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European Journal of Experimental Biology, 2012, 2 (5):1382-1388

ISSN: 2248 –9215
CODEN (USA): EJEBAU

Assessment of genetic variation in cucumber (*Cucumis sativus* L.) genotypes

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ABSTRACT

In this research, twenty genotypes of cucumber (Cucumis sativus) were studied that were collected from different geographical regions. The experiment was conducted as a randomized complete Block Design (RCBD) with four replications in Greenhouse. Data was collected on morphological features, yield and components of yield of cucumber which include: Total fruit yield per pickling, Fruit number per pickling, branch number per plant, plant height, distance between internode, length of branches, shoot diameter, leaf length, leaf width, fruit diameter, fruit length, plant vigor and fruit number per node. Analysis of variance showed that there was a high significant variation for all of the studied traits between genotypes. Mean comparison showed that the genotypes showed a broad phenotypic variation for studied traits, such that total fruit yield per pickling ranged from 474.3 g (Gohar) to 338.3 g (Tornado). Cluster analysis with Ward method divided the genotypes to four distinct groups. Genotypes in group number two (Gohar, Adrian451, Green majic, Sina) had the highest total fruit yield per pickling. For other traits, genotypes in groups number 1 and 2 (10 genotypes) showed best situation. Therefore, Selection of superior genotypes in view point of desirable morphologic traits, with high genetic distance could be selected for hybridization programs and recognition of best genotypes for different traits to produce new elite hybrids in cucumber.

Keywords: Analysis, Cluster, Cucumber, Genetic variation, Mean comparison.

INTRODUCTION

Assessment of genetic diversity could be suitable in crop breeding for diverse applications such as identifying diverse parental genotypes. Genetic diversity is the amount of heritable variability between varieties or populations of organisms [1]. Variability occurs from differences in DNA sequences, biochemical characteristics like protein structure or isoenzyme properties, physiological properties like growth rate, and morphological characters. Substantial effort has been directed towards collecting, preserving and evaluating genetic variability in crops [2].

The cucumber (*Cucumis sativus* L.) is a thermophiles and frost-susceptible horticultural crop usually cultivated in fields during spring-summer period [3] or in greenhouse in different seasons. Cucumber is believed to have been domesticated in India for 3000 years and in Eastern Iran and China probably for 2000 years [4]. Now it is grown all around the world [5, 6]. Cucumber is thought to be one of the oldest vegetable crops, being grown for at least five thousand years [7]. It is the fourth most important vegetable crop after tomato, cabbage and onion in Asia [8] and

Iran is in the third place of cucumber production in the world [9]. As a vegetable crop, *Cucumis sativus* has great economic importance [10]. In addition, cucumber is cultivated because its extract has soothing, cleansing and softening properties which are important for the cosmetics industry [11]. Genetic diversity in the primary center of origin (India) and secondary center for diversity (China) has been described [5]. Therefore, the study of phenotypic variation exhibited among cucumber genotypes could be suitable for plant breeding of cucumber

Breeding of yield in cucumber has been one of the important objectives of many cucumber breeding programs since 1900s [12, 13]. Biochemical markers such as isozyme [14, 15] have been used for evaluation of genetic diversity in cucumber. Molecular markers have extensively been used for assessment of genetic diversity in cucumber including restriction fragment length polymorphisms (RFLPs) [16] and PCR-based markers such as RAPD [17], inter-simple sequence repeat (ISSR) [18], simple sequence repeat (SSR) [19] and EST-SSR [20] have been employed for the characterization of genetic diversity in cucumber and mushroom [21], however it seems that evaluation of morphological traits is not used, abundantly, for estimation of genetic variation in it. Al-Rawahi *et al.* [22] studied the genetic diversity among twenty-four accessions of cucumber for morphological traits. Al-Rawahi *et al.* [22] clustered the genotypes to two main clusters. It seems that evaluation of genetic variation for morphological traits will help to provide valuable information about identification of new sources for high yield in cucumber germplasm in breeding programs. Also, observed results could suggest comparative advantages for further breeding options in cucumber.

The objective of this research was to perform a compare germplasm evaluation of some cucumber cultivars in Iran. Assessment of genetic diversity among genotype collections could exploit perfect information to enhance the development of better performing varieties of the cultivated species of cucumber.

