GENETIC DIVERSITY OF FORTY SUGAR CANE (Saccharum officinarum L.)VARIETIES BASED ON QUANTITATIVE TRAITS AND ISOPEROXIDASE ACTIVITY

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ABSTRACT

The present study was carried out at Sabahia Agricultural Research Station, Alexandria, Egypt. The main objectives of this study were to determine the genetic diversity of forty sugar cane varieties employing seven quantitative characters (stalk weight, stalk length, stalk diameter, number of internodes, total soluble solids, sucrose percentage and purity percentage); and to investigate peroxidase isozyme patterns (isoperoxidase). The field experiments were conducted at Sabahia Field Experiment Station during two successive seasons (2002 and 2003). The results obtained showed that there were highly significant differences among the forty studied varieties in all studied characters, and the cluster analysis for both quantitative traits and isozyme patterns was capable to differentiate the forty studied varieties into groups that could be used to choose parents for hybridization in breeding programs.

Key words: Sugar cane, isozyme, peroxidase, Euclidean distance, quantitative traits.

INTRODUCTION

Sugar cane, as a perennial grass plant, belongs to the *Saccharum officinarum* L. species as a member of *Saccharum* genus. *Saccharum* is a genus of between 6 to 37 species (depending on taxonomic interpretation) of tall grasses (Family: Poaceae, Tribe: Andropogoneae), native to warm temperate to tropical regions of the old world. Sugar cane has stout, jointed fibrous stalks, 2–6 m tall, and sap rich in sugar. However, very little is known about sugar cane genetics. The basic chromosome number estimation in *Saccharum* has been as diverse as x = 5, 6,8,10 or 12 and varied with species (Sreenivasan *et al*., 1987). The mating systems in natural populations have not been documented. Despite these drawbacks, sugar cane is an example of a very successful use of alien genetic resources. The interspecific hybridization programs that were carried out at the beginning of the 20th century, in Java and India, revolutionized sugar cane breeding. Modern sugar cane varieties result from interspecific hybridization and may contain more than 100 chromosomes contributed by up to five different species (Heinz, 1987). It encompasses very diverse euploid and aneuploid members (2n = 40 – 128), Lu *et al.* (1994). This wide range of chromosome number of sugar cane gives the breeders the chance to successfully practice his selection program.

Nowadays, commercial sugar cane cultivars are almost exclusively resulted from backcrosses, involving *S. officinarum* and *S. spontaneum*. However, only a few clones of these species were used. This narrow genetic base of modern hybrid varieties is surely one of the principal causes of the present slow rate of sugar cane breeding progress (Berding and Roach, 1987).

In Egypt, sugar cane provides about 75% of the sugar supply, while sugar beet provides around 25%. There is a gab of about 600.000 tons of sugar between the consumption and the production of sugar, and this amount is yearly imported from abroad and costs the country a lot of foreign currency. The plan of the government is to minimize this gab by the extension of both crops (cane and beet). In sugar cane, only vertical expansion could be applied because horizontal expansion needs a lot of irrigation water, which is relatively limited. To develop new desired sugar cane varieties and to improve the existing ones is the main goal of the breeder to achieve the vertical expansion in sugar cane.

Many of the desirable characters are quantitative, so multivariate statistical techniques have been suggested and utilized, to a limited extent, to measure genetic and phenotypic divergence among entries and genotypes to aid in planning crosses among genotypes belonging to different clusters (Whitehouse, 1969; Bhatt, 1970: Goodman, 1973; Sneath, 1976; Camussi *et al*., 1983). Characterization and quantification of genetic diversity, both within and among populations, has long been a major goal in evolutionary biology. In plant breeding programs, information concerning the genetic diversity within a crop species is essential for a rational use of genetic resources. It is particularly useful in the characterization of individual accessions and cultivars, in detecting duplications of genetic material in collections and as a general guide in the choice of parents for breeding hybrids.

Isozymes, which are multiple molecular forms of an enzyme, have recently been used to characterize cultivars in a number of crop species. Zhang *et al* (1993) and Heun *et al* (1994) used the isozymes to estimate the genetic diversity in the comparison of RFLP and RAPD techniques. Adam *et al* (1987) estimated the genetic distance, using the isozyme analysis. Several investigators studied the biochemical genetic assays to determine genetic markers in several plants (Abe *et al*, 1997 and Saleh, 1999). In sugar cane, isozyme variations have not been widely employed, although they were discussed for the first time as early as 1969 (Heinz, 1969). Glaszmann *et al.* (1989) used isozyme technique to identify molecular genetic markers usable in sugar cane

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breeding. They surveyed isozyme variations in samples of wild and noble clones, representing the species thought to be the closest relatives of the modern commercial clones.

The main purpose of the present investigation is to study the genetic diversity among forty sugar cane varieties, using both quantitative traits and isoperoxidase patterns to give more genetic information that could be used in the breeding program and as a general guide in the choice of parents for hybridization.

