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Original Research Paper

Assessing the genetic diversity of squash (*Cucurbita pepo* L.) genotypes based on agro-morphological traits and genetic analysis

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ABSTARCT

An experiment was conducted to estimate the genetic variability of 15 indigenous and exotic squash genotypes assessing 18 quantitative and 8 qualitative traits. Results showed that the accessions have high variability in qualitative traits like fruit size, fruit shape, fruit skin colour, lustre and fruit productivity, which allowed selection for considerable gains in these characteristics. The quantitative traits such as fruits yield per plant, fruit weight, length, diameter and total yield per hectare showed the greater phenotypic coefficient of variation (PCV) along with higher heritability which can helps to identify desirable genotypes. The obtained significant and positive correlation between fruit yield with number of leaves, nodes, fruit length, weight and number could assist in selection to improve this crop. Cluster analysis resulted in the formation of 4 groups, confirming the genetic variability among the studied genotypes. Eventually, the attained PCA analysis result revealed that the number of fruits per plant, fruit yield per plant, fruit length and days to first female flowering are the most discriminating traits which are accelerating the variability in squash genotypes. On the basis of the yield and its attributing traits, First Runner is the best genotype suited in this environment.

Keywords: Genetic analysis, genetic variability, heritability, morphological traits and squash

INTRODUCTION

Squash (*Cucurbita pepo* L.) is an economically important species of the Cucurbitaceae family that represents one of the most primitive genera (*Cucurbita)* in the plant kingdom (Tadmor *et al.*, 2005). Squash is monoecious vegetable crop and familiar with its different traditional name like Zucchini (Italy); Cucuzza (Saudi Arabia), Courgette (America); Marrow (Ireland and Britain) and Baby marrow (South Africa). It is grown throughout the temperate, sub-tropical, and tropical regions, native to eastern United States and Mexico and also cultivated worldwide for its fruits (Bisognin, 2002). The major economic value of this crop is based mainly on the culinary use of immature fruits which have relatively high nutritional and medicinal value as compared to other vegetable crops. Its nutritional profile consists of various organic compounds, nutrients, vitamins and minerals, that are responsible for providing all its impressive health benefits (Kulczynski and Gramza-Michałowska, 2019). It is also a very good source of carotenoids, important anti-inflammatory and anti-oxidant compounds (Deppe, 2015) and because of its low caloric value treated as weight loss diets (Fageria *et al.*, 2012). So, keeping its importance in mind, increase the global production is one of the important ways to ensure food security.

Bangladesh is one of the most densely populated country in the world having over 160 million people and based on its current growth trends a projected population will be over 200 million by 2050 (USAID, 2017). To meet up the food demand for its uprated population, increasing the crop production in per unit areas of land is the most effective ways to ensure the food security. Squash is one of the important vegetable crops which can assure the nutritional security from its present nutritional shortage (per capita deficiency of vegetables 158 g) in Bangladesh (Anon., 2018). Topographically, Bangladesh has diverse land area which is favorable for the crop diversification and production, however, squash can grow easily in any types of soil even in unproductive and marginal land areas. In addition, more economic growth can be achieved by producing vegetables like squash which will ultimately uplift the socio-economic condition of the farmers. Thus, there is urgent need to initiate research on squash especially for its vertical expansion and varietal improvement. Although, squash is becoming important vegetable crop in Bangladesh, there is little information available about its improvement and till date only a single variety has been recommended for winter season. In Bangladesh, few researchers have taken initiatives for studying its growth and effects of fertilizers on it (Akhter *et al.*, 2018; Baby *et al.*, 2021) but genetic variability study has not been taken up yet. Breeding for high-yielding crops require information available on the germplasm and the relationship among the agronomic traits as well as the degree of environmental influence (El-Hadi *et al.*, 2014). For its crop improvement, determining the extent of genotypic and phenotypic variability among geographical areas is important (Muralidhara and Narasegowda, 2014). Quantitative and qualitative determination (morphological characterization) of the degree of variation of traits present in genetic resources is important for vegetable breeding programs (Balkaya *et al.*, 2010; Gomes *et al.*, 2020). Morphological characterization is the first step followed by quantitative traits in the description and classification of genetic resources (Balkaya *et al.*, 2010). However, only a few studies have focused on variability analysis in relation to morphological and yield contributing quantitative traits with squash accessions. Therefore, the present research has been undertaken for the improvement of squash by assessing its genetic variability traits.

In Bangladesh, squash cultivation in summer season is challenging because of the severe attack of pests and diseases, excessive light and temperature, high rainfall and high labour cost etc. Meanwhile, a very few research works relating to its adaptability and variability have been conducted in Bangladesh especially in Sylhet region where huge amount of land has remained fallow (14% of the total land) for a long time (BBS, 2018). So, there is a great opportunity to increase squash production in this region to meet up the vegetable and nutritional requirement of the country. Considering the above points of view, the present study has been under taken to know the extent of genetic variability, heritability and genetic advance for different traits of squash genotypes in Sylhet region.

