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Morphological Diversity in Indigenous Cucumber Genotypes of Bangladesh

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Abstract- A study was conducted to assess the genetic diversity of locally collected cucumber germplasm by using morphological markers at the Plant Genetic Resources Centre of the Bangladesh Agricultural Research Institute, Gazipur, Bangladesh during March to August 2017. Quantification of variability for each character was done by using Shannon Weaver Diversity Index (SWDI). Characterization data were further subjected to Principal Component Analysis (PCA). The collection exhibited medium to high variations for both the qualitative and quantitative characters as reflected by mean diversity indices 0.62 and 0.63 respectively. Two phenological characters viz. days to harvest and days to pistillate flower respectively showed the weakest indices 0.26 and 0.32. First four principal components accounted for 80% of the observed variations. Analysis of the factor loading of component characters showed that fruit characters to be the major attributes of variation. Cluster analysis performed on the germplasm gave 3 distinct clusters. Genotypes AMA-406 and Ac-207 in cluster I found to be potential for breeding purpose.

Keywords: genetic diversity, PCA, Shannon diversity, phenology, variability.

I. INTRODUCTION

Cucumber (*Cucumis sativus* L.), a traditional vegetable of Bangladesh is considered a kitchen garden crop. Increasing market demands, less investment for cultivation and favorable marketing facilities has recently promoted cucumber as commercial crop. Cross pollinating nature and the growing environments has generated variability in cucumber at community levels (Smith and Smith, 1989). Being the products of on-farm selection by the farmers and co-adaption to their own habitats-the land races of cucumber represent a tremendous genetic resource needed for future crop improvement. Like many other cultivated indigenous varieties and wild relatives of crops, genetic erosion of cucumber is occurring in Bangladesh due to introduction of commercial varieties and infestation of insect-pests-diseases (Frankel, 1972; Harlan, 1975). In support of future breeding activities, PGRC, BARI is collecting and maintaining cucumber germplasm of different agro-ecological zones with

different agricultural practices. However, in the absence of requisite information on indigenous landraces of cucumber in Bangladesh, the present study was initiated to characterize and evaluate 33 locally collected cucumber accessions.

II. MATERIALS AND METHODS

The experiment was conducted at the Plant Genetic Resources Centre (PGRC), Bangladesh Agricultural Research Institute (BARI), Gazipur, during March to August, 2017. A total of 33 germplasm randomly selected for the study. Collection sites including the name of nearby district are presented in Table 1 and Figure 1. One accessions represented one treatment with two plants in a plot of 3m x 3m size and maintaining 0.5 m drains between the plots. Seeds were sown in polybags on 2 March, 2017. Around 35 day old seedlings were transplanted in field plots on 10 April, 2017. Appropriate cultural management practices were provided throughout the growth period of plants. Fruits were harvested when attained physiological maturity. Data on qualitative and quantitative characters were collected following standard descriptors list (Díez, et. al., 2008).

a) Data analysis

Descriptive statistics of data for each attributes were determined to compare the relative amount of variability of the traits.

