Morphological Characterization of Selected *Capsicum* Accessions and Development of Species Identification Key for Chili

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Abstract- *Capsicum is widely grown as an essential spice crop in Sri Lanka. The existing germplasms of Capsicum conserved at Plant Genetic Resource Centre (PGRC), Gannoruwa were initially identified as two species, Capsicum annuum, and Capsicum frutescens, instead, it is hypothesized that there may be additional genotypes within the existing germplasm collections. Hence, the present study was focused on evaluating twelve Capsicum accessions and to characterize morphologically including 24 qualitative and seven quantitative traits to assess the genetic diversity among plants. The experiments were carried out at the open field and protected house of PGRC, Gannoruwa by randomized complete block design with four replicates. Morphological characters were analyzed using analysis of variance (ANOVA) and multivariate methods. Significant variance among genotypes was obtained for most of the quantitative characteristics ($p<0.05$). Early flowering (41d) and fruiting (69d) was observed in accessions C-2018-12-232, NM-9-3-R1, NM-6-2-R2-B, and ACC# 1249B, while the remainder accessions exhibited late flowering (>47d) and harvest (>75d) nature respectively. Principal component (PC) analysis explained more than 73.12 % of total variability for the first three components. PC1 was highly positively correlated with seed beak prominence, corolla color, and seed shape, while PC2 was highly correlated positively with fruit color, shape at the blossom end, and fruit positions. Hence this study attempted to develop a species identification key for chili species by employing morphological traits, though seed shape can be considered as a power tool. Moreover, the dendrogram confirmed that the germplasms resemble into three chili species as C. annuum, C. chinense, and C. frutescens.*

Keywords: Capsicum accessions, Germplasms, Morphological characterization, Principal component analysis

I. INTRODUCTION

Capsicum spp. (2n=24), commonly called chili pepper is one of the most cultivated spice crops belongs to family Solanaceae. It is grown worldwide for its high economic importance as a spice and vegetable crop (Orobiyi *et al*., 2013). Genus *Capsicum* consists of around 32 species, among five of them (*C. Annuum, C. frutescens, C. chinense, C. baccatum, and C. pubescens*) were domesticated (Jing *et al*., 2013). Global estimated average production of chili peppers is 34.5 million tons (FAOSTAT, 2018). *Capsicum* is a great source of vitamins, minerals, amino acids and secondary metabolites such as carotenoids, ascorbic acid and flavonoids which have both nutritional and medicinal values (Pawar *et al*., 2011).

It is an evidence that chili pepper was originated in South America in 7500 BC (Andrews, 1999). During 15th and 16th centuries chili pepper was introduced to the Europe, Africa, and Asia. It was used as a substitute for expensive "Black Pepper" imported from Asia. Historically, chili pepper was utilized for decoration and seasoning purposes and now it is used in medicine and pharmaceutical industries (Paran and Van Der Knaap, 2007). The main chemical compound in chili pepper is capsaicin which is a lipophilic chemical (Lu *et al*., 2020; Zhang *et al*., 2020). Medicinally, capsaicin is being used to treat pain (Hall *et al*., 2020). At present, it is the most recommended tropical medication for arthritis (Kwenin *et al*., 2011). Further, various chili pepper varieties contain high levels of antioxidant vitamins A, C and E. *Capsicum* is rich in vitamin C, pro vitamin A, E, P (citrin), B1 (thiamine), B2 (riboflavin), and B3 (niacin) (Samira *et al*., 2013; Gupta, 2015). Antioxidants in foods have been the subject of extensive studies for cancer prevention (Antonious *et al*., 2009).

In Sri Lanka, chili pepper is an important cash crop cultivated especially in the northern and north-central provinces. Currently, the chili

pepper growing regions are Anuradhapura, Moneragala, Ampara, Puttalam, Vavuniya, Kurunegala, Hambantota and Mahaweli System H (DOA, 2020). Commonly cultivated chili pepper species are *Capsicum annuum* and *Capsicum frutescens*. They are consumed as spices for their specific flavor, aroma and pungency (Menike e*t al*., 2018). According to the Department of Agriculture (2020), Sri Lanka, annual per capita consumption of dry chili pepper is estimated as 2.84 kg and the national annual requirement of dry chili pepper is around 57,400 Mt. Although chili pepper is widely grown for dry chili production, a portion of the crop is harvested as green pods. Currently, the average area under chili pepper cultivation is around 13, 000 ha, with 2/3 of that being cultivated during *Maha* season (DOA, 2020).

