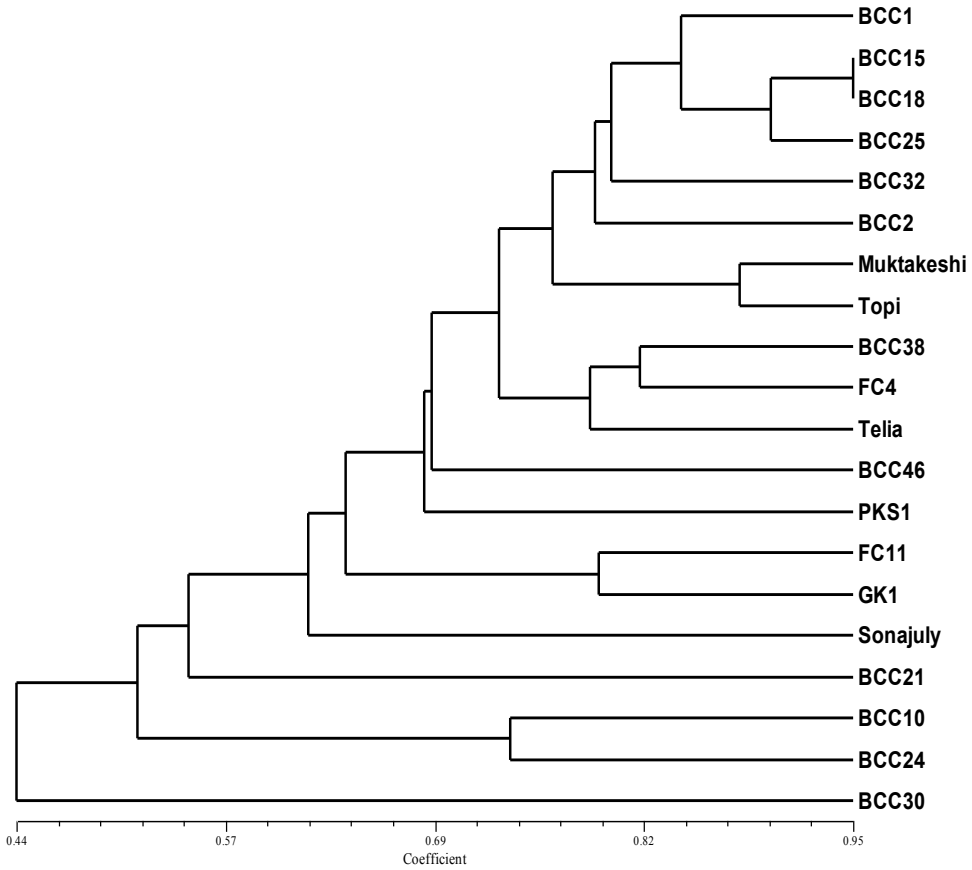


Fig. 4. Dendogram based on RAPD banding profile

Table 1: Passport information of *Colocassia esculenta*

SL. NO.	VARIETY NAME	IC NO.	DISTRICT	STATE
1	BCC1	361213	NADIA	WEST BENGAL
2	BCC2	361214	NADIA	WEST BENGAL
3	BCC10	361222	MIDNAPUR	WEST BENGAL
4	BCC15	361227	NADIA	WEST BENGAL
5	BCC18	361230	SOUTH DINAJPUR	WEST BENGAL
6	BCC21	361233	BIRBHUM	WEST BENGAL
7	BCC24	361236	BIRBHUM	WEST BENGAL
8	BCC325	361237	SOUTH DINAJPUR	WEST BENGAL
9	BCC30	361242	24 PGS.	WEST BENGAL
10	BCC32	361244	NADIA	WEST BENGAL
11	BCC38	-	HOOGHLY	WEST BENGAL
12	BCC46	-	HOOGHLY	WEST BENGAL
13	SONAJULY	-	CTCRI	ORISSA
14	MUKTAKESHI	-	CTCRI	ORISSA
15	TELIA	-	CTCRI	KERALA
16	TOPI	-	CTCRI	KERALA
17	FC-4	-	JORHAT	ASSAM
18	FC-11	-	JORHAT	ASSAM
19	GK-1	-	MURSHIDABAD	WEST BENGAL
20	PKS-1	-	JORHAT	ASSAM

