



## ESTIMATION OF GENETIC DIVERGENCE IN CHILLI PEPPER (*CAPSICUM ANNUUM* L.) GENOTYPES FOR MORPHOLOGICAL AND FRUIT TRAITS UNDER HOT CLIMATE OF UMERKOT, SINDH

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### ABSTRACT

Thirty-two genotypes of chilli from three different sources (local dandcuts, AVRDC and PGRI PARC) were evaluated for the presence of genetic divergence among them for different agro-economic traits i.e., plant height, stem girth, fruit length, fruit girth, number of fruits per plant, number of seed per fruit, fresh fruit weight and dry fruit weight at Arid Zone Agricultural Research Institute, Umerkot, Sindh during 2018. All these genotypes were found to be significantly different from each other for all the studied traits. For further diversity analysis, multivariate analyses like principal component analysis (PCA) and Cluster analysis were also performed to figure out the traits responsible for maximum variability and grouping of genotypes according to their similarities and differences for their future utilization in chilli breeding programs. PCA analysis showed that first four PCs exhibited 82.79% of the total variability among these 32 chilli genotypes. Cluster analysis showed four different sub-clusters and the genotypes from every single source merge in each sub-cluster, thus showing that chilli genotypes are from different geographical backgrounds. Based on these results, the chilli genotypes can be further utilized in future chilli improvement programs in Pakistan. Genotypes AZRI-DS-14, AZRI-DS-01 and AVPP-9804 showed better performance.

**Keywords:** chilli pepper, cluster analysis, genetic divergence, principal component analysis (PCA)

### INTRODUCTION

Chilli pepper or hot pepper (*Capsicum annuum* L.), a diploid species having chromosome  $2n=24$  is an important vegetable-cum-condiment crop of family *Solanaceae* and genus *Capsicum* (Zulaikha, 2019). Genus *Capsicum* has five cultivated and twenty five wild species (Moscone *et al.*, 2007). It is an important cash crop, used both as fresh vegetable and also processed and used as a condiment (Votava *et al.*, 2005). Due to its pungency this crop is considered as a chief ingredient in culinary preparation in Asian countries like Pakistan, India, Malaysia and Bangladesh (Farhad *et al.*, 2008). Besides its culinary usage chilli is also used as fresh food, condiments in dry form, pickle, sausages and as a guacamole in Mexico (Orellana-Escobedo *et al.*, 2013). Chilli pepper is nutritionally rich in

carbohydrates, lipids, proteins, mineral salts and fibers (Wahyuni *et al.*, 2013; El-Ghorab *et al.*, 2013). It is also an important source of vitamins (A, B, C and K) and phytochemical compounds like carotenoids, flavonoids, ascorbic acid and capsaicinoids (El-Ghorab *et al.*, 2013). These vitamins and phytochemicals are useful for cure of chronic disorders like asthma, cancer, used as immune system stimulator, antioxidant and antimicrobial (do Nascimento *et al.*, 2013). Fresh green chillies have more vitamin C than citrus and fresh red chilli pepper have more vitamin A than carrot (Osuna-García *et al.*, 1998; Marin *et al.*, 2004).

Total area of the world under chilli cultivation is about 1856.641 thousand hectares with a production of 4625.833 thousand tones (FAOSTAT, 2017). In Pakistan it was cultivated on 47.349 thousand hectares with a production of 126.943 thousand tones (MNFS and R, 2018-19). Sindh contributes 36.067 thousand hectares

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in area and 108.578 thousand tones in production followed by Punjab 6.842 thousand hectares in area and 11.698 thousand tones in production, Baluchistan 4.112 thousand hectares in area and 6.285 thousand tones in production and KPK contributes 0.328 thousand hectares in area and 0.382 thousand tones in production (Agriculture Statistics of Pakistan, 2019). Chilli shows good response to warm climatic situations but vulnerable to extreme temperature regimes (Dahal *et al.*, 2015). Kunri is the major area of chilli production in Sindh Province that is known as Asia's biggest market (Wahocho *et al.*, 2016). Presence of genetic diversity is a pre-requisite for improvement in any crop. However, varieties developed lose their genetic potential over the period of time due to continuous cultivation and genetic erosion, therefore, leading to decrease in per acre yield of the available genotypes. However, because of the popularity of the crop and extended history of cultivation at different parts of the world, sufficient genetic variability is present that can be utilized for generation of new and improved varieties (Pujar *et al.*, 2017). For varietal improvement, exploring the potential of base population and selection efficiency are the key factors. Therefore, current study was carried out to investigate thirty-two genotypes of chillies for different agronomic and fruit traits. This study will be helpful in designing a breeding program for new varieties development that

should sustain and produce higher yield than existing varieties.