MATERIALS AND METHODS

a) Materials:

Forty sugar cane varieties were used in this study. These varieties were imported from different origins and they differ in their flowering behavior, as shown in Table (1). Sugar cane varieties were grown in a randomized complete block design (RCBD) with four replicates.

b) Field experiment:

Cuttings, with two buds each were planted in the field of the Agricultural Research Station (Sabahia). Each variety was planted in plots contained 7 ridges, 1.5 meter wide and 5 meters long, the distance between each two cuttings was 30 cm. All cultural practices needed for growing sugar cane crop were applied for the two planting successive seasons. Planting took place on March in 2002 and 2003, as spring planting.

The observations and measurements were taken after 300 days from planting date of each season. Thirty plants were randomly taken from each experimental plot to measure the following vegetative characters:

1-Stalk weight (in g).
2-Stalk length (in Cm).
3-Stalk diameter (in Cm).
4-Number of internodes/stalk.
5-Total soluble solids (T.S.S.) percentage.
6-Sucrose percentage.
7-Purity percentage.

c) Statistical analysis:

The experimental design used in this investigation was randomized completed block design (RCBD) with four replicates, and the data were analyzed according to Steel and Torrie (1981). In order to detect patterns of genetic relationship in the varieties, data analysis on the means of clearly defined seven traits was initially performed, based on the Euclidean distance matrix. The output was analyzed, using an agglomerative hierarchical clustering method with complete linkage strategy. Firstly, the data were subjected to analysis to produce a matrix of dissimilarity values and the phenotypic distance between each pair of varieties was estimated as Euclidean distance. Secondly, cluster analysis was conducted on the Euclidean distance matrix with un-weighted pair-group method , based on arithmetic average (UPGMA) to develop a dendrogram, using the computer program, NTSYS-pc ver 2.1 (Rolhf, 2000).

d) Electrophoresis technique:

Peroxidase analysis:

Isozyme patterns of sugar cane varieties were done as the following procedure:

***Buffers*:**

0.23 M Tris - Citric acid buffer, pH 8.0 was prepared according to.

0.01 M Sodium acetate - acetic acid buffer, pH 5.0

Gel media:

Agar-starch-polyvinyl pyrolidine (P.V.P.) gel was prepared as that described by Sabrah and El-Metainy (1985).

Staining solution:

100 ml of 0.01 M sodium acetate - acetic acid buffer, pH 5.0 containing 0.1 gm benzidine were used and 0.5% hydrogen peroxide (H_2O_2) was added immediately before staining.

Procedure:

Leaf samples from just expanded leaves were homogenized in a cool mortar, and the homogenate was absorbed on filter paper strips, which placed on the origin line of agar gel plate for about one hour, electrophoretic started for two hours at current of 13–14 V/Cm. After separation, the peroxidase isozymes were stained and band scoring was done according to TOTAL LAB software V.1.11.Data were analyzed with the computer program, NTSYS-pc ver 2.1 (Rohlf,2000) to develop the cluster analysis.

RESULTS AND DISCUSSION

1. Field experiment:

1.1. Quantitative characters analysis:

Analysis of variance for the seven studied quantitative characters (stalk weight, stalk length, stalk diameter, number of internodes, total soluble solids, sucrose percentage and purity percentage) of the forty tested sugar cane varieties was presented in Table (2). Highly significant differences were noticed for the tested varieties in all examined traits. With regard to the interaction between varieties and years, highly significant differences were found for all traits under study. No significant differences in these characters were found between the two examined seasons, except for purity character,.

Mean values for different characters of forty sugar cane varieties for the two seasons are presented in Table (3).The data showed that G.T.54-9 variety had the highest values in stalk length, stalk diameter and total soluble solids characters. Ph.8013 variety had the highest values in number of internodes, stalk weight, sucrose percentage and purity percentage characters. But, these increased values were not

significant, compared to G.T.54-9 variety. While, C.34-33 variety had the lowest values in stalk weight, total soluble solids and sucrose percentage characters. Variety, Nco. 293, had the lowest values in purity percentage. L.61-49 variety had the lowest value in stalk length. These results are in good agreement with those reported by several authors [e.g. El-Manhaly (1987); Gaber *et al.* (1990); Tawfik *et al* (1997) and Younan *et al* (1997)] who found differences in some characters among some sugar cane varieties.

Varieties x years interactions were apparent in the rankings of the varieties in the two examined years. The evaluation of the studied varieties indicated that G.T.54-9, Ph8013, G85-37, G.87-249, G.75-368, Co.312, Co.421, Nco.310 and G.74-96. varieties gave a good performance for the seven studied characters over both years.

1.2. Cluster analysis based on quantitative traits:

Quantitative traits appear least suited as genetic markers because they are often modified by the environment subject to epistatic and pleiotropic effects, and coded by an unknown number of genes (Van Beuningen and Busch, 1997). An assumption, underlying the use of phenotypic similarity estimates based on quantitative traits, is that such estimates are an accurate reflection of genotypic similarity. Distance measures among varieties can be based on morphological or biochemical marker traits, pedigree information or quantitative morphological traits. The objectives of this study were to group sugar cane genotypes into clusters according to their morphological behavior.