Table 2: Chemical properties of the soil

Particulars	Value	Method applied
Organic carbon(%)	0.57	Walkey and Blac (Jackson, 1973)
Total nitrogen (%)	0.06	Kjeldahl Method (Jackson1 973)
Available phosphorus(Kg/ha.)	30.1	Olsen's Method (Jackson,1973)
Available potassium (kg/ha.)	115.7	Olsen's Method (Jackson,1973)
Soil P^H	6.5	Beckman's PH meter method in 1:25 soil suspension (Jackson,1973)

Table 3: Meteorological data pertaining to the period of field experimentation

Months 2012/2013	Temperature (°C)		Relative Humidity (%)		Rainfall (mm)
	Max	Min	Max	Min	
2012April	37.2	25.5	89.7	41.0	0.2
May	34.6	25.7	89	61.0	199.8
June	35.6	27.1	89.5	64.5	66.3
July	32.7	26.5	94.1	76.3	227.4
Aug	32.6	26.1	96.1	80.1	457.4
Sept	32.9	26.2	95.2	74.2	210.6
Oct	32.4	22.0	94.8	60.5	91.1
Nov	30.2	18.5	93.2	54.8	3.8
Dec	26.8	11.7	93.8	49.8	0
2013Jan	23.6	9.26	95.7	51.3	0
Feb	29.5	14.6	92.0	42.6	0
March	35.8	22.4	89.3	38.4	0

Table 4: Variation in leaf colour and shape observed in 20 taro genotypes

Sl. No.	Germplasm	Leaf colour	Leaf shape
1	BCC-1	Deep green	Lanceolate
2	BCC-2	Deep green	Lanceolate
3	BCC-10	Deep green	Cup
4	BCC-15	Deep green	Cup
5	BCC-18	Light green	Lanceolate
6	BCC-21	Deep green	Lanceolate
7	BCC24	Deep green	Cup
8	BCC-25	Deep green	Cordate
9	BCC-30	Bass green	Lanceolate
10	BCC-32	Bass green	Cup
11	BCC-38	Bass green	Lanceolate
12	BCC46	Purple green	Oval
13	Sonajuly	Deep green	Cordate
14	Muktakeshi	Deep green	Cup
15	Telia	Purple green	Oval
16	Topi	Green	Lanceolate
17	FC-4	Light green	Cordate
18	FC-11	Light green	Cordate
19	GK-1	Yellow green	Cordate
20	PKS1	Deep green	Lanceolate

Table 5 : Primers used during the study, their sequence and annealing temperature.

Primers	Primer sequence (5´-3´)	Annealing Temperature
OPA01	ATGGCCTTAC	30ºC
OPA02	GAGGGAGACC	36 ºC
OPA03	CGGTGGCGAA	34 ºC
OPA04	TTGCTTGGCG	32 ºC
OPA05	CTGGGGCCGT	36 ºC
OPA06	TACGTCTTGC	30 ºC
OPA07	GTCTCTCCC	34 ºC
OPA08	GTCGGTACCC	34 ºC
OPA09	ACAGGGAACG	32 ºC
OPA10	GCGTGACCCG	36 ºC

Table 6: Distribution of 20 genotypes of taro into different cluster based on D^2 Values

Cluster	No. of genotypes	Name of the genotype
I	4	BCC-15, BCC-38, BCC-48, Telia, G.K.-1
II	9	BCC-1, BCC-2, BCC-10, BCC-15, BCC-18, BCC-21, BCC-24,BCC-25,Topi
III	3	FC-4, FC-11, PKS-1
IV	1	BCC-32
V	2	BCC-30, Muktakeshi
VI	1	Sonajuly