MATERIALS AND METHODS

The experiment was conducted at the Research field of the Department of Genetics and Plant Breeding, Faculty of Agriculture, Sylhet Agricultural University, Bangladesh during the period October 2019 to January 2020. Fifteen indigenous and exotic genotypes (Table 1) of squash were used in this experiment that were collected from the different parts of Bangladesh as well as from the other countries.

The experiment was laid out in a Randomized Complete Block Design (RCBD) with three replications. The experiment was divided into three blocks and each consisted of 15 plots. Each unit plot size was 1 x 2.3 m^2. Altogether, there were 45 unit plots in experiment and required 300 m^2 land. Both row to row and plot-to-plot distances were 0.5 m. The treatments were randomly assigned to each of the block. Each unit plot had 5 pits and in each pit 2 seeds were sown. After germination only one plant was allowed to grow. The land was prepared by ploughing and cross ploughing and different intercultural operations were accomplished according to recommended BARI Squash variety (BARI, 2018). The data were recorded based on 18 quantitative yield contributing traits i.e. plant height in cm at first harvest (PH), stem diameter in cm at first harvest (SD), number of leaves at first harvest (NL), number of nodes at first harvest (NN), days to flower bud initiation (DFBI), days to first male flowering (DFMF), days to first female flowering (DFFF), number of male flowers from flowering to last harvest (NMF), number of female flowers from flowering to last harvest (NFF), viable pollen in percentage (VP), days to first harvest (DFH), nodes at first fruit harvest (NFFH), fruit length in cm (FL), fruit diameter in cm (FD), fruit weight in g (FW), number of fruits per plant (NFPP), fruit yield per plant in Kg (FYPP), total yield in t/ha (TY) and 8 qualitative traits *i.e.* plant vigor, pubescence, stem shape, flower color, fruit size, fruit shape, fruit skin color and luster.

The recorded data on various parameters were analyzed to find out the statistical significance of the experimental results. Mean and standard deviation were calculated using Microsoft Excel software 2010.

Table 1. Name and source of the Squash genotypes used in the experiment

Genotypes	Name of the genotypes	Origin	Remarks
G1	First Runner	South Korea	Indigenous
G2	Alaska	Australia	Indigenous
G3	Blossom House	Netherlands	Indigenous
G4	Balam House	USA	Indigenous
G5	Cheonlima	South Korea	Indigenous
G6	Hungnong Squash	South Korea	Indigenous
G7	Runner	USA	Indigenous
G8	SQ-001	Australia	Exotic
G9	SQ-002	Australia	Exotic
G10	SQ-003	Australia	Exotic
G11	SQ-004	Australia	Exotic
G12	SQ-005	Australia	Exotic
G13	SQ-006	Australia	Exotic
G14	SQ-007	Australia	Exotic
G15	SQ-008	Australia	Exotic

The significance of the difference between treatment means, coefficient of variation (CV) was calculated by the Least Significance Difference (LSD) test for the interpretation of the results (Gomez and Gomez, 1984). Then the tabulated results were analyzed using one-way analysis of variance (ANOVA) and statistical differences between the means were estimated using Duncan's multiple range test (DMRT) at 1% or 5% or 0.1% probability with the help of statistical "R" software.

Estimation of Genetic parameters

Estimation of genotypic and phenotypic variances: Genotypic and phenotypic variances were estimated according to the formula given by Johnson *et al.* (1955).

Estimation of coefficient of variability (genotypic and phenotypic coefficient of variation): Both phenotypic and genotypic coefficient of variability for all characters w estimated using the formula of Burton (1952). PCV and GCV were classified into three categories *viz*., Low (< 10%), Moderate (10-20%) and High (> 20%) as suggested by Sivasubramanian and Madhavamenon (1973).

Heritability in broad sense (h^2_{bs}): The broad sense heritability (h^2_{bs}) was estimated for all characters as the ratio of genotypic variance to the total of phenotypic variance as suggested by Hanson *et al*., (1956). Heritability estimates in cultivated plants could be placed in the categories viz. as Low (0-30%), Moderate (30-60%) and High (>60%) as suggested by Robinson (1966).

Genetic advance (GA): The expected genetic gain or advance for each character was estimated by using the method suggested by Johnson *et al*., (1955). Genetic advance was classified as high (>20%), moderate (10-20%) and low (<10%). Further the Genetic advance as per cent of mean was computed by using the formula which was given by Burton (1952). Genetic advance as per cent mean was categorized into groups *viz*., Low (< 10%), Moderate (10-20%) and High (> 20%) as suggested by Johnson *et al.* (1955).