MATERIALS AND METHODS

This experiment was conducted at Research Greenhouse of Agricultural Department of Islamic Azad University, Khorasgan (Isfahan) branch, in Iran, (51º36´ longitude and 32º63). Genotypes were planted in March 2011 for the spring season 2011, in pitmas, perlite, cocopit bed. The experiment was conducted as a Randomized complete Block Design (RCBD) with 4 replications. There were ten plants in every plot. In this study 20 genotypes from different geographical regions of world were evaluated that their name is presented in Table1. Studied traits were measured on 10 plants for each genotype in each replication and used from average of them in analysis.

Cultural practices:
The space between and within couple rows were 90 (cm) and 50 (cm), respectively, and 180 (cm) between every couple rows. Different fertilizers were used based on soil analysis, including: potassium nitrate, Ammonium nitrate, magnesium nitrate, Iron and the other mineral elements as sulphate solving in water. Irrigation was applied when needed and Dichlorvos, Trigard, Vertimec and Organic neem oil were applied for insect control. The greenhouse air temperature at the growing period was 25-30 C˚ day / 19-21 C˚ nights with a relative humidity of about 60%.

Different horticultural traits including: total fruit yield per pickling (TFY)(g), fruit number per pickling (FNP), branch number per plant (BP), plant height (PH) (cm), the length of branch (LB) (cm), shoot diameter (SHD) (cm), leaf length (LL) (cm), leaf width (LW) (cm), fruit diameter (FD) (cm), vigor of each plant (VP), fruit number per node (FNN), distance between internode in (DN) (cm) and fruit length (FL) (cm) were measured on 20 different genotypes of cucumber.

Each plot was harvested daily, if there was marketable fruit size in each plant. Then the number and the weight of total fruit in each plot were recorded. The length of the plant from the soil surface to the tip was measured after the final harvest. The length of nodes per vine on the main stem was measured from node number 10 to number 20. Number of branch per plant, length of branch and vigor of plant was rated on a 1 to 4 scale at the time of harvesting, based on the least to the most number and vigor in each plant. Length of 15 fruits harvested randomly in five days on three fruit in each day at edible maturity was recorded from base to the apex of fruit and averaged. Diameter of the same 15 fruits selected for recording the length, was measured in centimeter at maximum thickness with the help of vernier caliper. Length of leaf was measured from dom to tip and width of leaf was recorded at maximum point. The number of fruit per node was recorded based on more than 75% on nodes.

Statistical analysis
The collected data were subjected to analysis of variance (ANOVA) using general linear model (GLM) of Statistical Analysis System program [23]. Mean comparisons were conducted using the Fisher's least significant difference ($LSD_{0.05}$) test. The means of each trait were used for cluster analysis. Euclidean distance was used for cluster

analysis with the ward method by using SPSS software version 16. The number of group in dendrogram was recognized based on F-bill test (by SAS software) and discriminant function (by SPSS software).

RESULTS

The results of analysis of variance (ANOVA) showed a significant difference for all of the studied traits (Table 2). According to Table 3, the highest value for total fruit yield (TFY) was belonged to genotype of Gohar (474.25) (g). The means of TFY were ranged from 338.31 (g) (Tornado) to 474.24 (g) (Gohar). Fruit number per plant (FNP) ranged from 6.52 in (Green majic and Adrian-451) to 3.50 in Tornado and Karim.

Fruit number per node (FNN) was varied from 3.40 (Yalda-R2) to 1 (Karim). Although yalda-R2 showed the highest FNN, however TFY, PH, BP, LL, FL, SHD and LB were low in this genotype. Plant height (PH) had significant variation among 381.25 (cm) in Neda to 267.25 in Yalda R2 (cm) (Table 3).