Evaluation of genetic diversity is an essential tool for crop breeding programs to develop improved varieties having enhanced productivity and resistance to biotic and abiotic stresses. There are more than 600 accessions of *Capsicum* conserved at Plant Genetic Resource Centre (PGRC), Gannoruwa, Sri Lanka. Initially, these germplasms were identified as two species, *Capsicum annuum* (Amu miris) and *Capsicum frutescens* (Nai miris and Kochchi miris). But, it is hypothesized that there may be additional species within existing germplasm collections. It is necessary to correctly define and evaluate these accessions conserved in the gene bank in order to make them available for crop breeding programs. Genetic diversity of a population can be studied through germplasm characterization that can be performed using morphological, biochemical and molecular techniques. Plant Genetic Resources Centre, Gannoruwa is the main germplasm collection center of Sri Lanka and PGRC has developed species identification keys for several crops, but not for chili germplasms. Therefore, this study was aimed to characterize 12 *Capsicum* accessions conserved in PGRC, Gannoruwa morphologically to identify the diversity and latent potentials for use in *Capsicum* improvement program in Sri Lanka and to develop a key for easy identification of chili species.

II. MATERIALS AND METHODOLOGY

A. Experimental site and planting materials

A field trial was established at Plant Genetic Resources Center (PGRC), Gannoruwa, Sri Lanka (7^{o} 27'N and 80^{o} 60'E; altitude 473 m above sea level) from December 2020 to April 2021 with a total of twelve *Capsicum* accessions including *C. Annuum, C. frutescens* and *C. chinense* (Table 1) conserved at PGRC, Gannoruwa. Initially, *Capsicum* seeds were grown in germination trays inside a greenhouse. After three weeks, seedlings were transplanted to the raised type of field plots. The experiment was laid out in Randomized Complete Block Design (RCBD) with four replicates. Seedlings were grown according to the recommendation of DOA, Sri Lanka (45 cm X 60 cm).

Table 1: Accessions of Capsicum annuum, Capsicum frutescens and Capsicum chinense used in this study

No	Accessions	Treatments	Species
1	ACC# 1249B	T1	*Capsicum annuum*
2	NM-9-3-R1	T2	*Capsicum annuum*
3	NM-6-2-R2-B	T3	*Capsicum annuum*
4	C-2018-12-232	T4	*Capsicum annuum*
5	NM-3-2-R1	T5	*Capsicum chinense*
6	NM-6-2-R2-A	T6	*Capsicum chinense*
7	NM-3-4-R1	T7	*Capsicum chinense*
8	NM-6-R-2	T8	*Capsicum chinense*
9	C-2018-11-139	T9	*Capsicum frutescens*
10	C-2019-4-165	T10	*Capsicum frutescens*
11	ACC# 08149	T11	*Capsicum frutescens*
12	C-2018-12-246	T12	*Capsicum frutescens*

Table 2: Descriptors of qualitative and quantitative morphological traits used to characterize chili in Plant Genetic Resources Centre, Gannoruwa.