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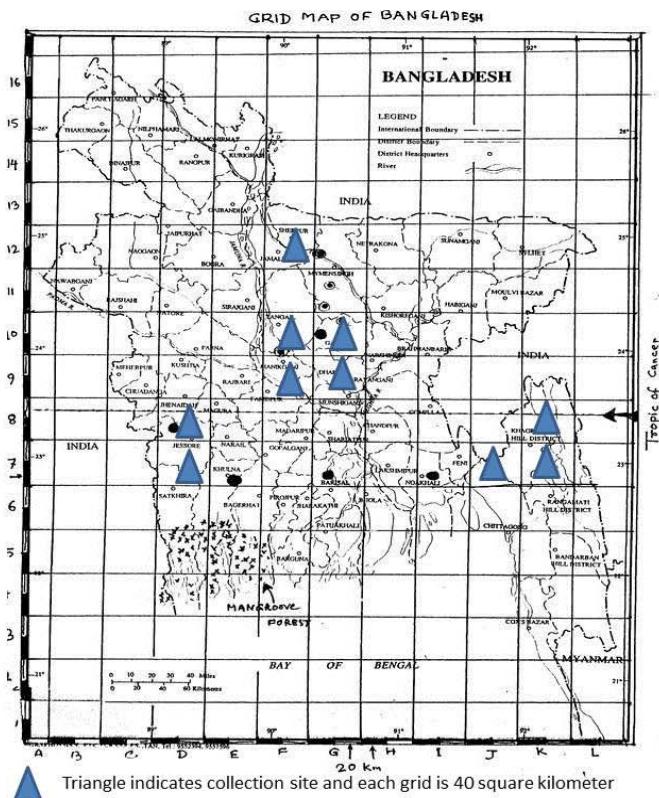
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Table 1: Thirty three cucumber germplasm collected from four regions of Bangladesh

District	Location on Grid Map	Germplasm	Number of entries
Dhaka	G9	AC-304, AH-54, AH-59, AH-60, AH-61	5
Gazipur	G10	BD-4241, BD-4260, BD-4321, IAH-117, IAH-126, AC-356, AH-63, AH-66,	8
Jashore	D7-8	AHI-35, AHI-70, AHI-83, AHI-94	4
Khagrachari	J7-K7-8	IAH-273, IAH-274, IAH-275, IAH-297, IAH-299, IAH-323, IAH-327, IAH-331	8
Manikgonj	F9	AC-207, AC-245	2
Sherpur	F12	AMA-406	1
Tangail	F10	AC-74, AC-100	2
Breeding Line	-	BD-9764, BD-10104, BD-10954	3
		Total=	33

**Figure 1:** Grid map showing the collection sites of germplasm

b) Shannon-Weaver Index of Diversity

Phenotypic diversity for both qualitative and quantitative traits was determined by using Shannon-Weaver Diversity Index (H'). H' ranges from 0 to 1, where 1 indicates the maximum diversity (Yu Li et al., 1996). H' is defined as:

$$H' = -\sum P_i \log_2 P_i \text{ where}$$

P_i is the proportion of the total number of genotypes belonging to the i^{th} class. The exact

descriptor states define the classes for the qualitative characters while the classes for quantitative characters are defined according to the procedure suggested by Yu Li et al, (1996) where each H' value was standardized.

For the quantitative characters, the overall genotypes mean (\bar{X}) and standard deviation (σ) was used to subdivide the population values (x_i) into 10 frequency classes ranging from class 1 (if $x_i \leq -2\sigma$) to

class 10 (if $x_i \leq \bar{X} + 2\sigma$), the class interval being 0.5σ . The relative frequencies for the different classes were used to calculate the diversity index. The H' for each character was calculated using MS Excel. Test statistic $H_0: H'=0$ H' classifies as high when $\geq H'0.66$, moderate when $H' = 0.34- 0.66$ and low variation when $H' < 0.33$ (Sourour and Hajar, 2009).

c) Multivariate Analysis

Multivariate analysis methods, including principal component analysis (PCA), clustering and discriminant analysis were carried out to assess the pattern of morphological variation and to group the population into classes. Differences among classes were revealed based on a set of quantitative variables. Dissimilarity of the population was determined by using Euclidian Distance.

d) Principal Component Analysis (PCA)

PCA estimates the structure of the correlation between the variables within a data set. It can identify the main variable(s) that significantly contribute to the variation within the data set. Average distances and dendograms were computed and constructed using SPSS package. Cluster analysis based on similarity matrices using the Unweighted Pair Group Method Arithmetic Average (UPGMA) was done.