## MATERIALS AND METHODS

### Experimental site

Current study was carried out at field area of Arid Zone Research Institute (AZRI), Pakistan Agricultural Research Council, Umerkot, Sindh, Pakistan. Experimental site was situated at 25.15°N and 69.32°E.

### Experimental material and design

Current experiment was carried out by using 32 genotypes; out of which sixteen genotypes were local dundicut (DS) selections, eight genotypes from Asian Vegetable Research and Development Center (AVRDC) and seven genotypes were from Plant Genetic Resources Institute (PGRI, PARC, Islamabad). Nursery was raised on raised bed at field area of Arid Zone Research Institute (AZRI) Umerkot, Pakistan Agricultural Research Council. Seedlings were then transplanted in field in mid of April, 2018 in a randomized complete block design (RCBD) having three replications. Row to row distance was maintained at 75 cm and plant to plant distance of 30 cm (Marama *et al.*, 2009). Four rows were transplanted for each genotype with a row length of 4 m. All agronomic and crop management practices were performed at different crop stages.



**Map.** Location map of chilli experiment at PARC-AZRI, Umerkot, Sindh, Pakistan

### Agronomic and fruit characterization of studied chilli genotypes

Data regarding vegetative and fruit traits from five randomly selected plants for all genotypes were collected from all three replications. Data for plant height (cm) was computed with the help of meter rod, number of main branches was counted manually, stem girth (mm), fruit length (mm) and fruit width (mm) were computed with the help of vernier caliper and number of fruits per plant, number of seeds per fruit, fruit fresh weight (g) and fruit dry weight (g) were also determined.

### Statistical analysis

Analysis of variance (ANOVA), descriptive statistics, cluster analysis and principal component analysis was performed by using R-Studio Version 1.2.1335.

**Table 1.** List of chilli pepper genotypes with the sources used in current experiment

Genotypes	Source
AZRI-DS-01	Local, PARC-AZRI
AZRI-DS-02	Local, PARC-AZRI
AZRI-DS-03	Local, PARC-AZRI
AZRI-DS-04	Local, PARC-AZRI
AZRI-DS-05	Local, PARC-AZRI
AZRI-DS-06	Local, PARC-AZRI
AZRI-DS-07	Local, PARC-AZRI
AZRI-DS-08	Local, PARC-AZRI
AZRI-DS-09	Local, PARC-AZRI
AZRI-DS-10	Local, PARC-AZRI
AZRI-DS-11	Local, PARC-AZRI
AZRI-DS-12	Local, PARC-AZRI
AZRI-DS-13	Local, PARC-AZRI
AZRI-DS-14	Local, PARC-AZRI
AZRI-DS-15	Local, PARC-AZRI
AZRI-DS-16	Local, PARC-AZRI
AZRI-DS-17	Local, PARC-AZRI
AVPP-9804	AVRDC
AVPP-1346	AVRDC
PBC-518	AVRDC
AVPP-0506	AVRDC
AVPP-0705	AVRDC
AVPP-0701	AVRDC
AVPP-9704	AVRDC
AVPP-0903	AVRDC
Pr-20530	PGRI, PARC
Pr-20528	PGRI, PARC
Pr-28340	PGRI, PARC
Pr-28345	PGRI, PARC
Pr-20531	PGRI, PARC
Pr-16161	PGRI, PARC

AZRI-DS = Arid Zone Research Institute- dundicut selection, AVRDC = Asian Vegetable Research and Development Centre, PGRI = Plant Genetic Resources Institute

## RESULTS

### Morphological variation

Knowledge about the genetic divergence is not only helpful in crop improvement program but

also helps in understanding the availability of germplasm diversity to certain area. Analysis of variance showed that considerable diversity is available among studied genotypes (Table 2). All studied traits showed highly significant genotypic mean square values.

There was a great variability observed for quantitative traits that includes agronomic and fruit traits (Table 3). Plant height was ranged from 41.5 to 82.5 cm. Number of main fruiting branches varied from 6 to 10 branches per plant. Stem girth varied from 10 to 32 mm. There was great variability observed for number of fruits per plant that varied from 31 to 259 fruits per plant. There was also considerable variability for fresh and dry fruit weight. Fresh fruit weight ranges from 54 to 382 g per plant while dry fruit weight ranges from 15 to 100 g per plant. There was also variability for fruit length and fruit width. Fruit length varied from 4.3 to 7 mm, while fruit width varied from 6.3 to 23.7 mm (Table 3).