Parameter	Descriptors
Qualitative characters	
(SBP) Seed beak prominence	1:Nub, 2: Medium
(CC) Corolla color	1:White, 2: Light yellow, 3: Yellow, 4: Yellow-green, 5: Purple with white base, 6: White with purple base, 7: White with purple margin, 8: Purple, 9: Other (specify)
(SS) Seed shape	1: Reniform, 2: circular with fish mouth, 3: teardrop
(CM) Calyx margin	1: Entire, 2:Intermediate, 3: Dentate
(LC) Leaf color	Recorded when in 50% of the plants the first fruit has begun to ripen. Based on 10 leaves on the main branches of the plant.
(PGH) Plant growth habit	Observed when 50% of the plants bear ripe fruits. 1: Prostrate, 2: Intermediate (compact), 3: Erect, 4: Other (specify)
(LS) Leaf shape	1: Deltoid, 2:Ovate, 3: Lanceolate
(FS) Fruit set	Recorded before harvest. 1: Low, 2: Intermediate, 3: High
(NA) Nodal anthocyanin (whole plant)	1: Green, 3: Green with purple, 4: Purple, 5: Dark purple
(BH) Branch habit	1: Sparse,2:Intermediate, 3:Dense
(LP) Leaf pubescence	Observed on the youngest mature leaves. Same stage as in 10.1: Sparse, 2: Intermediate, 3: Dense
(FCM) Fruit color maturity (ripe fruit color)	1: White, 2: Lemon-yellow,3: Pale orange-yellow, 4: Orange yellow, 5: Pale orange, 6: Orange, 7: Light red, 8: Red, 9: Dark red, 10: Purple, 11: Brown, 12: Black, 13: Other (specify)
(FS) Fruit shape	1: Elongate, 2: Almost round, 3: Triangular, 4: Campanulate, 5: Blocky, 6: Other (specify)
(FSB) Fruit shape at blossom end	Average of 10 fruits.1: Pointed, 2: Blunt, 3: Sunken, 4: Sunken and pointed., 5: Other (specify)
(FP) Fruit position	1: Up, 2: Down
(FCI) Fruit color at intermediate stage	Recorded on fruits just before the ripening stage. 1: White, 2: Yellow, 3: Green, 4: Dark green, 5: Orange, 6: Purple, 7: Deep purple, 8: Other (specify)
(FSP) Fruit shape at pedicel attachment	1: Acute, 2:Obtuse, 3: Truncate, 4:Cordate, 5: Lobate
(FP) Flower position	Recorded at anthesis.1: Pendant, 2: Intermediate, 3: Erect
(SP) Stem pubescence	Recorded on mature plants, excluding the first two nodes below the shoot. 1: Green, 3: Green with purple, 4: Purple, 5: Dark purple
(AC) Anther color	Observed immediately after blooming before anthesis. 1:White, 2:Yellow, 3:Pale blue, 4:Blue, 4:Purple, 5:Other (specify)
(SC) Stem color	Recorded on young plants before transplanting 1:Green, 2:Green with purple stripes, 3:Purple, 4:Other (specify)
(SE) Stigma excretion	In relation to anthers at full anthesis. Average of 10 stigmas from representative flowers selected from 10 random plants. 1:Inserted, 2:Same level, 3:Exerted
(CAC) Calyx annular constriction	At junction of calyx and pedicel. Observed at mature stage 1:Absent, 2:Present

(AS) Anthocyanin spots or strips on the fruit	Recorded just before the ripening stage 0:Absent, 1:Present
Quantitative characters	
(DF) Days to flowering (Days)	Number of days from sowing/ transplanting until 50 % of plants have at least one open flower.
(DF)Days to fruiting (Days)	Number of days from transplanting until 50 % of the plants bear mature fruits at the first and second bifurcation.
(NFA) Number of flowers per axil	1: One, 2: Two, 3: Three or more, 4: Many flowers in bunches but each in individual axil (fasciculate growth), 5: Other (specify)
(LW) Leaf width (cm)	Same stage as in 10 Measured on the widest part of the leaf. Use same leaves as in 13.
(MLL) Leaf length (cm)	Same stage as in 10. Average of 10 leaves.
(PH) Plant height (cm)	At flowering stage 1: <25, 2: 25-45, 3: 46-65, 4: 66-85, 5: >85
(TSW) Thousand seed weight (g)	(100×10)

B. Crop management

Field was fertilized with Urea, TSP and MOP at the rate of 200 Kg/ha, 220 Kg/ha and 130 Kg/ha respectively. Half of the urea, MOP and entire quantity of TSP were applied as basal dose and remaining 50 % of urea and MOP were added four weeks after planting. Plants were watered twice a day and weeding was done once a week manually. Gap-filling was done by replacing the unhealthy seedlings with healthy plants whenever necessary.