TABLE 1. Plant materials used for evaluation of cucumber genotypes used in this experiment

Code number	Genotype	Origin	Code number	Genotype	Origin
1	Green majic	Netherland	11	Adrian451	Netherland
2	Nasim	Netherland	12	Adrian salar	Turkey
3	Storm	Netherland	13	Neda	Turkey
4	Janeete	Netherland	14	Sina	USA
5	Karim	Russia	15	Tornado	Spain
6	Atilgan	France	16	Amiral	Spain
7	Vista	France	17	Sco4184	Denmark
8	Raneem	Netherland	18	Danish	Denmark
9	Zohal	Netherland	19	Yalda-R2	Netherland
10	Gohar	Netherland	20	Khassib-R2	Netherland

TABLE 2. Analysis of variance studied traits in cucumber genotypes

S.O.V.		Mean squares												
		¥TFY	FNP	FNN	BP	PH	LL	LW	FL	FD	DN	SHD	LB	VP
Replication	3	15028.6**	2.4	5.2**	0.01	13700.9**	24.02	19.5	2.2	0.09	471.06**	5.43**	1.08	0.3
Genotype	19	7132.5**	3.7**	1.4*	3.9**	4593.7**	15.5**	13.4**	4.1**	0.12*	252.19**	0.73**	3.5**	1.5**
Residual	57	3260.2	1.04	0.6	0.1	449.2	5.54	7.0	1.4	0.07	70.74	0.26	0.8	0.2

¥: TFY: Total fruit yield per pickling, FNP: Fruit number per pickling, FNN: fruit number per node, BP: branch number per plant, PH: plant height, LL: leaf length, LW: leaf width, FL: fruit length, FD: fruit diameter, DN: distance between internode, SHD: Shoot diameter, LB: length of branches, VP: Vigor of plant,

** and ** significant at P < 0.01 and P < 0.05, respectively.*

Different qualitative traits have been studied in different parts of the plant. Highest value for branch number per plant (BP) (3.87) and length of branch (LB) (3.87) was observed in genotype of Sina and the least value is belonged to Danish and Yalda R2 genotypes (Table 3). Vigor of plants had variation between 4 (Gohar) to 1.75 in Green-majic genotype (Table 3). Wehner and Guner [24] studied four cultigens under 10 planting days. In this research, number of nodes per branch was greatest on the lower branches, so branch length was probably dependent on the amount of time since formation. The number of branches per plant and nodes per branch was lowest at the intermediate planting dates. The tall indeterminate cultivar had the most branches and nodes, and the dwarf determinate inbred had the least.

The highest (36.37) and the least (29.12) values for length of leaf (LL) was observed in Tornado (36.37) (cm) and Nasim genotypes (29.12) (cm) (Table 3). In genotype Tornado, LW, PH and DN were also high; however fruit yield was the lowest. So this genotype had had good vegetative growth, but this growth could not result high fruit yield. Leaf width (LW) was varied from 34.37 (cm) in Gohar genotype to 26.82 (cm) in Sina genotype (Table 3). Length of branches (LB) varied from 3.87 (cm) in Sina to 1.12 (cm) in Danish and Yalda R2 (Table 3). Shoot diameter (SHD) varied from 2.03 (cm) in Green majic to 1(cm) in Sina. This result showed a high variation among evaluated genotypes for length of branches. Fruit diameter (FD) had a variation between 3.07 (cm) in Khassib-R2 to 2.32 (cm) in Atilgan (Table 3). The highest (17.27) (cm) and the least (12.88) (cm) values for fruit length was denoted to Vista and Khassib R2 genotypes, respectively (Table 3). Fruit length and fruit diameter are marketable traits that could have influence on marketing of cucumber and the acceptance range of them is different in different countries. Khasib showed the highest amount of TFY and FD, however the lowest amount of FL. The size of fruit is marketable trait in Iran. Chen et al. [25] evaluated different traits related to stem, leaf, fruit and flower in some hybrids between wild and cultivated cucumber. They reported that the diameter of the main stem and the shape and size of the leaves were intermediate when compared with their parents. Also the length× diameter and

length:diameter of fruits were different between diploid and tetraploid hybrids as tetraploid fruits were shorter in length.

TABLE 3. Mean comparisons of studied traits in cucumber genotypes.