**Table 2.** Analysis of variance (ANOVA), coefficient of variation (CV %) for thirty two studies genotypes

Dependent Variable	MS <sub>g</sub>	MS <sub>g x r</sub>	MS <sub>e</sub>	CV %
PH	171.938	9.399	62.068	4.54
NMB	2.769	0.421	1.833	7.73
SG	57.671	10.018	3.208	15.49
NFPP	6643.622	141.951	130.167	10.05
FFWt	9993.883	514.163	3619.333	13.00
FDWt	679.091	78.137	67.333	16.97
FL	289.000	23.875	262.823	19.68
FW	58.936	1.911	4.875	9.64
NS	1083.005	34.457	729.333	10.89

MS<sub>g</sub> = Mean square for genotypes, MS<sub>g x r</sub> = Mean square for genotypes x replication, MS<sub>e</sub> = Mean square for error, PH = Plant height, NMB = Number of main branches, SG = Stem girth, NFPP = Number of fruits per plants, FFWt = Fresh fruit weight, FDWt = Fruit dry weight, FL = Fruit length, FW = Fruit width (mm), NS = Number of seeds per fruit

### Principal component analysis

The principal component analysis (PCA) is useful statistical attribute from multivariate analysis that can prove useful for future breeding and population development program. First four PCs with eigen value > 1, explained 82.79% variability out of the total variation existed in studied chilli genotypes (Figure 1). Here it can be seen in this figure that maximum variability can be explained by first four principal components. Scree plot explained variance percentage obtained by each component. Maximum variance is accounted for first four PCs after that semi curve line obtained which shows little variance existed in remaining

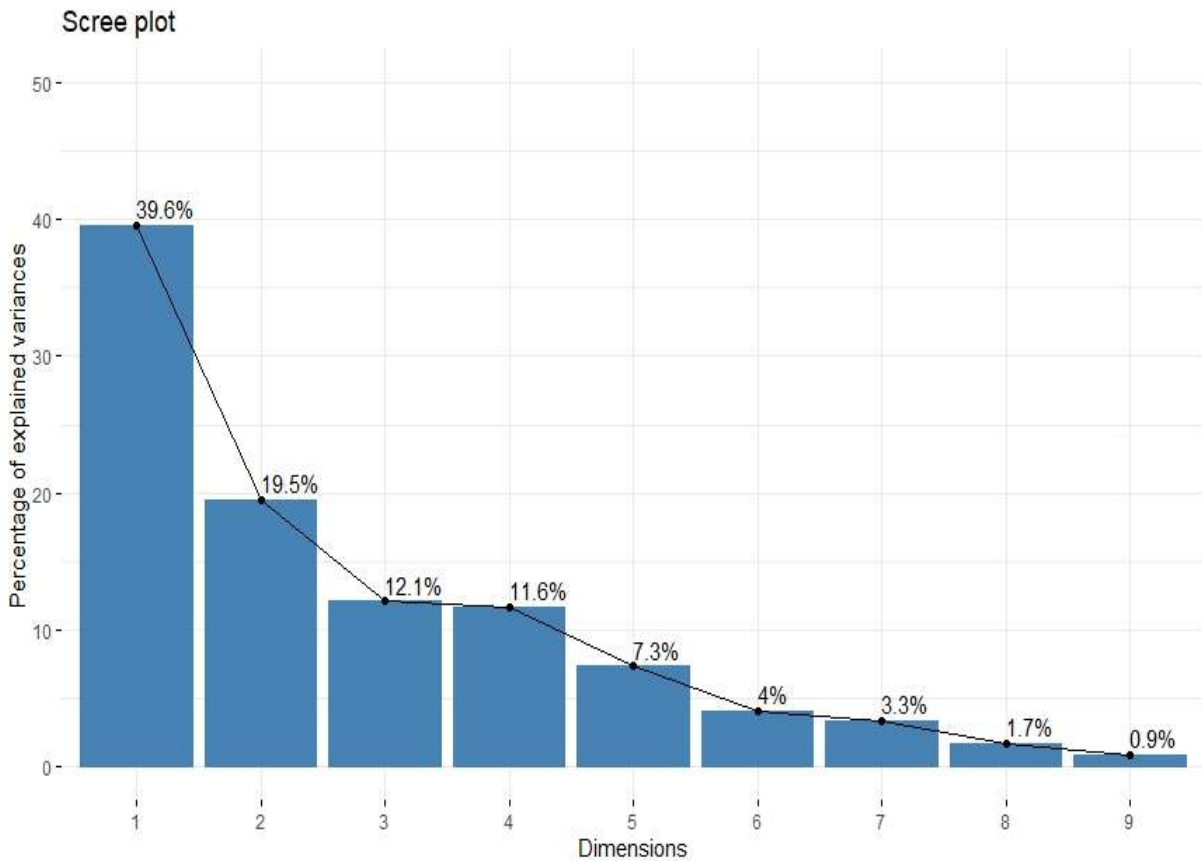
principal components (Figure 2). First eigen vector alone showed variation of about 39.6% out of total explained variation. Principal components results showed all the traits studied had a positive contribution except fruit length. Fruit length has a negative correlation with fruit width, plant height and number of seeds per plant (Figure 2). PC Biplot diagram illustrates the diversity among studied chilli genotypes and