The mean of the observations of all traits was averaged over the two years. The mean values were normalized prior to cluster analysis by dividing such values by the standard deviation and subtracting the mean for each trait. The matrix of Euclidean distance for all varieties was computed. The dissimilarity coefficient is based on interval measure data collected for the stable quantitative traits. Cluster analysis was, then, conducted on the Euclidean distance matrix with the un-weighted pair-group method, based on arithmetic averages (UPGMA) employing the NTSYS-pc ver. 2.1 software. Cluster analysis resulted in the varieties grouping is presented in Figure (1).

The cluster analysis was capable to differentiate the forty sugar cane varieties into seven groups. Group No.1 contained seven varieties (Co.281, Co.360, Bo.47, Bo.18, H.86-486, F.144 and M.253-48). Group No.2 contained ten varieties that existed in two sub clusters (C.63-46, Bo.22, H.86-471, Cp.27-93, Cp.44-101, Cp.66-346, F.116, H.86-197, Q.87 and L.61-49). While, F.135 variety equipped group No.3 alone. The remaining twenty-two varieties were grouped in No.4 (Co.312, Co.421, Nco.310, G.85-37, G.87-249 and G. 75-368), group No. 5 (G.74-96, Cp.30-29, G.84-47, M.35-15, Kassoer, Cp.36-13, L.62-96, Ph.98-97, Poj.2878, Q.58, Poj.105 and L.60-25), group No.6 (C.34-33 and Nco.293) and group No.7 contained two varieties (G.T.54-9 and Ph.8013). Euclidean distance, using quantitative traits between all pairs of varieties, ranged from 0.06 to 2.19. The lowest value between two varieties (0.06) was found between C.63-46 and Bo.22. Several investigators used the quantitative traits to study genetic diversity on several crops (Abd El-Hameed , 2004 and Saleh and Attalah, 2005).

2. Isozymes assay:

Isozymes had been defined by Shaw (1969) as multiple forms of enzymes in the same organism and having similar or identical catalytic activities. He classified isozymes into primary and secondary types. The primary type was distinctly different molecules, and was presumably produced from different genetic sites, while the secondary isozymes resulted from alteration in the structure of single polypeptide chain *in vitro* and many of these were artifacts. The primary isozymes were the only ones, which had a biological significance. Weising *et al.* (1995) reported that isozymes were enzymes that converted the same chemical substrate, but were not necessarily products of the same gene. Isozymes may be active at different life stages or in different cell compartments.

Early research on sugar cane genotypes had indicated that isozymes could be used to differentiate sugarcane clones (Fautret and Glaszmann, 1988). Cordeiro (2001) reported that the benefit from using isozyme method was the ability to identify and differentiate between closely and distantly related sugar canes.

2.1. Peroxidase analysis:

Leaf samples of forty sugar cane varieties were collected for isozymes analysis to differentiate between them with respect to gene expression. Four plates contained the forty plant samples for peroxidase assay every plate contained ten samples (Figures 2,3,4 and 5).

Scoring bands were carried out by using the computer program software, TOTALLAB V.1.11. The obtained data showed differences in band numbers, band volume, peak height and R.f. parameter in the investigated materials whereas:

- **Band volume:** It indicates the value resulting from the interaction between band area and band density. It refers to the amount of isozyme, which was expressed from a given gene.
- **Peak height:** It refers to the density of the band and this indicates the activity of the isozyme.
- **R.f. (Retardation factor):** It refers to the position of band from the original line to its position as relative number–typically between 0 and 1.

The data indicated that there were eight bands migrated towards the cathode (Table 4), while there were six bands migrated towards the anode (Table 5). Band existence, band volume, peak height and R.f.

Table (1): Sugar cane varieties; their origin and latitude.

Varieties	Nation	Breeding station	Latitude
Bo.18 Bo.22 Bo.47	India	Bihar	25° 59' N
Cp.27-93 Cp.30-29 Cp.36-13 Cp.44-101 Cp.66-346	U.S.A.	Canal point	26° 0' N
C.34-33 C.63-46	Cuba	Cuba, Central Jaranu	22° 04' N
Co.281 Co.312 Co.360 Co.421	India	Coimbatore	11° 0' N
F116 F135 F144	Taiwan	Formosa	23° 0' N
G.74-96 G.75-368 G.84-47 G.85-37 G.87-249 G.T.54-9	Egypt Taiwan (T)	Alexandria Sabahia Station	31° 12' N
H.86-197 H.86-471 H.86-486	U.S.A.	Hawaii	21° 30' N
Kassoer	India	Coimbator	11° 0' N
L.60-25 L.61-49 L.62-96	U.S.A.	Louisiana	29° 0' N
M35-15 M253-48	Mauritius	Mauritius	20° 15' S
Nco.293 Nco.310	South Africa	Natal	29° 0' S
Ph.8013 Ph.98-97	Philippines	Philippines	21° 10' N
Poj.105 Poj.2878	Indonesia	Java	7° 38' S
Q.58 Q.87	Australia	Queensland	19° 32' S

Table (2): Analysis of variance for the seven studied quantitative characters .