C. Data collection and statistical analysis

Data were recorded on 24 qualitative and 7 quantitative morphological characters (Table 2) and analyzed using Statistical Package for Social Sciences (SPSS). Analysis of variance and two multivariate analysis methods: Principal Component Analysis (PCA) and hierarchical cluster analysis were performed to analyze the data.

III. RESULTS AND DISCUSSION

A. Analysis of variability in quantitative morphological traits

Analysis of variance revealed significant differences among genotypes for most of the quantitative characteristics viz; plant height, leaf length and width, days taken to flowering and fruiting, number of flowers per axil and 1000 seed weight at 0.05 % probability level (Table 3). Chili accession NM-6-2-R2-B (T3) had the highest plant height (74.5 cm) while NM-3-4-R1 (T7) and NM-6-R-2 (T8) both had the lowest value (35.25 cm). It shows that the former accession seems to display tall and bushy types while the latter two were with dwarf appearances. The mean length and width of leaves were also showed diversified in nature. The highest leaf length (13.13 cm) and width (8.93 cm) was noticed in T8 while the lowest length (7.83 cm) and width (2.6 cm) was observed in ACC# 08149 (T11) and C-2018-12-232 (T4) respectively. This indicated that the T8 possessed broader long leaves, while on contrary, T11 and T4 had narrower short leaves. The 50% flowering dates showed an interesting feature, the accessions ACC# 1249B (T1), NM-9-3-R1 (T2), NM-6-2-R2-B (T3) and C-2018-12-232(T4) had displayed early flowering nature (41 days) whilst the remainder treatments required greater than 47 days (Table 3). The diversified nature in this greater important agronomic trait elaborates the persistence of two distinct groups can be seen within the tested chili accessions viz; the early flowering versus the late flowering habits. Moreover, the number of flowers per axil were compared, four accessions displayed significantly lower number of flowers with one flower per axil (T1, T2, T3 and T4), while T5 and T7 had produced an average of 2.75 flowers per axil. Similar trends were also be seen for the days taken for fruiting. Typically, early flowering accessions had produced physiologically matured pods at around 69 days, conversely, the remainder treatments required greater than 75days to harvest. From these observations, it can be elucidated that both groups of chili accessions require additional 28 days from flowering, to obtain the first harvest, hence it seems that both growth and development of pods constricted within a month. In recent

studies, it has been reported that the commercial elite varieties of chili cultivated by Sri Lankan farmers require 63 to 80 days taken for 50 % flowering while fruiting is expected to obtain between 75 to 110 days after field planting respectively (DOA, 2020). The remarkable finding of our present study elucidates the latent potentials of chili accessions preserved in the PGRC, Gannoruwa in early flowering and fruiting which is considered as preferred traits than those of requiring increased days, thus, these promising lines can be used in crop improvement programs.

Further, the dried seeds were obtained through processing the physiologically matured pods and significant differences were seen for seed traits. The highest 1000 seed weight (5.96 g) was recorded in T3 whilst the lowest (1.64 g) was recorded in T5. These observations may be explained that the T3 was an early flowering with lower number of flowers per axil (one per axil), in contrary, T5 displayed with late flowering (>47d) and produced increased number of flowers per axil (2.75), hence the photosynthetic resource allocation may differ as a result of sink size. Thereby T3 had greater chances for producing larger sized seeds than that of T5 accession. Aminifard *et al*. (2010) state that heavier seeds may provide superiority in germination and the establishment as they have more resources to enable them to emerge from greater depth. Even though same soil and other environmental conditions were been applied throughout this study, chili accessions showed significant differences in most of the morphological characteristics, suggesting that the promising traits displayed by the germplasms were due to the variations in genetic make-up among the accessions. Considering the above morphological and important agronomic traits, chili accessions can be selected for the crop improvement programs.