III. RESULTS AND DISCUSSION

The accessions under study were uniform in respect of growth habit, presence of pubescence on stem, leaf size and presence of tendrils. Diversities were mostly noticed in fruit characters.

a) Descriptive Statistics

The coefficient of variation (%CV) compares the relative amount of variability between traits (Sharma, 1988). The highest %CV was estimated in fruit length and fruit weight which were followed by %CVs of number of fruit per plant and internode length (Table 2). These results imply that fruit length, fruit weight, number of fruits, internode length of vine had higher amount of exploitable genetic variability among the studied cucumber characters. It also means that there is greater potential for favorable advance in selecting these attributes compared to others (Eid, 2009; Ndukauba et al., 2015). Ene et al., (2016), similarly reported high %CV in fruit characters of cucumber genotypes. Conversely, the lowest %CV recorded for the attributes days to harvest, days to staminate flower had low exploitable variability and as a result had less potential for favorable advance in selecting when compared to others.

b) Phenotypic Diversity

Shannon Weaver Diversity Indices (H') was estimated to assess the diversity in the morphological and phenological attributes. The lowest and highest

diversity indices for the qualitative characters ranged from 0.19 (blossom-end fruit shape) to 0.84 (leaf intensity of green color) with an average 0.67 (Table 3). The mean diversity indicates existing of high variation within the collection in terms of qualitative characters (Sourour and Hajar; 2009).

In quantitative attributes the estimated H' ranged from 0.26 (days to harvest) to 0.87 (fruit length) with an average value 0.63. The diversity values of two phenological attributes were 0.32 (days to pistillate flower) and 0.50 (days to staminate flower); the other attributes showed higher diversity. However, medium degree of variation existing for quantitative attributes within the collection as reflected by the mean diversity value of 0.63 (Table 4).

Shannon diversity index groups the genotypes into 1 to 10 frequency classes based on overall mean and standard deviation for a particular trait. Germplasm included in higher cluster for a particular trait obviously superior to those included in lower clusters. In the present study, genotypes AC-207 and AMA-406 were found to be included in cluster X (10) in respect of fruit weight, fruit length and fruit breadth (Table 6). However, the genotypes were inferior in respect of bearing habit. This means the accessions are potential breeding materials and may also be used as variety if improvement occurs in bearing habit.

c) Principal Component Analysis

PCA explains the variability of a set of random variables in terms of new set of variables with reduced dimensionality and with little loss of information as possible. The principal component analysis resulted in reduction of 10 original variables to four independent linear combinations, principal component of variables which accounted for 80% of the observed variation (Table 6). The rest of the components accounts for 20% of the variation. The results are indication of maximum linearity of the data which means the presence of high variability in the germplasm under study.

Factor loading analysis showed that Principal Component-1 (Prn1) comprised four characters, namely leaf length, leaf width, fruit length, fruit width and fruit weight while Prn2 included leaf length and number of fruit per plant. Highest loading 85% was found in fruit width and fruit weight in Prn1, days to number of fruit per plant in Prn2 and days to female flower in Prn3 contributed highest loading 71% (Table 7).

d) Cluster Analysis

Seven variables of 33 genotypes were selected on the basis of principal component of variation subjected to Unweighted Paired Group Method Arithmetic Average (UPGMA) cluster analysis using SPSS -15. From the resulting dendrogram in 1-5 scale measurement, the population was divided into 3 clusters (Figure 1). The largest cluster was cluster II, which included 20 accessions and second largest was cluster

III, which included 11 accessions. Cluster I, included 2 accession (Table 8).

The inter-cluster values were maximum in between clusters I and III (383.239) followed by clusters I and II (226.618) and minimum distance was in between II and III (118.654). The intra-cluster distance was maximum in cluster III (35.89) and minimum in cluster I (21.37) indicating homogeneity of accession in cluster I. Length of fruit, fruit width and fruit weight in clusters I were found to contribute maximum diversities (Table 9). In cucumber fruit is the most economically important part, thus has undergone intensive selection by man. The significant differences observed among the genotypes for all the traits suggest the presence of variability among the genotypes. Afangideh and Uyoh (2007) also reported the presence of variation among cucumber genotypes collected from different agro-ecological zones.