Correlation estimation

Simple correlation coefficient (r) among 12 important parameters of Squash accessions was estimated according to Singh and Chaudhury (1985). Again, cluster analysis (CA) was carried out according to Mahalanobis (1936). It divides genotypes into groups on the basis of a data set into some number of mutually exclusive groups. Furthermore, principal component analysis (PCA) was computed from correlation matrix and genotype scores obtained for the first components with roots greater than unit (Jeger *et al.,* 1983). It provides two dimensional plots, which helps in separating different populations involved. Contribution of the different characters towards variability was discussed from the latent vectors of the first three principal components. However, Mean data for each character was subjected to multivariate analysis techniques *viz*., principal component analysis (PCA), cluster analysis (CA) and also the simple correlation coefficient analysis were done by computer using the STATA 14.0 software.

RESULTS AND DISCUSSION

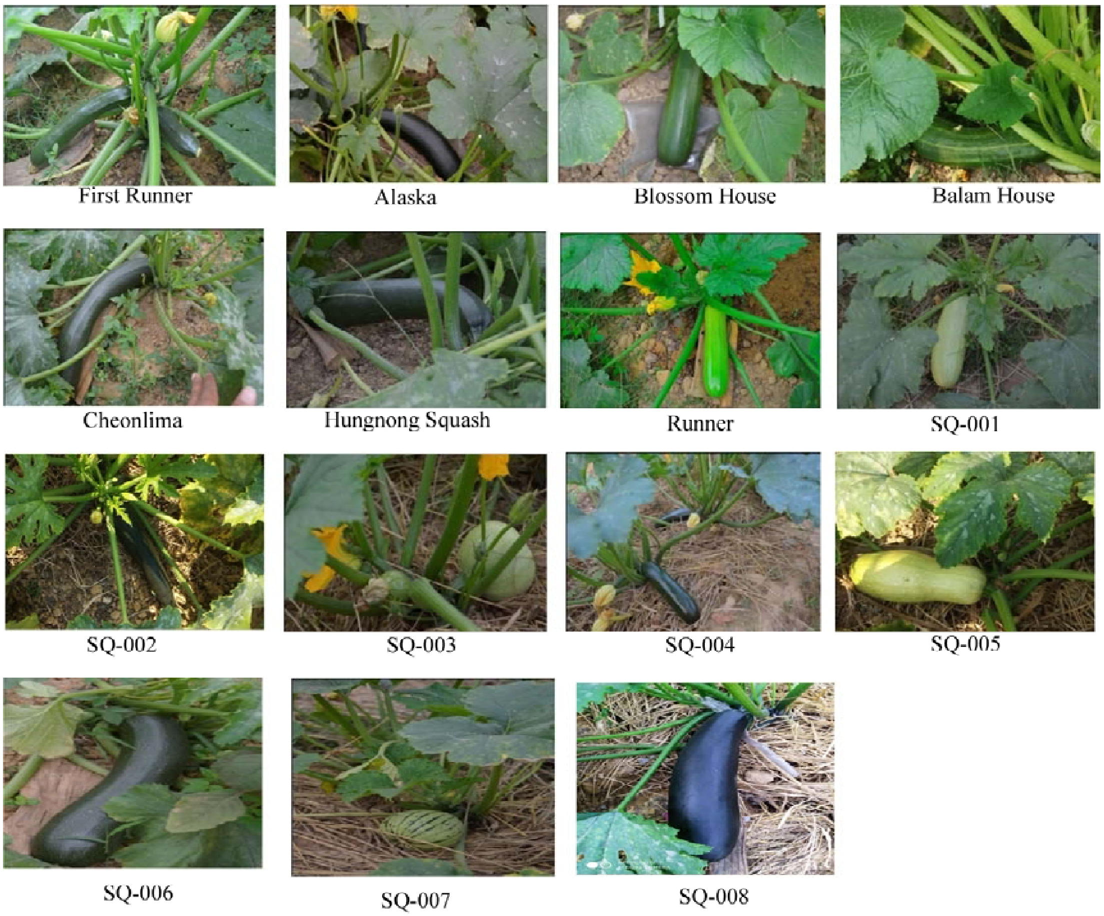
Mean performance or genotypes for vegetative characters

In this experiment, fifteen indigenous and exotic squash genotypes have been characterized according to morphological traits and genetic analysis. Although morphological characteristics depends on its external factors but it is parallelly important to support these morphological variations along with their genetic studies. Results of mean performance of different squash genotypes based on different agronomic and yield contributing traits indicated that there was a significant difference in mean performance among all the genotypes. This difference could be resulted from the genetic variation among the studied squash genotypes which is also supported with the results of other previous studies on squash (Gomes *et al.*, 2020; Tsivelikas *et al.*, 2009; Villanueva-Verduzco *et al.*, 2020). A wide genetic diversity was also reported in the experimental results of Egusi-melon (Olaniyi *et al*., 2011) and cucumber (Arunkumar *et al*., 2011).