B. Principal component analysis of morphological traits

Genetic relationship among *Capsicum* spp. was investigated using Principal Component Analysis (PCA) that is used to describe grouping of variables. The Eigenvalues revealed that the first 3 principal components accounted for 73.12% of the total variance. Component 1, 2, and 3 accounted respectively, for 45.37%, 19.45%, and 7.93 % of the total variance (Table 4).

Table 4: Eigenvalues and percentage of total variance explained by the first 3 components of PCA

	Component 1	**Component 2**	**Component 3**
Eigen values	11.89	5.05	2.06
Variance explained %	45.37	19.45	7.93
Cumulative variance %	45.37	65.18	73.12

Table 3: Average performance of quantitative characteristics in Capsicum accessions used in this study

Treatment	**Plant height (cm)**	**Mature leaf length (cm)**	**Mature leaf width (cm)**	**Days taken to flowering (Days)**	**No. Flower/Axil**	**Days taken to fruiting (Days)**	**1000 seed weight (g)**
T1	56.2±0.8^{a}	8.5±0.1^{ab}	2.8±0.1^{a}	43.0±1.1^{ab}	1.0±0.0^{a}	72.0±1.1^{ab}	3.98±0.052^{d}
T2	52.5±1.0^{b}	9.8±0.1^{abc}	3.2±0.1^{a}	40.5±0.5^{a}	1.0±0.0^{a}	69.5±0.5^{a}	4.56±0.083^{e}
T3	74.5±0.6^{a}	11.4±0.1^{cde}	4.2±0.1^{ab}	40.75±0.5^{a}	1.0±0.0^{a}	69.5±0.5^{a}	5.96±0.077^{f}
T4	52.5±1.0^{b}	8.5±0.0^{a}	2.6±0.0^{a}	40.0±0.0^{a}	1.0±0.0^{a}	68.0±0.0^{a}	2.48±0.182^{b}
T5	40.0±1.0^{c}	10.9±1.5^{bcd}	6.3±1.4^{cd}	50.0±1.0^{c}	2.7±0.2^{c}	79.0±1.0^{c}	1.64±0.124^{a}
T6	41.5±0.6^{c}	11.7±0.2^{cde}	7.7±0.2^{cd}	49.2±1.2^{c}	2.2±0.2^{c}	79.2±1.2^{c}	3.47±0.102^{c}
T7	35.2±0.6^{d}	12.1±0.2^{cde}	8.1±0.2^{cd}	48.0±1.1^{bc}	2.7±0.2^{c}	78.0±1.1^{c}	3.65±0.111^{cd}
T8	35.2±0.7^{d}	13.1±0.1^{e}	8.9±0.3^{d}	50.0±1.1^{c}	2.5±0.3^{c}	78.0±1.1^{c}	2.43±0.103^{b}
T9	57.7±0.5^{a}	11.6±0.2^{cde}	6.4±0.2^{bc}	47.5±1.4^{bc}	2.5±0.3^{c}	75.5±1.4^{bc}	3.61±0.078^{cd}
T10	56.2±0.5^{a}	11.4±0.1^{cde}	6.4±0.1^{bc}	50.5±1.4^{c}	2.2±0.2^{c}	80.5±1.4^{c}	3.63±0.043^{cd}
T11	42.2±0.8^{c}	7.8±0.1^{a}	2.8±0.1^{a}	49.2±1.2^{c}	2.5±0.3^{c}	76.2±1.2^{bc}	2.89±0.133^{b}
T12	55.7±0.5^{ab}	12.0±0.2^{cde}	7.0±0.2^{cd}	49.0±0.0^{c}	2.0±0.4^{ab}	78.0±0.0^{c}	3.43±0.040^{c}

The values are means of replicates ± standard error mean (SEM); Within a column, means followed by the same letter are not significantly different at p=0.05.