IV. CONCLUSION

The results of the study indicating the existing of medium to high variation in both qualitative and quantitative characters. To enhance variation further in the collection, future collection trips should consider the characters which are components of lower PCs and those characters which are not identified to belong to the retained PCs. Study on geographical pattern of diversity should be done to capture maximum diversity existing in the country.

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Table 2: Mean and standard deviation for 33 land race genotypes for different characters

	Minimum	Maximum	Mean	Standard deviation	CV%
Internodes length (cm)	2.62	10.65	6.95	1.88	27.08
Leaf length (cm)	7.39	16.86	12.46	2.21	17.72
Leaf width (cm)	8.29	19.66	15.47	2.52	16.27
Days to staminate flower	36	40	37.62	2.24	5.95
Days to pistillat flower	43	48	46.65	3.47	7.44
Fruit length (cm)	10.4	20.92	11.52	4.12	35.72
Fruit width (cm)	4.8	7.86	5.16	0.95	18.33
No. of fruit/pl	4	6.5	5.27	1.69	31.93
Days to harvest	84	89	84.75	2.65	3.13
Fruit weight (g)	242.8	528	251.68	88.43	35.14

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Table 3: Stem and mature leaf colors of cucumbers under study

Descriptor	Observed Phenotype	H'
Stem color		
	Light green	0.79
	Dark green	
Leaf intensity of green color		0.84
	Light	
	Medium	
Stem end fruit shape		
	Obtuse	0.61
	Acute	
Fruit skin texture		
	Spiny	0.68
	Smooth	
Blossom end fruit shape		
	Flat	0.19
	Tapered	
Fruit shape		
	Oblong	
	Oval	0.74
	Ovate	
Fruit skin color at table maturity		
	Light green	
	Green	0.71
	Yellowish green	
	Blackish green	
	Whitish green	
Fruit skin color at maturity		0.79
	Brown	
	Yellow	
	Average=	0.67

SWDI (Shannon weaver diversity index)

Table 4: Quantitative descriptor of germplasm

Character	H'
Internodes length (cm)	0.71(H)
Leaf length (cm)	0.65(M)
Leaf width (cm)	0.73(H)
Days to staminate flower	0.50(M)
Days to pistillat flower	0.32(L)
Fruit length (cm)	0.87 (H)
Fruit width (cm)	0.75 (H)
No. of fruit/pl	0.76(H)
Days to harvest	0.26(L)
Fruit weight (g)	0.78 (H)
Average=	0.63

Table 5: Distribution of genotypes in 10 clusters in order of richness of traits of fruit

Cluster	Fruit weight (g)	Fruit Number	Fruit length (cm)	Fruit width (cm)
I	0	0	0	0
II	0	0	AH-63, AC-100	AC-254
III	AC-100, AH-59, AH-54, AH-63	IAH-327, AC-100, AH-61, AH-63, AHI-70, IAH-274, IAH-275, IAH232, AMA-406	AH-59, IAH-273, AH-54, IAH-275	AH-63, IAH-33
IV	IAH-117, AC-74, BD-9764, AHI-94, AC-245, BD-10954, AH-66, AHI-94, AH-54, AH-59, IAH-331	AH-66, AHI-94, AH-54, AH-59	AC-245, IAH-274, IAH-327, AC-74, IAH-323, AC-356	AC-100, AC-74, AHI-94, BD-4260, BD-9764, IAH-117, AC-356
V	IAH-297, BD-10104, AHI-83, BD-4260, AH-66, BD-4321, AC-356, AH-61, BD-4241	AC-74, IAH-126, IAH-273, IAH-297, IAH-299, BD-10104, AC-245, IAH-331	BD-10954, AH-66, BD-9764, IAH-299, AHI-83	BD-4321, AH-66, IAH-299, AHI-35, AHI-275, IAH-126, IAH-274, AHI-70, BD-4241, AHI-83, IAH-273
VI	IAH-274, AC-304, IAH-126, AH-60, AHI-35, BD-4321, BD-10954, IAH-117, AC-207	BD-4321, BD-10954, IAH-117, AC-207	IAH-297, IAH-126, BD-4321, AHI-94, IAH-117	BD-10954, BD-10104, IAH-297, AH-54, AH-59
VII	IAH-273, IAH-299	BD-4260	BD-10104, IAH-331, AHI-35, AC-304, AHI-70	AC-304, AH-61, IAH-323, IAH-327
VIII	AHI-70, IAH-323, IAH-327	AC-304, AC-356, AH-60, AHI-35, AHI-83	AH-60, BD-4260, AH-61, BD-4241	0
IX	0	BD-4241, BD-9764	0	0
X	AC-20, 7AMA-406	0	AC-207, AMA-406	AH-60, AC-207, AMA-406
(H')	78%	76%	87%	75%