Source of variation	d.f.	Mean squares						
		Stalk length (cm)	Stalk diameter (cm)	Number of internodes / Stalk	Stalk weight (g)	Total solible solids (%)	Sucrose (%)	Purity (%)
Blocks	3	5650.33	1.92	94.25	297661.57	39.082	16.193	1.586
Years	1	2611.87 $^{N.S.}$	0.098 $^{N.S.}$	9.75 $^{N.S.}$	91226.28 $^{N.S.}$	2.403 $^{N.S.}$	11.885 $^{N.S.}$	163.573 *
Blocks x Years	3	728.47	0.229	25.84	75715.47	6.803	5.119	9.729
Varieties	39	6789.11 **	1.545 **	33.21 **	273606.84 **	24.58 **	27.622 **	483.81 **
Varieties X Years	39	210.34 **	0.037 **	1.87 **	9636.52 **	0.941 **	0.726 **	30.88 **
Error (combined)	234	55.93	0.016	0.84	2430.92	0.211	0.236	6.262

N.S. Not significant at 0.05 level of probability.
* Significant at 0.05 of probability.
** Significant at 0.01 of probability.

Table (3): Mean values for the seven studied quantitative characters in the tested varieties (average of two seasons; 2002 and 2003)

Characters / Varieties	Stalk length (cm)	Stalk diameter (cm)	Number of internodes	Stalk weight (g)	Total soluble solids (%)	Sucrose (%)	Purity (%)
Co.281	198.38	4.31	10.01	384.0	16.86	10.24	59.37
Co.312	219.53	3.62	14.58	757.38	20.76	14.4	74.54
Co.421	216.03	3.62	14.03	721.38	20.13	14.29	73.99
Bo.22	183.39	4.0	11.06	426.88	17.64	11.54	62.68
G.74-96	206.53	3.64	12.39	569.88	18.78	13.03	69.57
G.75-368	236.9	3.17	14.85	887.0	20.81	14.68	75.64
Poj.105	168.9	3.73	12.56	612.0	20.2	13.74	73.06
Cp.66-346	167.74	3.83	11.45	504.54	17.93	12.48	65.12
G.84-47	205.75	3.75	11.78	522.88	18.23	12.61	66.79
Bo.47	190.04	4.32	9.81	376.50	16.85	9.90	55.67
Cp.44-101	205.77	3.92	11.14	433.5	17.8	12.24	64.59
Cp.27-93	195.93	4.04	10.76	414.13	17.26	11.13	61.67
M.253-48	161.03	4.24	10.38	406.25	17.16	10.59	61.01
Cp.30-29	197.30	3.63	12.39	578.5	18.80	13.09	70.09
C.63-46	180.28	4.03	11.01	423.13	17.59	11.36	62.36
Co.360	182.74	4.27	10.14	393.38	17.07	10.38	59.60
Ph.8013	256.21	4.56	16.46	939.5	21.39	15.28	79.73
Nco.310	215.56	3.49	13.88	683.38	20.08	13.89	73.06
Cp.36-13	170.95	3.60	12.51	602.5	19.42	13.24	71.05
Bo.18	183.99	4.52	9.35	330.88	16.31	9.68	54.91
C.34-33	164.56	5.16	7.99	252.88	14.11	8.05	47.47
F.144	154.18	4.27	10.13	387.13	16.96	10.4	59.59
F.135	161.41	3.53	8.60	307.13	16.10	8.80	52.50
Nco.293	205.56	4.95	8.44	253.38	16.04	8.75	51.79
F.116	165.15	3.86	11.31	483.25	17.84	12.43	64.71
H.86-471	177.60	3.94	11.04	432.25	17.66	11.84	64.41
G.87-249	238.68	3.41	15.15	888.0	20.94	14.84	76.54
G.T.54-9	266.75	5.16	15.49	933.13	21.83	15.18	78.94
H.86-197	174.33	3.81	11.58	504.54	17.93	12.53	65.20
M.35-15	178.49	3.70	11.84	544.88	18.41	12.68	67.86
L.61-49	143.73	3.75	11.6	514.0	18.04	12.56	65.46
L.60-25	164.25	3.52	13.6	679.75	20.03	13.75	72.62
G.85-37	248.69	3.40	15.47	897.5	21.2	15.06	77.09
L.62-96	166.96	3.41	12.44	591.63	19.20	13.14	70.40
Ph.98-97	181.31	3.41	12.5	597.0	19.37	13.34	70.1
Kassoer	187.95	3.68	11.88	562.0	18.56	12.79	69.49
Poj.2878	185.24	3.43	13.06	646.63	19.96	13.39	71.78
Q.58	189.35	3.48	13.38	673.75	19.99	13.49	72.52
H.86-486	145.83	4.07	10.51	411.38	17.18	11.11	61.50
Q.87	174.33	3.65	11.08	485.88	17.86	12.53	64.95
L.S.D.0.05	20.10	0.84	1.89	136.0	1.30	1.20	7.70
L.S.D.0.01	26.40	1.11	2.50	178.0	1.80	1.60	10.10

Table (4): Analysis of electrophoretic data of peroxidase isozymes obtained from forty sugar cane varieties in the cathodal migration .