Table 5: Values represent the correlation coefficients for the three first principal components in the chili accessions. Correlation with absolute values ≥0.5 in bold, Principal Component with the Eigenvalue of <3 was not considered

Traits	**Component 1**	**Component 2**
Seed beak prominence	**.961**	.241
Corolla color	**.961**	.241
Seed shape	**.946**	-.256
Calyx margin	-.946	.256
Leaf color	-.906	-.221
Days to flowering	**.857**	.242
Days to fruiting	**.835**	.283
Number of flowers axil	**.772**	.326
Plant growth habit	-.719	-.674
Leaf shape	-.719	-.674
Mature leaf width	**.658**	.534
Fruit set	-.570	-
Nodal anthocyanin	-.479	-.142
Mature leaf length	.460	.341
Branch habit	-.446	-
Leaf pubescence	.362	-
Fruit color maturity	.283	**.927**
Fruit shape	.283	**.927**
fruit shape at blossom end	.262	**.833**
Fruit position	-.450	**.798**
Plant height	-.386	-.708
Fruit color intermediate	-	**.620**
fruit shape at pedicel attachment	.253	**.606**
Flower position	.486	-.600
Thousand seed weight	-.371	-.474
Stem pubescence	.128	-.306

The first principal component (PC1) was positively correlated with traits (Table 5) such as seed beak prominence (r=0.961), corolla color (r=0.961), seed shape (r=0.946), days to flowering (r=0.857) and days to fruiting (r=0.835) and a mild positive correlation was observed in mature leaf width (r=0.658). This suggests that these traits vary together, if one increases the remaining one tend to increase as well. Based on the correlation of 0.961 and 0.946, this PC1 is primarily a measure of seed beak prominence, corolla color and seed shape. Calyx margin (r= -0.946), leaf color (r= - 0.906), plant growth habit (r= -0.719), leaf shape (r= -0.719), fruit set (r= -0.570), nodal anthocyanin (r= -0.479), branch habit (r= -0.446), fruit position (r= -0.450), plant height (r= -0.386) and thousand seed weight (r= -0.371) were negatively correlated with PC1. The second principal component (PC2) was positively correlated with fruit color maturity (r=0.927), fruit shape (r=0.927), fruit shape at blossom end (r=0,833) and fruit position (r=0.798) and had mild positive correlation with fruit color intermediated (r=0.620) and fruit shape and pedicel attachment (r=0.606). Negative correlation was showed by seed shape (r= -0.256), leaf color (r= -0.221), plant growth habit (r= -0.674), leaf shape (r= -0.674), nodal anthocyanin (r= -0.142), plant height (r= -0.708), flower position (r= -0.6), thousand seed weight (r= -0.474) and stem pubescence (r= -0.306) (Table 5).

C. Hierarchical cluster analysis of quantitative traits

The genotypes were grouped into two major clusters and 4 sub-clusters. Cluster one contained

2 sub-clusters and one of them had 5 accessions (one *C. frutescens* accession *and* four *C. chinense* accessions). Though accession ACC# 08149 belongs to *C. frutescens* species, it showed close relationship with *C. chinense*. The other one was further divided into two sub-clusters where each of them contained 3 accessions (three *C. frutescens* and three *C. annuum*). Cluster 2 had only one accession (NM-6-2-R2-B) that was *C. annuum* (Figure 1).

D. Hierarchical cluster analysis of qualitative traits

Cluster analysis of qualitative traits grouped genotypes into two major clusters and they were further divided into 3 sub-clusters. Cluster one contained 2 sub-clusters, each having 4 accessions (4 *C. frutescens* accessions *and* 4 *C. chinense* accessions) and cluster 2 consisted 4 accession (*C. annuum)*. These findings confirm that there are three species of chili as *C. annuum* (Amu miris), *C. chinense* (Nai miris) and *C. frutescens* (Kochchi miris) conserved at PGRC gene bank which was initially identified as two species *C. annuum* (Amu miris) and *C. frutescens* (Nai miris and Kochchi) (Figure 2). The distance parents that have different genetic constitution could be used in breeding programs in future.

E. Development of species identification key

Plant Genetic Resources Centre, Gannoruwa is the main germplasm collection center of Sri Lanka. So far PGRC has developed species identification keys for several crops but not for chili germplasms. In this research, species identification keys for chili was developed (Figure 3) by studying the published scientific literatures (Sudré *et al*., 2010; Mongkolporn and Taylor, 2011; Ibiza *et al*., 2012; Ballina-Gomez *et al*., 2013; Carrizo *et al*., 2013; Nsabiyera *et al*., 2013; Occhiuto *et al*., 2014).