Table 7: Average intra and inter cluster distance (D^2 values) of the taro genotype

Cluster	I	II	III	IV	V	VI
I	12.77					
II	20.68	13.39				
III	20.53	34.24	8.70			
IV	35.01	21.13	50.89	0.00		
V	25.11	16.73	37.67	20.03	16.23	
VI	17.12	18.99	23.40	32.69	21.85	0.00

Table 8: Mean values of different character of each clusters

Character	Cluster I	Cluster II	Cluster III	Cluster IV	ClusterV	Cluster VI
Length of main sucker	56.78	80.78	55.70	84.72	79.32	83.50
Girth of main sucker	13.48	13.30	10.93	14.72	16.92	11.30
No. of side sucker	3.04	3.03	3.08	4.27	4.48	4.32
No. of Petioles per clump	7.62	9.23	6.98	13.62	14.60	7.54
Length of leaf lamina	31.15	32.54	36.70	29.34	35.69	40.50
Bredth of leaf lamina	24.20	20.99	27.08	21.73	21.81	29.10
No. of side tuber	12.83	15.61	6.02	20.47	12.98	11.95
Wt. of side tuber	221.69	270.61	112.49	378.97	214.78	125.20
Corm yield	13.30	16.29	3.37	24.20	15.15	15.00

Table 9: Mean values of 20 taro genotypes for different characters

Characters / Genotypes	Length of main sucker	Girth of main sucker	No. of side sucker	No. of Petioles per clump	Length of leaf lamina	Breadth of leaf lamina	No. of side tuber	Wt. of side tuber	Corm yield
BCC-1	83.40	15.88	3.86	12.24	37.44	23.52	16.95	382.67	18.46
BCC-2	71.24	13.44	2.72	9.49	32.41	21.27	19.17	334.27	13.09
BCC-10	81.95	14.71	2.90	8.68	31.52	22.39	17.70	328.00	14.38
BCC –15	91.23	12.61	3.58	10.47	31.57	21.73	12.15	153.40	14.55
BCC-18	91.61	14.47	3.54	7.31	34.68	21.80	11.27	170.00	15.22
BCC-21	65.59	12.39	2.45	8.66	28.55	21.75	17.47	284.07	18.47
BCC-24	72.51	13.83	3.23	10.74	36.28	19.44	18.43	370.10	19.10
BCC-25	82.74	12.92	2.24	6.67	27.05	18.68	16.97	265.00	17.76
BCC-30	66.94	18.30	4.57	16.56	35.88	20.35	15.17	192.88	12.80
BCC-32	84.72	14.72	4.27	13.62	29.34	21.73	20.47	378.97	24.20
BCC-38	58.57	12.17	3.63	6.17	35.40	25.60	12.75	182.33	15.88
BCC-46	46.30	16.31	3.00	7.53	27.13	18.93	16.40	227.00	11.20
Sonajhuli	83.50	11.30	4.32	7.54	40.50	29.10	11.95	125.20	15.00
Muktakeshi	91.70	15.54	4.39	12.65	35.50	23.27	10.78	236.67	17.51
Telia	71.69	12.31	3.18	8.46	26.49	25.44	15.60	257.43	13.88
Topi	86.77	11.88	2.73	8.83	33.37	18.39	10.42	148.00	15.57
FC-4	57.53	10.57	4.23	6.90	37.40	27.87	6.47	149.33	3.46
FC-11	53.97	10.81	3.24	9.04	38.37	29.54	4.93	85.47	2.61
G.K.-1	50.57	13.13	2.36	8.33	35.58	26.83	11.75	220.00	12.24
PKS-1	55.60	11.41	1.78	4.99	34.34	23.83	6.67	102.67	4.03
Mean	72.40	13.43	3.31	9.24	33.43	23.07	13.67	229.67	13.97
SE (Mean)	1.98	0.54	0.41	0.52	1.48	0.74	0.91	15.87	0.99
CD at 5%	4.02	1.11	0.83	1.06	3.01	1.52	1.84	32.21	2.02