In case of vegetative characters, results showed a great significant variation for all the characters among the squash genotypes (Table 2). The highest plant height at first harvest was found in SQ-002 (36.05 cm) and the lowest was in Balam House (32.17 cm). Diameter of stem during first harvest was highest in First Runner (13 cm) and the lowest was in Balam House (9.21 cm). Number of leaves, considered as an important parameter for fruit yield, was the maximum in two genotypes *i.e*., Cheonlima (25) and SQ-001 (25). Additionally, the maximum number of nodes per plant was recorded in First Runner (14.3) and the minimum was recorded from Balam House (11.53). Different types of leaves, flowers and fruits were observed in studied squash genotypes those are presented in Figure 1. Therefore, it was observed that, the squash genotypes showed a wide range of variation in their growth-related morphological traits. Variation in morphological (Ozturk *et al.*, 2021) as well as anatomical features (Balkaya *et al.*, 2010) is a common phenomenon among different *Cucurbita* species. Additionally, Esho and Jasim (2020) found a wide range of variability for number of nodes for the first female flower in Squash.

Moreover, considering the reproductive (flowering) characters, the results showed a significant variation on days to flower bud initiation, days to first male and female flowering, number of male and female flowers and viable pollen rate for all the squash genotypes (Table 3). The genotype Runner took minimum days to first flower bud initiation (19.29 days) while the genotypes First Runner took the lowest day to first male flowering (30.54 days) and the genotype Runner was the earliest genotypes to first female flowering (35.73 days). In most of the genotypes, female flowers were emerging before male flowers with some exceptions (Table 3). Significant difference for days to female flowering was also reported by Nahar *et al.*, (2016) in sweet gourd genotypes. In addition, the male flower numbers outnumbered the female flower numbers during the experimental period for all the genotypes. Both the male and female flowers formed simultaneously right from the outset. Additionally, pollen viability was of special interest to see the degree of influence it exerts upon fruit and seed setting. The percentage of pollen viability helps us in selecting the parents for crossing in a hybridization program. The mean values of fertile (viable) pollen showed statistically almost similar results for all the 15 genotypes (Table 3). This result indicated a high possibility of cross pollination among the genotypes and this could lead a high level to of genetic variability in squash genotypes.

Some other yield contributing traits considering the fruiting characters *i.e*., days to first harvest, number of nodes at first harvest, length and diameter of fruit per plant, fruit weight, number of fruits per plant, fruit yield per plant and total yield showed a significant variation among the genotypes (Table 4). The days to first harvest ranged from 54 to 62.67 days in Runner and Balam House respectively with a mean value of 58.3 days. Low variation was observed among the genotypes with respect to number of nodes at first fruit



First Runner

Alaska

Blossom House

Balam House

Cheonlima

Hungnong Squash

Runner

SQ-001

SQ-002

SQ-003

SQ-004

SQ-005

SQ-006

SQ-007

SQ-008

Fig. 1. Variation in leaves, flowers and fruits of fifteen squash genotypes

harvest (range: Runner 5.03 to Blossom House 6.5; mean: 5.78). The maximum fruit length was seen in the genotypes First Runner (45.58 cm), Hungnong Squash (45.42 cm) and Cheonlima (44.1 cm) while the minimum length of fruit was observed from SQ-007 (27.62 cm). Similar findings of significant variation on days to first harvest, number of nodes at first harvest and fruit length were also reported by Esho and Jasim (2020) in squash, Mohsin *et al.* (2017) in pumpkin and Nahar *et al.* (2016) in sweet gourd. Significant variation in fruit diameter (range: Alaska 17.47 cm to SQ-003 38.11 cm) was found among squash genotypes. Significant variation was also observed by Balkaya *et al.* (2010) in winter squash from the black sea region of Turkey. The individual fruit of First Runner (1165.5 g) had the highest weight followed by SQ-001 (1013.03 g) and Hungnong Squash (1002.08 g). The lowest single fruit weight was recorded in Runner (742.24 g). The variation of fruit weight could be due to the genetical, physiological and environmental influence. Abdein *et al.* (2021) reported similar results in respect of single fruit weight in summer squash. Number of fruits per plant was the maximum in case of Runner (10.2) proceeded to First Runner (10) and SQ-001 (9.53). Accession Balam House produced the minimum number (6.2) of fruits per plant. Rana *et al.* (2016) also observed significant variation in number of fruits per plant among cucumber genotypes. The yield of fruits per plant eventually contributes the total yield of fruit for each genotype. Among the studied squash genotypes, total fruit yield was varied significantly. The maximum total yield of fruit was obtained in First Runner (89.43 t/ha) preceded to SQ-001 (74 t/ha) and Cheonlima (63.48 t/ha) which was statistically different from other accessions, whereas the minimum total yield of fruit was obtained in case of Balam House (35.78 t/ha). These results corroborated with the findings of Akhter *et al.* (2018) in squash and Abdein *et al.* (2017) in sweet gourd. Uddain *et al.* (2019) also observed significant variation among the different genotypes of Zucchini squash in respect of weight of fruits per plant.