Genotypes	Band 1			Band 2			Band 3			Band 4			Band 5			Band 6			Band 7			Band 8		
	Vol.	Peak Heig.	R.f.	Vol.	Peak heig.	R.f.	Vol.	Peak heig.	R.f.	Vol.	Peak heig.	R.f.	Vol.	Peak heig.	R.f.	Vol.	Peak heig.	R.f.	Vol.	Peak heig.	R.f.	Vol.	Peak heig.	R.f.
1	-	-	-	93.89	0.12	0.15	233.91	0.25	0.29	-	-	-	709.6	0.80	0.50	577.1	0.65	0.73	-	-	-	-	-	-
2	-	-	-	439.5	0.60	0.17	518.97	0.63	0.29	-	-	-	1077.1	1.34	0.54	-	-	-	242.7	0.33	0.80	-	-	-
3	-	-	-	362.7	0.52	0.16	236.1	0.46	0.24	-	-	-	874.1	0.97	0.54	-	-	-	128.9	0.16	0.79	-	-	-
4	-	-	-	368.8	0.51	0.15	341.0	0.53	0.24	20.83	0.05	0.32	655.7	0.92	0.52	-	-	-	-	-	-	-	-	-
5	-	-	-	493.8	0.51	0.15	360.1	0.52	0.23	115.4	0.20	0.32	964.5	0.99	0.54	-	-	-	202.2	0.22	0.76	-	-	-
6	-	-	-	392.1	0.42	0.16	271.7	0.41	0.22	159.1	0.32	0.30	891.7	0.89	0.56	-	-	-	-	-	-	-	-	-
7	-	-	-	455.3	0.44	0.16	533.0	0.61	0.26	-	-	-	760.4	0.80	0.56	-	-	-	-	-	-	123.8	0.24	0.90
8	182.59	0.23	0.13	163.8	0.29	0.19	-	-	-	-	-	-	933.2	0.96	0.56	-	-	-	-	-	-	-	-	-
9	-	-	-	272.5	0.35	0.18	-	-	-	486.0	0.56	0.31	974.6	1.09	0.55	-	-	-	-	-	-	230.9	0.31	0.88
10	-	-	-	411.5	0.54	0.18	491.6	0.54	0.28	-	-	-	911.5	1.04	0.55	-	-	-	-	-	-	-	-	-
11	82.97	0.14	0.09	-	-	-	320.93	0.46	0.28	-	-	-	278.5	0.49	0.59	-	-	-	-	-	-	74.34	0.12	0.87
12	29.56	0.05	0.07	34.13	0.05	0.19	-	-	-	-	-	-	55.54	0.10	0.57	-	-	-	-	-	-	-	-	-
13	84.44	0.13	0.10	-	-	-	115.83	0.19	0.23	-	-	-	157.5	0.26	0.58	-	-	-	-	-	-	-	-	-
14	97.86	0.14	0.08	132.26	0.19	0.20	-	-	-	-	-	-	140.2	0.24	0.58	-	-	-	34.45	0.06	0.85	-	-	-
15	326.5	0.52	0.10	-	-	-	293.6	0.34	0.25	-	-	-	221.2	0.35	0.57	-	-	-	-	-	-	-	-	-
16	79.02	0.15	0.11	116.13	0.19	0.19	-	-	-	-	-	-	209.2	0.34	0.56	-	-	-	-	-	-	-	-	-
17	156.2	0.17	0.12	-	-	-	-	-	-	-	-	-	199.8	0.24	0.58	-	-	-	-	-	-	32.9	0.08	0.90
18	91.94	0.14	0.12	-	-	-	-	-	-	-	-	-	165.7	0.24	0.57	-	-	-	72.4	0.13	0.83	-	-	-
19	99.83	0.16	0.11	-	-	-	118.5	0.19	0.23	-	-	-	94.96	0.17	0.56	-	-	-	-	-	-	-	-	-
20	325.36	0.32	0.11	-	-	-	-	-	-	-	-	-	264.24	0.37	0.53	-	-	-	49.8	0.09	0.77	-	-	-

Cont. Table (4): Analysis of electrophoretic data of peroxidase isozymes obtained from forty sugar cane varieties in the cathodal migration