Genotype	TFY¥	FNP	FNN	BP	PH	LL	LW	FL	FD	DN	SHD	LB	VP
1	404.05	6.52	1.83	3.75	375.06	31.62	31.25	14.73	2.50	89.87	2.03	3.5	1.75
2	430.05	3.92	2	1.25	294.6	29.12	28.81	14.51	2.51	86.87	1.96	1.62	3
3	398.94	3.58	1.33	3.12	332	30.31	30.18	14.77	2.58	78.25	2	1.87	2.5
4	421.33	5.45	1.95	1.92	329.3	29.75	30.31	14.86	2.6	78.62	1.99	1.5	2.7
5	350.01	3.50	1	3.62	376.4	34.43	33.93	16.12	2.46	87.37	1.98	3.5	2.7
6	357.99	3.76	1.04	3.5	373.8	33.75	33.50	15.4	2.32	101.5	1.90	3.62	2.71
7	415.98	3.96	2.04	1.37	315.25	30.06	29.68	17.27	2.70	84.56	2	2.37	2.5
8	377.59	4.74	1.37	3.25	321.8	31.25	31.12	15.62	2.42	86.25	1.95	1.62	2.5
9	363.83	4.07	2.20	3.12	365.6	33.75	32.87	16.81	2.40	92.81	1.98	3.37	3.37
10	474.25	5.11	2.62	2.37	327.3	33.75	34.37	15.53	2.43	89.31	1.98	1.87	4
11	451.72	6.52	2.66	3.62	327	35.5	32.12	15.48	2.67	88.25	1.22	3.25	3.75
12	364.23	3.91	2.41	1.87	317.5	31.12	29	14.95	2.77	96	1.17	2.12	3.87
13	343.52	3.58	2.54	2.37	381.25	32.87	30.5	14.72	2.65	105.5	1.10	2.25	3
14	439.02	5.36	2.27	3.87	370.05	30.87	26.82	15.63	2.77	100.2	1	3.87	2.12
15	338.31	3.50	2.12	3.37	331	36.37	32	15.82	2.67	102.2	1.25	3.37	2.5
16	360.32	3.76	1.95	2.87	294.25	31.62	28.25	14.87	2.87	92.25	1.05	2.75	2.62
17	367.49	3.96	2.74	3.37	374	33.62	33.50	15	2.67	104.7	1.17	3.62	2.87
18	420.95	4.74	2.68	1.12	327	32.37	30.5	13.97	2.70	86.50	1.05	1.12	3.37
19	354.26	4.07	3.41	0.87	267.25	30.50	31.25	13.56	2.70	94.75	1.17	1.12	3.5
20	459.93	5.65	2.14	1.50	287	32.25	31.75	12.88	3.07	89.75	1.35	1.5	3.87
LSD (5%)	80.84	1.44	1.18	0.79	30.01	3.33	3.76	1.67	0.37	11.90	0.73	1.27	0.63
Phenotypic CV(%)	14.4	22.8	39.3	37.6	10.13	7.30	8.5	7.8	10.2	9.16	32.9	35.9	38
Genotypic CV(%)	12	18	20.9	21.5	9.62	4.91	4.03	5.45	4.2	7.3	21.2	33.2	33
Phenotypic variance	3357.5	0.94	0.36	99.2	1148.2	3.8	3.35	1.03	0.03	63.04	0.17	0.88	0.38
Genotypic variance	2270.7	0.68	0.19	0.96	1035.9	2.5	1.58	0.68	0.01	45.36	0.11	0.68	0.33

¥: TFY: Total fruit yield per pickling, FNP: Fruit number per pickling, FNN: fruit number per node, BP: branch number per plant, PH: plant height, LL: leaf length, LW: leaf width, FL: fruit length, FD: fruit diameter, DN: distance between internode, SHD: Shoot diameter, LB: length of branches, VP: Vigor of plant

TABLE 4- Analysis of variance and mean comparison between groups of cluster analysis in cucumber genotypes

Traits	Mean square between groups	Mean			
		Group 1	Group 2	Group 3	Group 4
TFY	6749.7**	353.5^{d}	442.3^{a}	408.8^{ab}	391.9b^{c}
FNN	0.63*	1.94^{ab}	2.34^{ab}	1.74^{b}	2.52^{a}
BP	3.45**	3.23^{a}	3.4^{a}	2.18^{b}	1.65^{b}
LB	3.59**	3.29^{a}	3.12^{a}	1.8^{b}	1.72^{b}
FL	2.67*	15.6^{a}	15.3^{a}	15.4^{a}	14.1^{b}
FD	0.09*	2.53^{b}	2.59^{b}	2.56^{b}	2.82^{a}
DN	236.7**	99.1^{a}	91.9^{a}	82.9^{b}	91.8^{b}
PH	4955.7**	367.1^{a}	350^{a}	318.6^{b}	299^{b}
FNP	3.77**	3.73^{b}	5.88^{a}	4.33^{b}	4.43^{b}
LL	16.19**	34.1^{a}	32.9^{ab}	31.6^{c}	30.1b^{c}
LW	26.9*	32.7^{a}	31.6^{ab}	30^{b}	30.2^{b}
VP	0.57^{ns}	2.87^{ab}	2.9^{ab}	2.65^{b}	3.45^{a}
SHD	0.57*	1.55^{ab}	1.56^{ab}	1.99^{a}	1.16^{b}