different traits studied in current research (Figure 2). Based on nine morphological and fruit attributes, studied genotypes scattered in four quadrants. Genotypes that are close to each other forming small angle are closely related and having most of the features common to each other. While, genotypes on bi-plot that make an angle more than ninety are diverse genotypes.

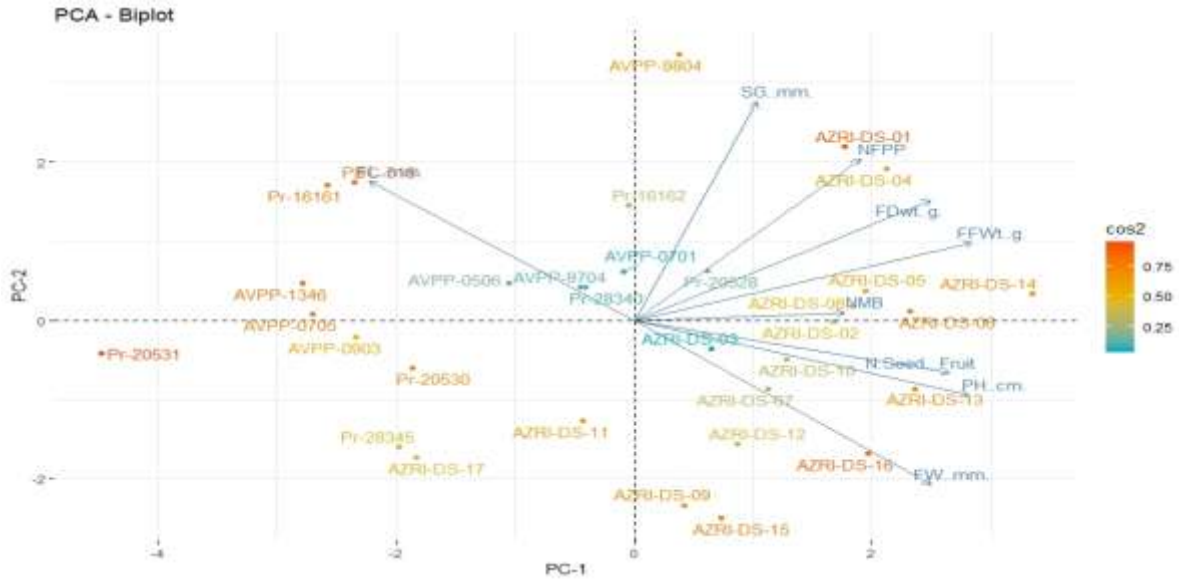
**Table 3.** Descriptive Statistics for thirty two studied genotypes in current experiment

Trait	N	Minimum	Maximum	Mean	Std. Error	Std. Deviation	Variance
PH	96	41.5	82.5	66.53	0.81	7.92	62.79
NMB	96	6.0	10.0	8.47	0.11	1.10	1.20
SG	96	10.0	32.0	20.20	0.51	4.96	24.64
NFPP	96	31.0	259.0	1.18	4.78	46.86	2.20
FFWt	96	54.0	382.0	1.72	6.12	59.98	3.60
FDWt	96	15.0	100.0	51.28	1.67	16.33	266.56
FL	96	4.3	57.0	28.43	1.09	10.72	114.94
FW	96	6.3	23.7	14.12	0.46	4.47	20.01
NS	96	14.0	87.0	54.24	2.01	19.68	387.24
Valid N (listwise)	96						