Genotypes	Band 1			Band 2			Band 3			Band 4			Band 5			Band 6			Band 7			Band 8		
	Vol.	Peak heig.	R.f.	Vol.	Peak heig.	R.f.	Vol.	Peak heig.	R.f.	Vol.	Peak heig.	R.f.	Vol.	Peak heig.	R.f.	Vol.	Peak heig.	R.f.	Vol.	Peak heig.	R.f.	Vol.	Peak heig.	R.f.
21	95.83	0.43	0.06	-	-	-	-	-	-	-	-	-	195.9	1.54	0.54	-	-	-	91.4	0.46	0.81	-	-	-
22	93.78	1.83	0.07	-	-	-	54.3	0.85	0.28	-	-	-	247.5	1.41	0.56	-	-	-	-	-	-	-	-	-
23	200.8	1.37	0.08	-	-	-	167.9	1.56	0.29	-	-	-	345.7	0.41	0.57	-	-	-	-	-	-	-	-	-
24	29.3	0.81	0.10	-	-	-	-	-	-	-	-	-	225.3	0.40	0.57	-	-	-	73.2	1.37	0.83	-	-	-
25	527.3	2.01	0.10	-	-	-	465.9	1.65	0.27	-	-	-	231.1	1.30	0.57	-	-	-	147.7	0.72	0.84	-	-	-
26	90.0	1.1	0.08	-	-	-	83.5	0.82	0.27	-	-	-	450.4	1.63	0.56	-	-	-	-	-	-	-	-	-
27	76.1	0.99	0.09	-	-	-	-	-	-	-	-	-	299.8	0.21	0.55	-	-	-	35.06	1.51	0.83	-	-	-
28	148.3	1.0	0.10	-	-	-	-	-	-	27.3	0.27	0.35	263.1	0.25	0.54	-	-	-	69.45	1.42	0.84	-	-	-
29	72.2	0.72	0.07	-	-	-	149.4	1.05	0.24	-	-	-	215.0	1.41	0.54	-	-	-	-	-	-	-	-	-
30	90.4	0.92	0.09	-	-	-	44.8	0.58	0.26	-	-	-	203.6	1.34	0.52	-	-	-	58.9	0.19	0.84	-	-	-
31	414.8	0.14	0.11	-	-	-	-	-	-	-	-	-	1216.7	0.56	0.54	323.7	0.03	0.67	-	-	-	-	-	-
32	-	-	-	-	-	-	-	-	-	-	-	-	1272.7	0.70	0.54	474.3	0.36	0.67	246.9	0.10	0.76	-	-	-
33	425.1	0.23	0.12	-	-	-	-	-	-	-	-	-	857.3	0.63	0.53	-	-	-	330.9	0.20	0.76	-	-	-
34	544.9	0.32	0.11	578.0	0.19	0.20	-	-	-	-	-	-	1530.1	0.75	0.54	-	-	-	327.6	0.09	0.76	382.3	0.25	0.90
35	676.7	0.27	0.14	-	-	-	-	-	-	-	-	-	1452.1	0.64	0.54	383.2	0.06	0.65	-	-	-	457.1	0.44	0.89
36	587.8	0.54	0.14	-	-	-	-	-	-	599.3	0.19	0.32	1399.1	0.67	0.53	-	-	-	380.4	0.15	0.75	436.5	0.41	0.88
37	-	-	-	402.5	0.08	0.20	-	-	-	-	-	-	1120.6	0.53	0.54	370.0	0.11	0.65	269.3	0.05	0.75	-	-	-
38	-	-	-	-	-	-	-	-	-	-	-	-	1579.4	0.65	0.55	-	-	-	398.3	0.11	0.77	274.0	0.10	0.89
39	418.8	0.17	0.13	-	-	-	-	-	-	-	-	-	1513.0	0.46	0.56	-	-	-	43.3	0.11	0.76	368.5	0.25	0.88
40	-	-	-	422.4	0.09	0.20	-	-	-	-	-	-	1529.4	0.56	0.54	-	-	-	501.4	0.22	0.75	369.4	0.34	0.87

Table (5): Analysis of electrophoretic data of peroxidase isozymes obtained from forty sugar cane varieties in the anodal migration.

Genotypes	Band 1			Band 2			Band 3			Band 4			Band 5			Band 6		
	Volume	Peak height	R.f.	Volume	Peak height	R.f.	Volume	Peak height	R.f.	Volume	Peak height	R.f.	Volume	Peak height	R.f.	Volume	Peak height	R.f.
1	-	-	-	-	-	-	61.5		0.52	-	-	-	109.8		0.64	100.2		0.73
2	186.3		0.19	-		-	-	-	-	-	-	-	457.3		0.69	-	-	-
3	29.7		0.20	-	-	-	186.6		0.53	-	-	-	-	-	-	517.3		0.76
4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	624.7		0.78
5	25.5		0.15	-	-	-	176.6		0.53	-	-	-	482.5		0.69	-	-	-
6	9.01		0.15	19.2		0.38	-	-	-	-	-	-	374.1		0.69	-	-	-
7	34.99	-	0.16	36.0		0.39	-	-	-	-	-	-	407.1		0.65	334.1		0.74
8	-	-	-	-	-	-	-	-	-	448.5		0.60	-	-	-	992.3		0.79
9	99.6		0.17	34.5		0.40	-	-	-	-	-	-	-	-	-	610.1		0.72
10	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	465.3		0.79
11	-	-	-	-	-	-	86.2	0.22	0.55	-	-	-	253.4	0.50	0.67	-	-	-
12	-	-	-	-	-	-	85.95	0.17	0.49	-	-	-	-	-	-	-	-	-
13	41.97	0.07	0.30	-	-	-	74.5	0.18	0.50	56.30	0.10	0.59	-	-	-	-	-	-
14	-	-	-	99.2	0.25	0.48	-	-	-	141.5	0.35	0.58	-	-	-	-	-	-
15	-	-	-	23.6	0.10	0.48	54.8	0.15	0.51	144.2	0.16	0.16	-	-	-	-	-	-
16	-	-	-	43.7	0.12	0.39	20.2	0.06	0.50	140.4	0.22	0.62	-	-	-	-	-	-
17	-	-	-	-	-	-	-	-	-	114.5	0.23	0.58	100.16	0.21	0.68	-	-	-
18	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	84.85	0.14	0.71
19	47.39	0.07	0.29	83.8	0.11	0.43	-	-	-	-	-	-	179.5	0.18	0.69	-	-	-
20	-	-	-	-	-	-	-	-	-	-	-	-	86.5	0.18	0.65	130.98	0.22	0.74

Cont. Table (5): Analysis of electrophoretic data of peroxidase isozymes obtained from forty sugar cane varieties in the anodal migration.