TFY: Total fruit yield per pickling, FNP: Fruit number per pickling, FNN: fruit number per node, BP: branch number per plant, PH: plant height, LL: leaf length, LW: leaf width, FL: fruit length, FD: fruit diameter, DN: distance between internode, SHD: Shoot diameter, LB: length of branches, VP: Vigor of plant.

Means followed by the same letter was not significantly different at 0.05 level using LSD test.

,: F-test significant at P<0.05 and P<0.01 respectively; ns: not significant*

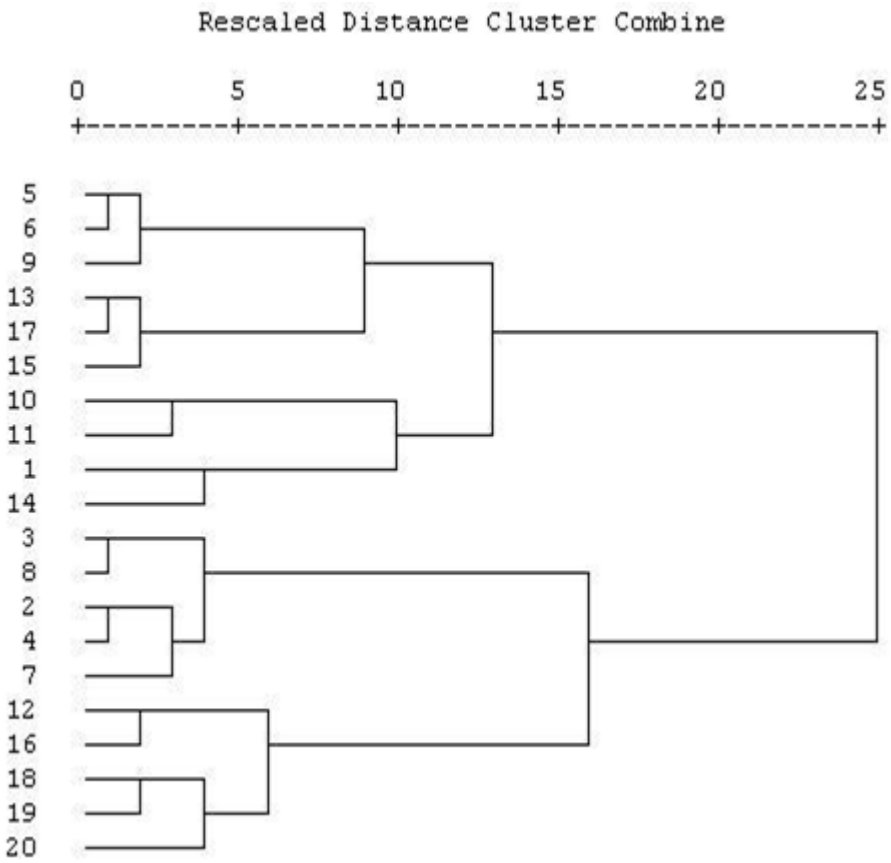

Figure 1. Ward-based dendrogram showing genetic relationship among 20 cucumber genotypes based on morphological, yield and its components traits.

Distance of internode had a broad variation that ranges from 105.5 (cm) in Neda to 78.25 (cm) in Storm (Table 3). Ramirez *et al.* [26] showed that multiple fruiting was associated with high leaf area. On the other hand, line with highest stem length had a greater proportion of dry weight into leaves, stems and roots and number of fruit per plant. Line Calypso had produced the same amount of total dry weight and fruit weight as the line with the highest amount of these traits, in spite of having a smaller leaf area that can be related to increase leaf photosynthetic rates.