PH = Plant height, NMB = Number of main branches, SG = Stem girth, NFPP = Number of fruits per plants, FFWt = Fresh fruit weight, FDWt = Fruit dry weight, FL = Fruit length, FW = Fruit width (mm), NS = Number of seeds per fruit



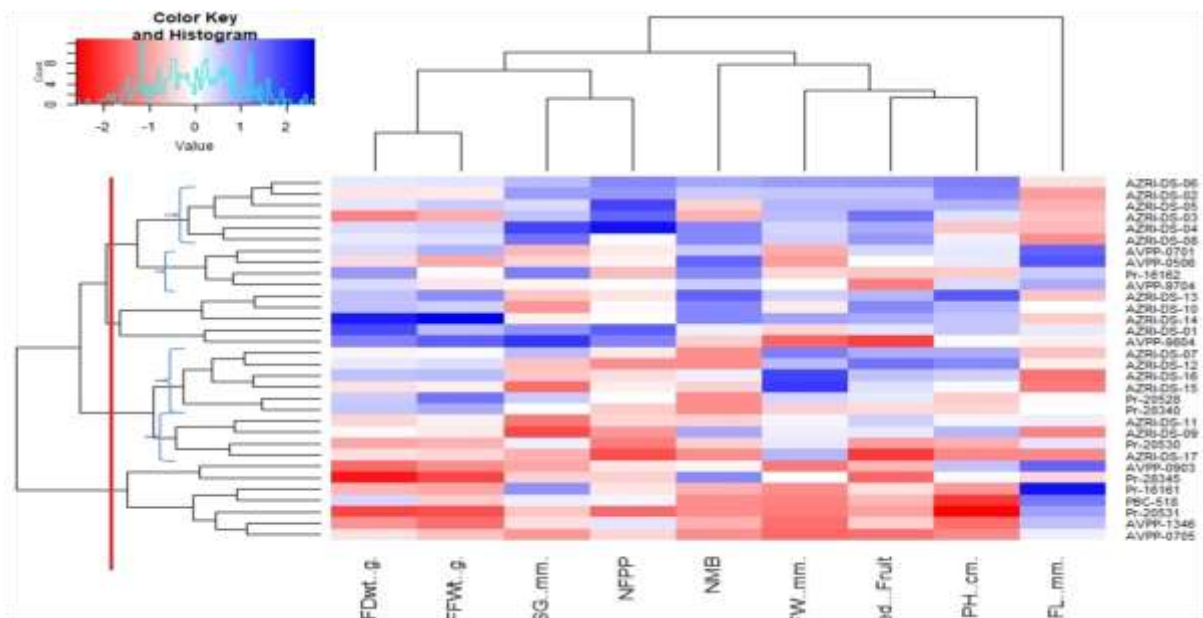
**Figure 1.** Scree plot, showing eigen value (explained variances) on Y-axis and factors (PCs) on X-axis.



**Figure 2.** Principal component analysis. PH = Plant height, NMB = Number of main branches, SG = Stem girth, NFPF= Number of fruits per plants, FFWt = Fresh fruit weight, FDWt = Fruit dry weight, FL = Fruit length, FW = Fruit width (mm), NS =Number of seeds per fruit

**Table 4.** Distribution of thirty two chilli genotypes into four diverse clusters

Clusters	No. of genotypes	Name of genotypes
I	10	AZRI-DS-06, AZRI-DS-02, AZRI-DS-05, AZRI-DS-03, AZRI-DS-04, AZRI-DS-08, AVPP-0701, AVPP-0506, Pr-1612, AVPP-9704
II	05	AZRI-DS-13, AZRI-DS-10, AZRI-DS-14, AZRI-DS-01, AVPP-9804
III	10	AZRI-DS-07, AZRI-DS-12, AZRI-DS-16, AZRI-DS-15, Pr-20528, Pr-28340, AZRI-DS-11, AZRI-DS-09, Pr-2530, AZRI-DS-17
IV	07	AVPP-0903, Pr-28345, Pr-16161, PBC-518, Pr-20531, AVPP-1346, AVPP-0705



**Figure 3.** Cluster heat map generated by UPGMA for diversity analysis of thirty two chilli genotypes. PH = Plant height, NMB = Number of main branches, SG = Stem girth, NFPF = Number of fruits per plants, FFWt = Fresh fruit weight, FDWt = Fruit dry weight, FL = Fruit length, FW = Fruit width (mm), NS = Number of seeds per fruit