Table 2. Mean performance of squash genotypes for vegetative characters at first harvest

Genotypes	Plant height (cm)	Stem diameter (cm)	Number of leaves	Number of nodes
G1=First Runner	34.13^{bcde}	13^{a}	24.6^{ab}	14.3^{a}
G2=Alaska	33.33^{def}	12.13^{b}	22^{cde}	12.93^{bcd}
G3=Blossom House	33.68^{cdef}	11.45^{cde}	22.2^{cde}	11.73^{ef}
G4=Balam House	32.17^{f}	9.21^{j}	20.27^{f}	11.53^{ef}
G5=Cheonlima	33.61^{cdef}	11.6^{bcd}	25^{a}	14.27^{a}
G6=Hungnong Squash	32.5^{1ef}	11.63^{bcd}	23.27^{bcd}	12.07^{def}
G7=Runner	34.69^{abcd}	11.89^{bc}	23.87^{ab}	13.53^{abc}
G8=SQ-001	35.11^{abcd}	10.9^{efg}	25^{a}	14^{ab}
G9=SQ-002	36.05^{a}	11.29^{cde}	21.2^{ef}	13.93^{ab}
G10=SQ-003	34.31^{abcde}	11.36^{cde}	21.67^{ef}	11.87^{def}
G11=SQ-004	35.92^{ab}	10.03^{hi}	23.47^{abc}	11.47^{f}
G12=SQ-005	34.59^{abcd}	11.05^{def}	21.47^{ef}	11.87^{def}
G13=SQ-006	35.23^{abc}	10.48^{fgh}	21.8^{def}	12.6^{cde}
G14=SQ-007	34.03^{cde}	9.75^{ij}	21.07^{ef}	13.6^{abc}
G15=SQ-008	34.61^{abcd}	10.38j^{hi}	21^{ef}	12.93^{bcd}
Mean	34.27	11.08	22.52	12.84
SD	0.99	0.32	0.75	0.54
LSD	1.81	0.66	1.57	1.09

Means followed by the same letter (s) in a column do not differ significantly

Table 3. Mean performance of squash genotypes for various flowering characters

Genotypes	Days to flower bud initiation	Days to first male flowering	Days to first femal	Number of male flowers	Number of female flowers	Viable pollen (%)
G1=First Runner	20.86^{def}	30.53^{f}	35.80^{h}	18.73^{defg}	13.13^{b}	91.1^{abc}
G2=Alaska	22.20^{bcd}	32.47^{ef}	37.73^{g}	18.4^{fg}	12.87^{bc}	89.75^{bc}
G3=Blossom House	23.69^{ab}	31.73^{ef}	40.20^{cde}	18.53^{efg}	10.67^{ef}	87.13^{d}
G4=Balam House	24.58^{a}	33.87^{e}	41.80^{b}	18.93^{defg}	10.67^{ef}	84.47^{e}
G5=Cheonlima	22.70^{bc}	32.26^{ef}	40.33^{cde}	19.33^{bcdef}	12.8^{bc}	89.89^{abc}
G6=Hungnong Squash	22.89^{abc}	33.13^{e}	41^{bcd}	20.6^{a}	10.6^{ef}	89.11^{cd}
G7=Runner	19.29^{f}	37.73^{cd}	35.73^{h}	20.13^{ab}	14.13^{a}	90.28^{abc}
G8=SQ-001	21.49^{cde}	36.53^{d}	39.93^{def}	18.40^{fg}	12.93^{bc}	90.42^{abc}
G9=SQ-002	20.05^{ef}	42.87^{a}	43.06^{a}	20.0^{6abc}	12^{cd}	89.88^{abc}
G10=SQ-003	22.67^{bc}	43.47^{a}	36^{h}	19.73^{abcd}	11^{e}	92.36^{a}
G11=SQ-004	22.08^{bcd}	42.33^{a}	41.2^{bc}	19.6^{abcd}	12.8^{bc}	89.99^{abc}
G12=SQ-005	21.99^{bcd}	42.40^{a}	39.06^{f}	19.06^{cdef}	12.53^{bc}	91.02^{abc}
G13=SQ-006	22.59^{bc}	39.27^{c}	41.27^{bc}	19.46^{bcde}	12.8^{bc}	91.79^{ab}
G14=SQ-007	21.67^{cde}	39.53^{bc}	36.27^{h}	18.73^{defg}	9.87^{f}	89.91^{abc}
G15=SQ-008	22.48^{bcd}	41.80^{ab}	39.53^{f}	17.93^{g}	11.27^{de}	89.86^{abc}
Mean	22.08	37.33	39.26	19.18	12	89.80
SD	0.92	0.52	0.53	0.53	0.48	1.28
LSD	1.71	2.33	1.07	1.06	0.99	2.54