Genotypes	Band 1			Band 2			Band 3			Band 4			Band 5			Band 6		
	Volume	Peak height	R.f.	Volume	Peak height	R.f.	Volume	Peak height	R.f.	Volume	Peak height	R.f.	Volume	Peak height	R.f.	Volume	Peak height	R.f.
21	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
22	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
23	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
24	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	189.5	0.82	0.80
25	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	296.1	0.63	0.82
26	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	420.5	0.75	0.77
27	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	230.6	0.81	0.80
28	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	73.6	0.80	0.82
29	131.0	0.85	0.06	-	-	-	-	-	-	-	-	-	-	-	-	89.3	0.72	0.83
30	-	-	-	-	-	-	135.9	0.58	0.51	-	-	-	157.3	0.81	0.69	-	-	-
31	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
32	-	-	-	-	-	-	-	-	-	-	-	-	99.1	0.41	0.67	-	-	-
33	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	85.2	0.37	0.88
34	-	-	-	-	-	-	-	-	-	-	-	-	227.0	0.57	0.68	-	-	-
35	-	-	-	-	-	-	-	-	-	-	-	-	128.1	0.35	0.67	-	-	-
36	-	-	-	-	-	-	-	-	-	-	-	-	408.5	0.86	0.66	-	-	-
37	-	-	-	-	-	-	-	-	-	-	-	-	133.9	0.37	0.66	-	-	-
38	-	-	-	-	-	-	-	-	-	124.1	0.39	0.60	-	-	-	128.9	0.52	0.87
39	-	-	-	-	-	-	-	-	-	171.0	0.51	0.60	-	-	-	312.6	0.41	0.86
40	-	-	-	-	-	-	-	-	-	211.1	0.46	0.60	-	-	-	257.0	0.39	0.86

Table (6): Matrix analysis of peroxidase isozymes for forty sugar cane studied varieties in the cathodal and anodal migration.

	Genotypes / Band No.	Co.281	Co.312	Co.421	Bo.22	G.74-96	G.75-368	Poj.105	Cp.66-346	G.84-47	Bo.47	Cp.44-101	Cp.57-93	M.253-48	Cp.30-29	C.65-46	Co.360	Ph.8013	Neo.310	Cp.36-13	Bo.18	C.34-33	F.144	F.135	Neo.293	F.116	H.86-471	G.87-249	G.T.54-9	H.86-197	M.15-15	L.61-49	L.60-25	G.85-37	L.62-96	Ph.90-97	Kamoor	Poj.2878	Q.58	H.86-486	Q.87
-	1	0	0	0	0	0	0	0	1	0	0	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	0	1	1	1	1	0	0	1	0
	2	1	1	1	1	1	1	1	1	1	1	0	1	0	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	1	0	0	1
	3	1	1	1	1	1	1	1	0	0	1	1	0	1	0	1	0	0	0	1	0	0	1	1	0	1	1	0	0	1	1	0	0	0	0	0	0	0	0	0	0	0
	4	0	0	0	1	1	1	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	1	0	0	0	0	0
	5	[illegible]																																							
	6	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	0	0	1	0	1	0	0	0
	7	0	1	1	0	1	0	0	0	0	0	0	0	0	1	0	0	0	1	0	1	1	0	0	1	1	0	1	1	0	1	0	0	1	1	0	1	1	1	1	1	1
	8	0	0	0	0	0	0	1	0	1	0	1	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	0	1	1	1
+	1	0	1	1	0	1	1	1	0	1	0	0	0	1	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0
	2	0	0	0	0	0	1	1	0	1	0	0	0	0	1	1	1	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	3	1	0	1	0	1	0	0	0	0	0	1	1	1	0	1	1	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0
	4	0	0	0	0	0	0	0	1	0	0	0	0	1	1	1	1	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	1
	5	1	1	0	0	1	1	1	0	0	0	1	0	0	0	0	0	1	0	1	1	0	0	0	0	0	0	0	0	0	1	0	1	0	1	1	1	1	0	0	0	0
	6	1	1	1	1	0	0	1	1	1	1	0	0	0	0	0	0	0	1	0	1	0	0	0	1	1	1	1	1	1	0	0	0	1	0	0	0	0	1	1	1	1

Whereas: (0): Refers to band absence – (1) Refers to band presence.