Table 6: Eigen values of 10 principal components for quantitative characters of cucumber germplasm producing oblong fruits

Component	Initial Eigen values		
	Total	% of Variance	Cumulative %
1	2.921	29.212	29.212
2	2.298	22.980	52.192
3	1.655	16.554	68.746
4	1.133	11.333	80.079
5	.723	7.232	87.312
6	.481	4.813	92.125
7	.368	3.675	95.800
8	.259	2.591	98.391
9	.108	1.076	99.467
10	.053	.533	100.000

Table 7: Initial cluster centre

Traits	Component			
	1	2	3	4
Internodes length (cm)	-.275	.207	-.118	.847
Leaf length (cm)	.636	.615	.310	.133
Leaf width (cm)	.652	.598	.218	.189
Days to male flower	.311	-.739	.409	.322
Days to female flower	.315	-.569	.711	.177
Fruit length (cm)	.747	.098	-.545	.039
Fruit width (cm)	.859	-.116	-.344	.136
No. of fruit/plant	.137	.600	.564	-.097
Days to harvest	.672	-.117	.241	-.436
Fruit weight (g)	.817	-.357	-.317	.036

Extraction Method: Principal Component Analysis.

Table 8: Number of genotypes in each cluster

Cluster	Number of genotype	Genotype
I	2	AC-207, AMA-406
II	20	BD-4241, BD-4260, BD-4321, BD-10104, AC-304, AC-356, AH-60, AH-61, AH-66, AHI-35, AHI-70, AHI-83, IAH-126, IAH-273, IAH-274, IAH-275, IAH-297, IAH-299, IAH-323, IAH-327
III	11	BD-9764, BD-10954, AC-74, AC-100, AC-245, AH-54, AH-59, AH-63, AHI-94, IAH-117, IAH-331

Table 9: Distance between final cluster Centres

Cluster	1	2	3
1	<u>21.37</u>	264.618	383.239
2		<u>41.141</u>	118.653
3			<u>35.895</u>

Table 10: Distribution of Germplasm in different cluster collected from different geographic allocation

District	Location on Map	Cluster-I		Cluster-II		Cluster-III	
		Accession/Collectors Number	Nos	Accession /Collectors Number	Nos	Accession /Collectors Number	Nos
Dhaka	G9	0	0	AC-304, AH-60, AH-61,	3	AH-54, AH-59	2
Gazipur	G10			BD-4241, BD-4260, BD-4321, AC-356, AH-66, IAH-126,	6	AH-63, IAH-117	2
Jashore	D7-8			AHI-35, AHI-70, AHI-83,	3	AHI-94	1
Khagrachari	J7-K7-8			IAH-273, IAH-274, IAH-275, IAH-297, IAH-299, IAH-323, IAH-327	7	IAH-331	1
Manikgonj	F9	AC-207	1	0	0	AC-245	1
Sherpur	F12	AMA-406	1	0	0		0
Tangail	F10			0	0	AC-74, AC-100	2
Breeding Line	-		2	BD-10104	1	BD-9764, BD-10954	2
					20		11