Means followed by the same letter (s) in a column do not differ significantly

Table 4. Mean performance of squash genotypes for various fruit characters

Genotypes	Days to first harvest	Nodes at first fruit harvest	Fruit length (cm)	Fruit diameter (cm)	Fruit weight (g)	No of fruits per plant	Fruit yield per plant (Kg)	Total yield (t/ha)
First Runner	58.47^{cde}	5.53^{cde}	45.58^{a}	19.67^{ef}	1165.50^{a}	10.2^{a}	11.92^{a}	89.43^{a}
Alaska	58.33^{cde}	5.58^{bcd}	34.43^{b}	17.47^{f}	797.60^{gh}	8.90^{de}	7.14^{efg}	53.53^{ef}
Blossom House	61.80^{a}	6.50^{a}	31.30^{de}	20.06^{ef}	816.70^{efg}	7.00^{h}	5.73^{h}	42.98^{h}
Balam House	62.67^{a}	6.34^{ab}	31.10^{de}	19.39^{ef}	810.10^{fgh}	6.20^{i}	4.77^{i}	35.78^{i}
Cheonlima	58.87^{cd}	5.77^{abc}	44.10^{a}	19.72^{ef}	948.90^{bc}	8.90^{def}	8.46^{c}	63.48^{c}
Hungnong Squash	59.8^{bc}	5.90^{abcd}	45.42^{a}	19.19^{ef}	1002.80^{b}	7.93^{g}	7.99^{cde}	59.98^{cd}
Runner	54.00^{h}	5.03^{e}	27.62^{f}	21.19^{e}	742.24^{h}	10.00^{ab}	7.22^{efg}	54.13^{ef}
SQ-001	55.80^{g}	5.19^{de}	32.2^{cd}	25.48^{cd}	1013.00^{b}	9.50^{bc}	9.95^{b}	74.00^{b}
SQ-002	61.07^{ab}	$5.8a^{bcd}$	34.4^{b}	21.02^{e}	909.30^{cd}	9.10^{cd}	8.31^{cd}	62.33^{cd}
SQ-003	55.93^{fg}	6.06^{abc}	11.84^{g}	38.11^{a}	921.10^{bcd}	8.13^{g}	7.47^{def}	56.05^{de}
SQ-004	57.73^{de}	6.28^{abc}	34.48^{b}	24.59^{d}	896.90^{cde}	8.30^{fg}	7.47^{def}	56.05^{de}
SQ-005	56.87^{efg}	6.16^{abc}	30.06^{e}	20.99^{e}	822.30^{efgh}	8.27^{g}	6.80^{fg}	51.0^{fg}
SQ-006	60.00^{bc}	5.92^{abc}	34.10^{bc}	18.85^{ef}	863.90^{defg}	8.93^{d}	7.90^{cde}	58.88^{cd}
SQ-007	55.60^{gh}	5.06^{e}	11.43^{g}	23.77^{b}	873.50^{defg}	7.00^{h}	6.34^{gh}	47.55^{gh}
SQ-008	57.53^{def}	5.55^{bcd}	31.7^{de}	27.42^{c}	987.18^{bc}	8.50^{ef}	8.21^{cd}	61.55^{cd}
Mean	58.29	5.78	31.97	22.93	904.74	8.46	7.71	57.78
SD	0.84	0.43	0.97	1.18	49.66	0.28	0.55	4.18
LSD	1.69	0.79	1.92	2.65	93.10	0.55	0.92	6.88

Means followed by the same letter (s) in a column do not differ significantly

Variability of yield contributing characters

The identification and utilization of an extensive germplasm is the prerequisite for improvement of a specific crop by adapting an appropriate plant breeding program. Regarding these, precise and exhaustive descriptions of the genotypes with the patterns of their genetic diversity can promote the introgression of current squash genetic base. In variability studies, high value of coefficient of variation (%CV) was found in number of nodes per plant at first harvest (5.11%), fruit yield per plant (7.13%), fruit diameter (6.89%), and fruit weight (6.14%). On the other hand, the lowest CV value was recorded in days to first female flowering (1.63%). The estimated genotypic variance (σ^2g) was higher than their corresponding environmental variances (σ^2e) for all the traits, except for plant height and number of nodes at first harvest that was very negligible (Table 5). Among the 15 accessions, the high magnitude of genotypic coefficient of variation (GCV) along with phenotypic coefficient of variation (PCV) were recorded for fruit diameter followed by the fruit yield per plant, total yield/ha and number of female flowers per plant. Very low level of GCV along with PCV was found in case of viable pollen percentage along with plant height at first harvest. Most of the characters had low GCV values than PCV values indicated considerable influence of environment in the expression of all the traits (Table 6). High GCV indicates the presence of exploitable genetic variability for the traits, which can facilitate selection (Muralidhara and Narasegowda, 2014; Yadav *et al.*, 2009).