The highest and the lowest genetic variation (CV) (%) were observed for VP and LW, respectively. The highest and the least value for phenotypic variation was observed to VP and LL, respectively (Table 3). Small difference between phenotypic and genetic coefficient of variations for most of the studied traits indicated that most of the observed variation was compromised from genetic factors.

Cluster analysis

Clustering based on studied traits separated the genotypes into four main groups (Fig. 1). There were 6, 4, 5 and 5 genotypes in these groups. This result indicated that there is a significant difference between evaluated genotypes for morphological traits. Cluster 1 included Karim (Russia), Atilgan (France), Zohal (Netherland), Neda (Turkey), Sco4184 (Denmark) and Tornado (Spain). Cluster 2 included Gohar (Netherland), Adrian-451 (Turkey), Green majic (Netheland) and Sina (USA). Cluster 3 included Storm, Raneem, Nasim and Janeete (Netheland) and Vista (France). Finally, cluster 4, included Adrian salar (Turkey), Amiral (spain), Danish (Denmark), Yalda-R2 and khassib-Rr2 (Netheland). This result revealed that there is no any overlap for geographical dispersion and measured traits. Of course, there were only Netherland genotypes in group number 3.

Analysis of variance was carried out to compare the means of groups (Table 4). The results showed that the difference between groups was significant for all of the studied traits except for VP (Table 4). The highest mean for LW, PH, LB, FL, DN and LL were denoted to cluster number 1 (Table 4). The genotypes with suitable morphological traits were clustered in group number 1. According to Table 4, the highest mean for TFY, FNN, and FNP was belonged to cluster number 2. Hence, these superior genotypes such as Green-majic and Gohar are recommended for fruit yield improvement, as a high yield parent in hybridization programs of cucumber. In cluster number 3, the genotypes did not show superior characteristics, except for SHD. The highest value for FNN, FD, and

VP were belonged to cluster number 4. These genotypes are originated from different geographical regions. It could be concluded that the genotypes in cluster number 2, could be used for enhancement of fruit yield and its components in cucumber breeding.

DISCUSSION

The result of this research could be compromised from a broad genetic diversity among selected germplasm. It seems that there is a high variation for FNP among evaluated genotypes. Genotypes with high TFY also showed high FNP with the exception of genotype Green majic with low TFY. To give attention to other traits in this genotype revealed that FL, FD and FNN were low that caused yield reduction. Fruit number was found to be more stable measure of productivity than fruit weight for cucumber [6].

For some genotypes, some traits were high and other traits were in low amount. This result confirms that consideration of few traits is not suitable way for selecting best genotypes. Because different traits have interaction between them, and direct and indirect effects of some traits make main traits such as fruit yield.

Some genotypes with highest TFY, showed slight PH and DN. It is clear that other traits such as FNP and FNN are more important than traits related to vegetative growth. FNP and FNN are the most important components of TFY in cucumber. Cramer and Wehner [27] studied four cucumber populations over two seasons and years. They found that fruit yield and its components such as number of branches per plant, number of different fruit size per plant and percentage of pistillate nodes differed between populations and environments. Some populations showed the highest amount of different traits as the lowest amount for other traits. For example, NCWBS population had a lower mean yield and poorer fruit quality and higher average fruit set. On the other hand, there was no difference between populations for some traits such as branch number and nodes per branch.

The superior genotypes for fruit yield and its components could be used as superior genotypes in cucumber cultivation, and also can be recommended for fruit yield improvement, as a high yield parent in hybridization programs of cucumber. Gohar, Khasib-R2, Adrian-451, Nasim and Sina showed the highest amount of fruit yield and fruit number. On the other hand, the other traits also were high in these genotypes that emphasize on better use from environmental conditions such as high photosynthesis by large leaves and good vigor. Over all, these genotypes in view point of TFY could be high yield genotypes for its cultivation in Iran. Fan *et al.* [28] used from long-fruited (length: diameter ratio) and number of fruits for selecting the best genotypes in three cycles of phenotypic mass selection. There were significant differences detected among cycles of phenotypic selection. A positive response to three cycles of phenotypic selection was detected for length: diameter ratio.

Overall, there were high genetic variation between these genotypes and can be used for different aims in greenhouses and also in breeding programs.

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