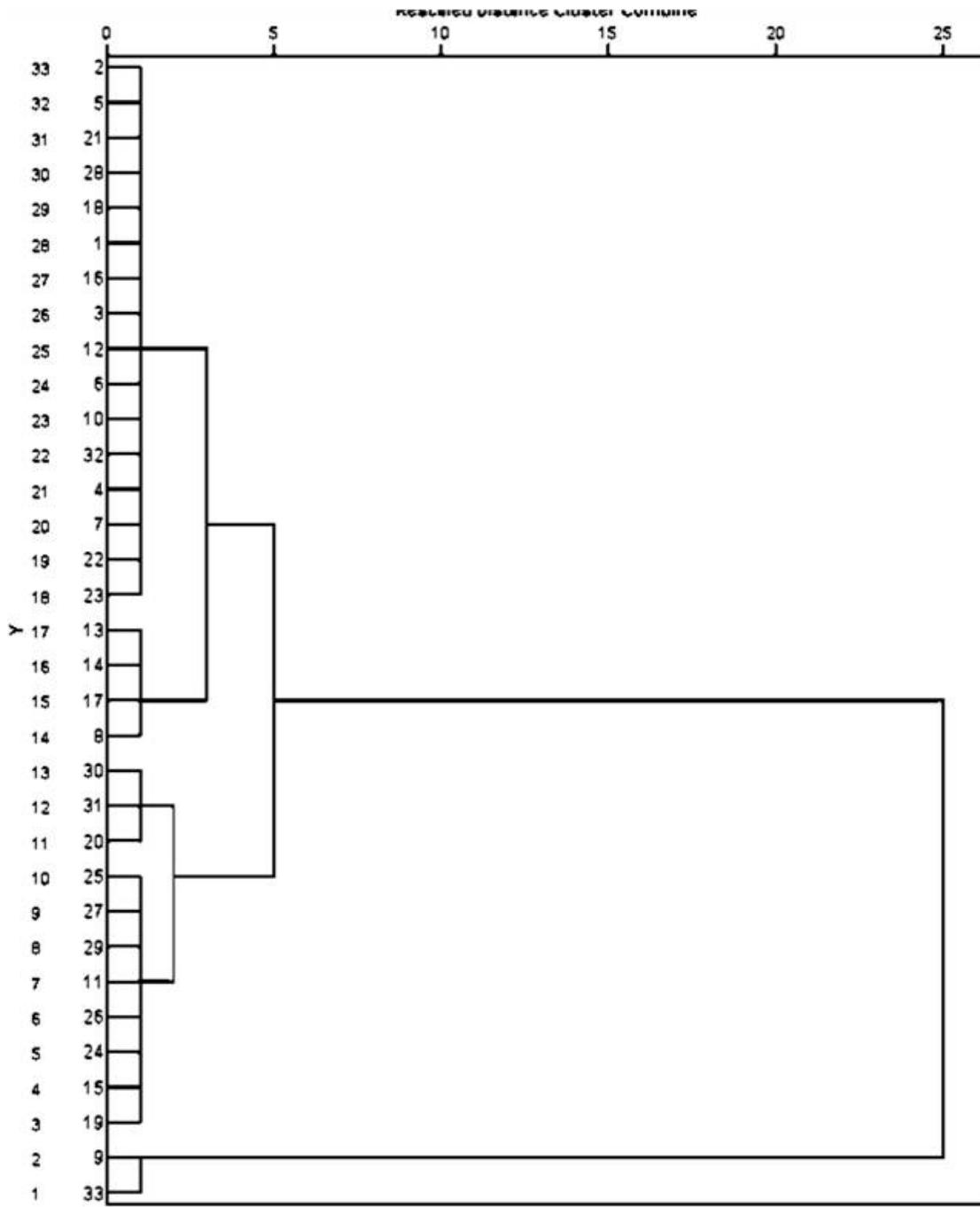


Figure 2: Dendrogram using average linkages of germplasm (between groups)