Heritability estimation gives an insight into the extent of genetic control to express a particular trait and phenotypic reliability in predicting its breeding value (Ndukauba *et al.*, 2015, Nahar *et al.*, 2016). The heritability in combination with genetic advance (GA)

Table 5. Estimates of genetic parameters for various characteristics in squash genotypes

Parameters	Mean	MSS	CV %	σ^2_g	σ^2_{ph}	σ^2_e
Plant height (cm) at first harvest	34.27	3.67**	3.16	0.83	2.01	1.17
Stem diameter (cm) at first harvest	11.08	2.90***	3.55	0.91	1.07	0.16
Number of leaves at first harvest	22.52	7.26***	4.17	2.13	3.01	0.88
Number of nodes at first harvest	12.84	3.19***	5.11	0.92	1.35	0.43
Days to flower bud initiation	22.08	5.26***	4.64	1.40	2.45	1.05
Days to first male flowering	37.33	65.33***	3.73	21.13	23.07	1.94
Days to first female flowering	39.26	17.29***	1.63	5.63	6.04	0.41
Number of male flowers	19.18	1.71***	3.29	0.44	0.84	0.40
Number of female aflowers	12.00	4.56***	4.95	1.40	1.76	0.35
Viable pollen (%)	89.80	10.80***	1.69	2.83	5.14	2.31
Days to first harvest	58.30	18.23***	1.74	5.73	6.76	1.03
Nodes at first fruit harvest	5.78	0.63*	8.24	0.14	0.36	0.23
Fruit length (cm)	31.97	96.72***	3.59	31.8	33.12	1.32
Fruit diameter (cm)	22.93	92.99***	6.89	30.17	32.67	2.50
Fruit weight (g)	904.74	34814***	6.14	10573	13668	3095
Number of fruits per plant	8.46	3.74***	3.87	1.21	1.32	0.11
Fruit yield per plant (Kg)	7.71	8.57***	7.13	2.76	3.06	0.30
Total yield (t/ha)	57.78	477.72***	7.12	153.6	170.5	16.93

* Significant at 5% level of probability; ** Significant at 1% level of probability and; *** Significant at 0.1% level of probability.

increases the intensity of selection in a breeding program. High heritability indicates less environmental influence in the observed variation (Abdein *et al.,* 2017). Thus, genetic advance measures the difference between the mean genotypic values of the original population from which these are selected. Almost all the attributes showed high heritability except nodes at first harvest (38.89%) and plant height (41.49%). The highest estimates of genetic advance (in percent of mean) were determined for total yield/ha, fruit yield per plant, fruit weight, fruit length, fruit diameter and number of fruits per plant (Table 6).

Correlation analysis (Table S1), the trait plant height had significant positive correlation with days to first male flowering, number of female flowers, number of fruits per plant, and fruits yield per plant. Other attributes such as number of leaves at first harvest was negatively and significantly correlated with the days to first male flowering. The number of nodes at first harvest showed positive and significant correlation with number of female flowers, single fruit weight, number of fruits per plant and yield of fruits ton per hectare. The number of female flowers per plant had significant and positive correlation with number of

Table 6. Estimation of heritability and genetic advance (GA) in squash genotypes

Parameters	GCV	PCV	ECV	Herita-bility	GA (5%)	GA (% mean)
Plant height (cm) at first harvest	2.67	4.14	1.47	41.49	1.21	3.53
Stem diameter (cm) at first harvest	8.61	9.34	0.73	85.05	1.81	16.34
Number of leaves at first harvest	6.48	7.71	1.22	70.76	2.53	11.23
Number of nodes at first harvest	7.47	9.05	1.58	68.15	1.63	12.70
Days to flower bud initiation	5.36	7.10	1.73	57.14	1.84	8.33
Days to first male flowering	12.31	12.87	0.56	91.58	9.07	24.28
Days to first female flowering	6.04	6.26	0.22	93.21	4.72	12.02
Number of male flowers	3.46	4.78	1.32	52.38	0.99	5.16
Number of female flowers	9.86	11.04	1.18	79.73	2.18	18.14
Viable pollen (%)	1.87	2.53	0.65	55.05	2.57	2.86
Days to first harvest	4.11	4.46	0.35	84.76	4.54	7.79
Nodes at first fruit harvest	6.47	10.38	3.91	38.89	0.48	8.31
Fruit length (cm)	17.64	18.00	0.36	96.01	11.38	35.61
Fruit diameter (cm)	23.95	24.93	0.98	92.35	10.87	47.42
Fruit weight (g)	11.37	12.92	1.55	77.36	186.31	20.59
Number of fruits per plant	13.00	13.58	0.58	91.67	2.17	25.64
Fruit yield per plant (Kg)	21.55	22.69	1.14	90.2	3.25	42.16
Total yield (t/ha)	21.45	22.60	1.15	90.07	24.23	41.93

fruits per plant. Fruit length had significant and positive correlation with fruit weight, number of fruits per plant and fruit yield per plant and also significantly and negatively correlated with fruit diameter. One of the most important traits of fruit weight was significantly and positively correlated with fruit yield per plant. Highly significant and positive association of fruit yield per plant was recorded with the plant height, number of leaves per plant, number of nodes at first harvest, fruit length, fruit weight and number of fruits per plant. Similar findings were noticed by Gomes *et al.* (2020) in Brazilian germplasm of winter squash and Mohsin *et al.* (2017) in pumpkin.