If any chili accession has Calyx annular constriction present, intermediate flower and fruit position, calyx small teeth, seed shape circular with fish mouth morphological characters, it can be grouped as ***C. chinense*** species while the chili plant with calyx annular constriction absent, teeth lacking, pedicel slender, erect flower, seed shape teardrop morphological characters, it can be ***C. frutescens*** species.

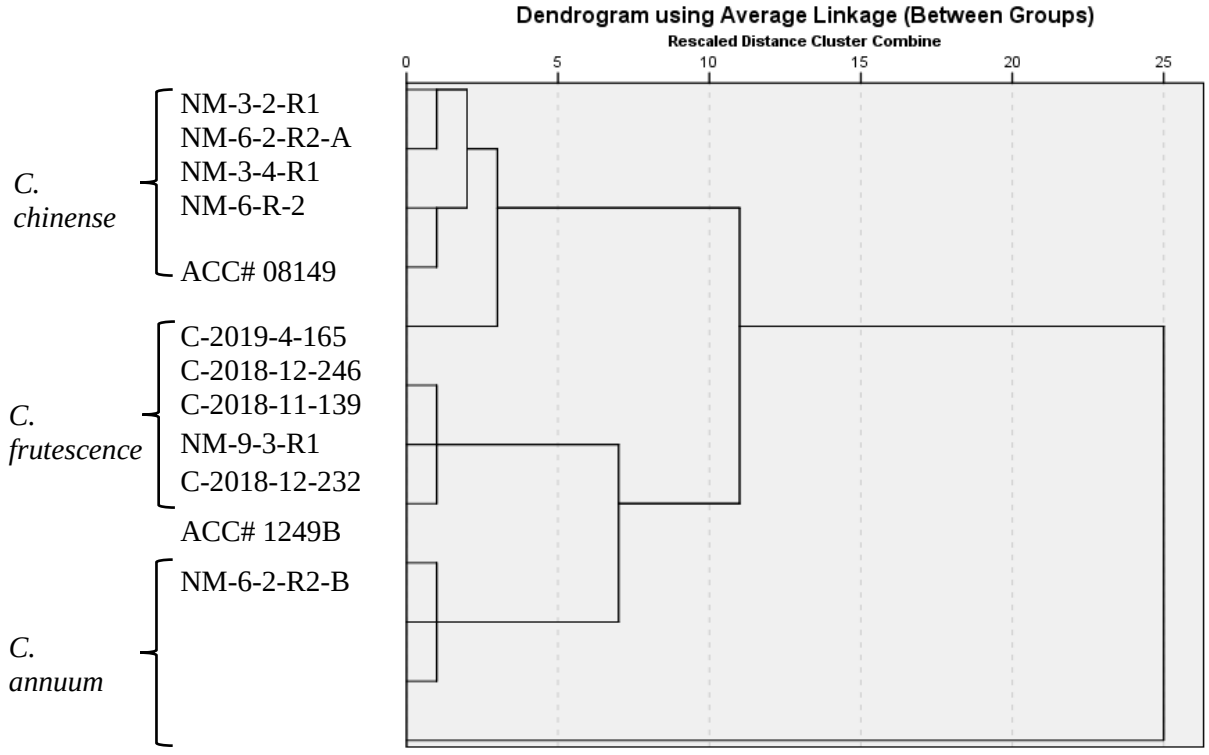

Figure 1: Dendrogram generated based on quantitative characteristics in 12 *Capsicum* accessio