### Cluster analysis

Cluster heat map was generated through Euclidean distance matrix computed by UPGMA analysis for nine morphological and fruit traits of thirty two chilli genotypes, to study the diversity among these genotypes acquired from different sources in 2018. Heat map diagram generated from the morphological and fruit data of chilli genotypes studied during cropping season 2018, divided these genotypes into four different clusters (Figure 3). First cluster contains 10 genotypes that can further be divided into two sub-clusters, in sub-cluster I, 6 genotypes fall that are from local dundicut selection source from AZRI i.e., AZRI-DS-02, AZRI-DS-03, AZRI-DS-04, AZRI-DS-05, AZRI-DS-06 and AZRI-DS-08 while in sub-cluster II, genotypes AVPP-0701, AVPP-0506, Pr-16162 and AVPP-9704 Figure 3. Cluster II contains five genotypes viz. AZRI-DS-01, AZRI-DS-10, AZRI-DS-13, AZRI-DS-14 and AVPP-9804. Cluster III contains two sub-clusters, sub-cluster I contains 6 genotypes namely AZRI-DS-07, AZRI-DS-12, AZRI-DS-01, AZRI-DS-16, Pr-20528 and Pr-28340, while sub-cluster II contains four genotypes namely AZRI-DS-11, AZRI-DS-09, Pr-2530, AZRI-DS-17. Cluster IV contains seven genotypes i.e AVPP-0903, Pr-16161, PBC-518, Pr-20531, AVPP-1346 and AVPP-0705. It was observed from that genotypes from different sources merges in mega clusters but these can be found in different sub-clusters on the basis of their diversity.

### DISCUSSION

Chilli is an important vegetable cum cash crop of Sindh province, as it produces high quality red (dry) chilli with its unique export quality. Limited research work has been carried out in vegetables seed sectors, especially in chillies. It is the need of time to study the diversity in this crop especially due to ever changing climatic situations. Diversity study is imperative in any crop improvement and population development program. Enhancement of gene pool diversity and its characterization is a foremost goal of a breeder to strengthen breeding program (Bianchi *et al.*, 2020). Current study was aimed to study diversity in hot pepper (red chilli) genotypes collected from three different sources. Significant diversity was observed for different morphological and fruit related traits. There exists a great variability between different traits like fruit length and weight. Similar findings were also observed by (Castro and Dávila, 2008; Chowdhury *et al.*, 2015; Naegele *et al.*,

2016; Arumingtyas *et al.*, 2017; Moreira *et al.*, 2018). Baba *et al.* (2016) while studying on *Capsicum chinense* also found great diversity for morphological and fruit parameters. They also study different molecular markers to study genetic diversity in available germplasm.

Principal component analysis and cluster analyses are the two important multivariate analysis approaches. This analysis can be of great help for breeders to select genotypes for launching breeding and improvement program. As it can be seen there was considerable variability existed by analyzing our collected germplasm by PCA and cluster analysis. It was observed that genotypes from different sources were placed in different clusters and showed different relationships by principal components on the basis of studied traits. Genotypic selection can be made on these approaches to strengthen our germplasm and to improve our breeding program. Diversity of chilli was also studied by Hasan *et al.* (2014) by cluster analysis through  $D^2$  statistics and Principal component analysis. They also studied that broad spectrum variability can be achieved by selecting genotypes from diverse clusters for different morphological and fruit quality traits. Utilizing distantly related parents can be helpful for population improvement and future breeding program as this approach can provides higher heterosis or transgressive segregants. On the basis of results of current study, it can be established that characterization of available germplasm is important and basic criteria to understand the genetic variability of existing population and also for the development of effective strategies germplasm conservation, improvement and breeding purpose.

### CONCLUSION

Present study provides us the considerable information for selection of diverse parents to improve our existing cultivars and breed new cultivars with traits of our interests. Some of the study genotypes like AZRI-DS-14, AZRI-DS-01 and AVPP-9804 showed better performance for different fruit traits and showed diversity from other genotypes. These genotypes can further have potential to use in future breeding programs for improving fruit yield and fruit quality traits for enhancement of ultimate yield of the chilli crop.

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## AUTHOR'S CONTRIBUTION

**A. Memon:** Performed the experiments and data collected

**R. Ahmad:** Analyzed data and wrote the manuscript

**M. S. Debar:** Design the study and review manuscript

**A. K. Pathan:** Design the study and Review manuscript

**D. Ibrar:** Analyzed data and wrote the manuscript

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