In cluster analysis (CA), the cluster means of 15 accessions of squash showed that the mean values of the clusters varied in magnitude for maximum characters (Table S2. The cluster II showed the highest total yield value along with the second highest fruit length and fruit diameter value, the highest number of fruits per plant value and the highest yield per plant value, which could contribute to total yield. From the clustering comparison of the means, it was found that cluster II expressed the best agronomic quantitative yield contributing traits and yield potentials. Comparing the means of all clusters it was showed that First Runner from cluster I, Cheonlima and SQ-001 from cluster II, SQ-008 from cluster IV and SQ-006 from cluster III expressed the best quantitative and qualitative traits and yield potentials which could be effective for the improvement of yield of squash (Fig. S1). Gomes *et al.* (2020) reported similar results in Brazilian germplasm of winter squash. Ene *et al.* (2016) also reported similar findings in cucumber genotypes. This suggests that the genotypes of squash of the same origin have diverse and broad genetic basis.

Principal component analysis (PCA) is an important multivariate technique used to examine associations between characters and measures the genetic variability of genotypes (Balkaya *et al.*, 2010; Ene *et al.*, 2016). The three principal components (PC1, PC2 and PC3) can be retained to describe the variability among the squash genotypes (Table S3). The first three components explain 63% of the total genetic variation while the first two principal components accounted for 53% and first component accounted for 32.4% of the total genetic variation among the 18 attributes describing 15 different genotypes. The first component (PC1) described 32.4% of the total variation, second component (PC2) explained 20.6% of the total variability and the third component (PC3) evaluated only 10.02% of the total variation. The PC1 was positively and strongly associated with the plant height (0.18), stem diameter (0.29), number of leaves (0.29), number of nodes (0.28), number of female flower (0.30), viable pollen percentage (0.25), fruit length (0.13), fruit weight (0.21), number of fruits per plant (0.39) and fruit yield per plant (0.36). The PC2 was positively and highly associated to days to first harvest (0.36) and fruit length (0.47). In case of PC3, it was strongly associated with plant height (0.47), days to first male flowering (0.39), days to first female flowering (0.49), number of male flower (0.31), number of female flowers (0.24) and fruit length (0.16). Whereas, morphological (qualitative) characterization showed that limited variability present in the genotypes in respect of some characters viz., plant vigor, stem and leaf pubescence, and flower colour. Significant variability was observed in case of fruit shape, fruit size, fruit skin color and luster respectively. This finding corroborated with the findings of El-Hadi *et al.* (2014) in squash and partly agrees with the results by Khawla *et al.* (2019) in Tunisian squash and Nahar *et al.* (2016) in sweet gourd.

Qualitative characterization

Selection of qualitative traits is also very important in successful crop breeding program. Significant variation was found under this research in relation to different qualitative traits of squash accessions. Most significant variation was found in fruit size, fruit shape and fruit skin color followed by lustre (Table S4). The fruit colour, size, shape was morphologically different because of the genetic makeup present in the studied genotypes (Fig. 1). A similar morphological variation of qualitative traits was reported by Uddain *et al.* (2019) in Zucchini squash, Muralidhara and Narasegowda (2014) in pumpkin, Nahar *et al.* (2016) in sweet gourd and Ene *et al*. (2016) in cucumber.

CONCLUSION

High heritability coupled with high genetic advance observed for total yield/ha, fruit yield per plant, fruit weight, fruit length and fruit diameter in this set of germplasm indicated that, these traits will be the main contributing factors for further crop improvement programme. Significant and positive association of fruit yield per plant with number of leaves per plant, number of nodes at first harvest, fruit length, fruit weight and number of fruits per plant suggested that, these traits were inter-related and collectively contributed to the final yield of squash. The principal component analysis showed that number of fruits per plant, fruit yield per plant, fruit length and days to first female flowering were the most discriminating factors that accounted for the genetic diversity of squash and would be considered for squash improvement program. Considering yield performance, it can be recommended that First Runner is the highest yielding genotype in similar environment which might be used as a parent in developing high yielding variety of squash. The genotype Runner can be used as a parent for developing early fruiting variety whereas Alaska would be preferable for improving the appearance of fruits.

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