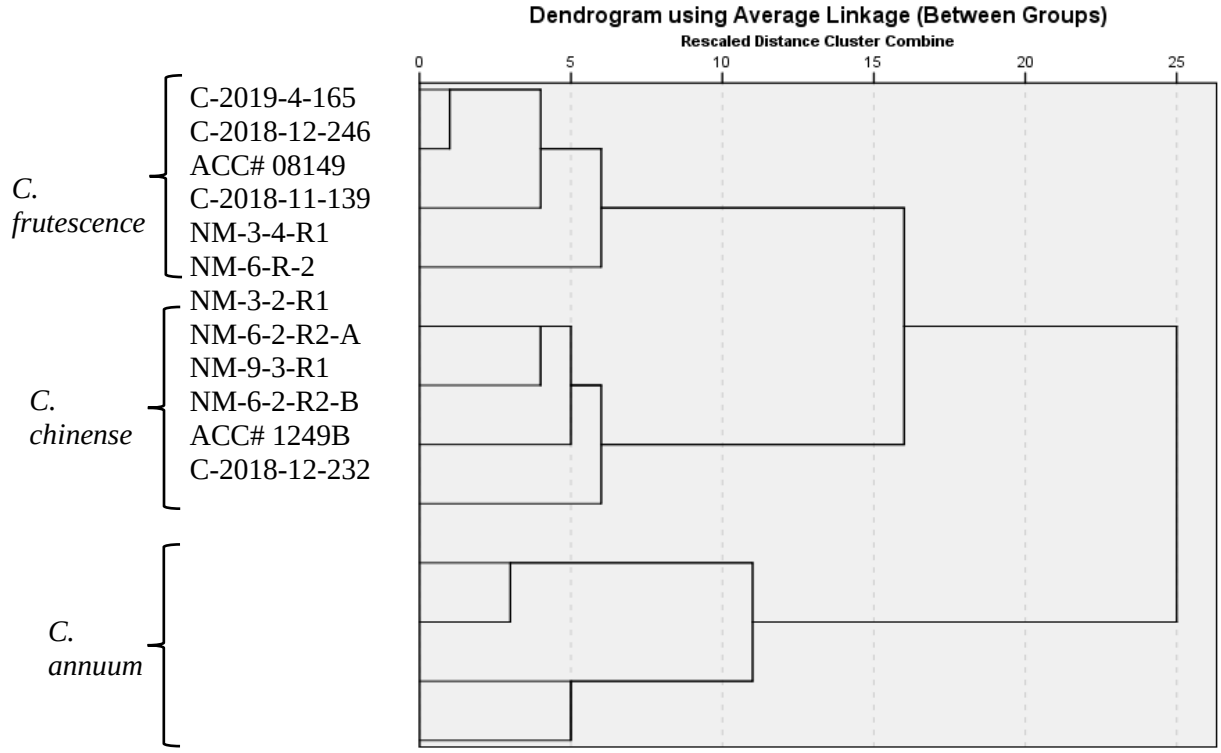

Figure 2: Dendrogram generated based on qualitative characteristics in 12 Capsicum accessions

a. Calyx annular constriction present, intermediate flower and fruit position, calyx small teeth, seed shape circular with fish mouth …..……..….**1. *C. chinense***
a. Calyx annuler constriction absent
b. Calyx teeth lacking, pedicel slender, erect flower, seed shape teardrop…….**2. *C. frutescens***
b. Calyx large teeth, pendent and erect flower position, seed shape reniform……...…….**3. *C. annuum***

Figure 3: Species development key for chili

Chili plant having calyx annular constriction absent, small teeth, pendent and erect flower position, seed shape reniform morphological characters, it can be ***C. annuum*** species.

After developing this key diagram, it was used to separate the accessions in to species. This developed "key" is useful for identify chili species by looking morphological characters. This technique is quick, scientific, consistent and inexpensive. Also this characterization is useful for varietal improvement, seed certification, and seed production programs.

IV. CONCLUSION

Here, a method for detecting species identification key for chili was developed based on the morphological characteristics. This provides the first stage toward linking plant growth and morphological traits in which seed traits can be used as a powerful tool to determine the types of crop plant either belonging to one of such three groups, viz; *Capsicum annuum* (Amu miris), *Capsicum chinense* (Nai miris) and *Capsicum frutescens* (Kochchi miris). Although, the present analysis based on morphological traits may be crude and time-consuming, while there are precise and rapid cutting-edge technologies for genetic diversity analysis, dealing with DNA molecular markers and further database generated through

such techniques will increase the precision for chili characterization in future.

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