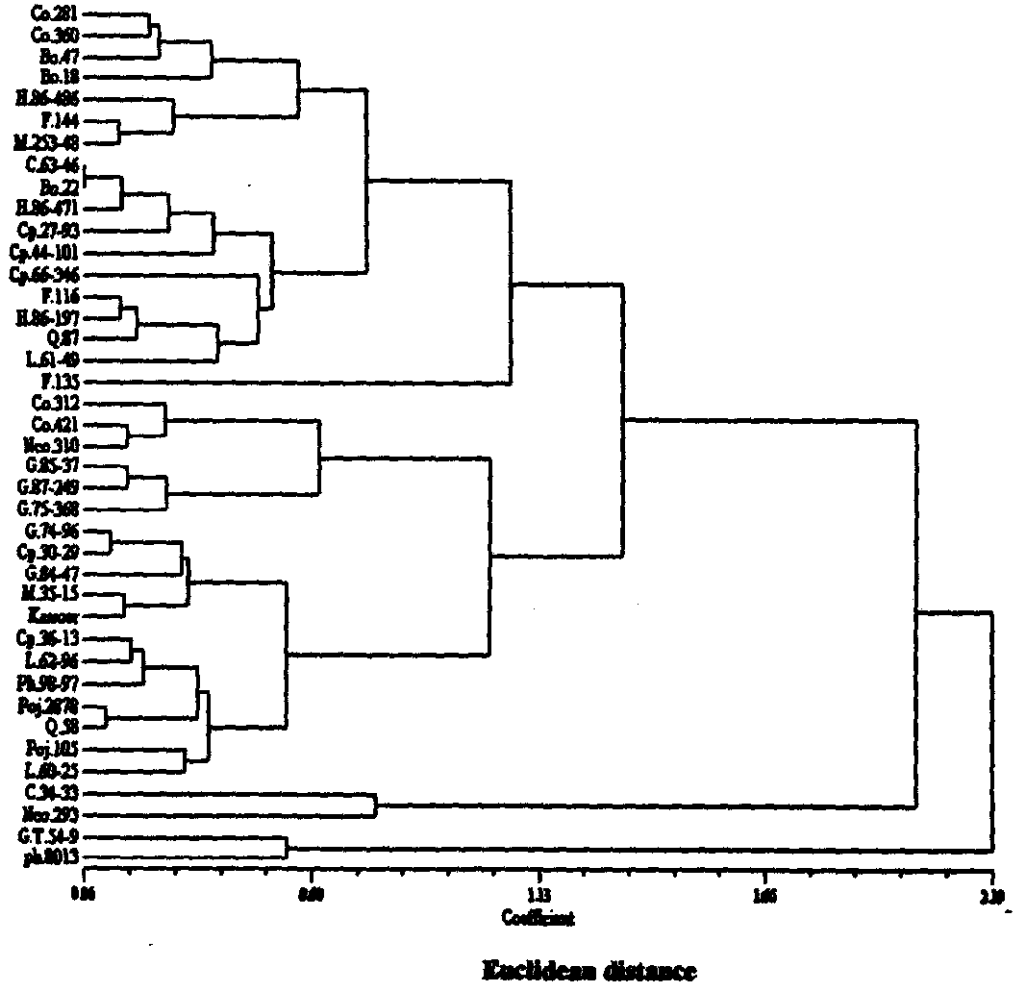

Figure (1): Dendrogram of forty sugar cane varieties based on seven quantitative traits.

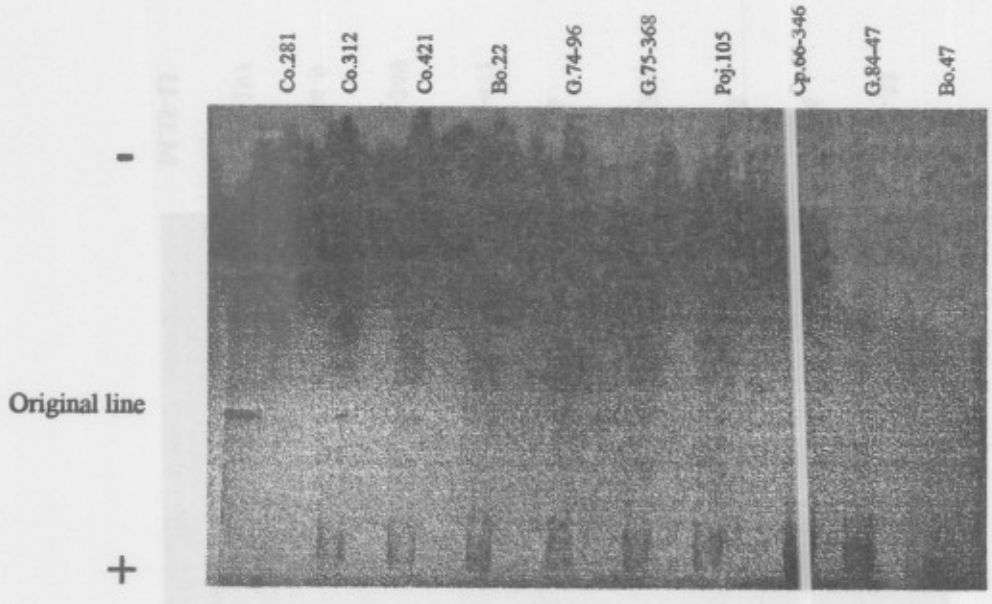

Figure (2): Electrophoretic patterns of peroxidase isozymes for ten sugar cane varieties (1-10).

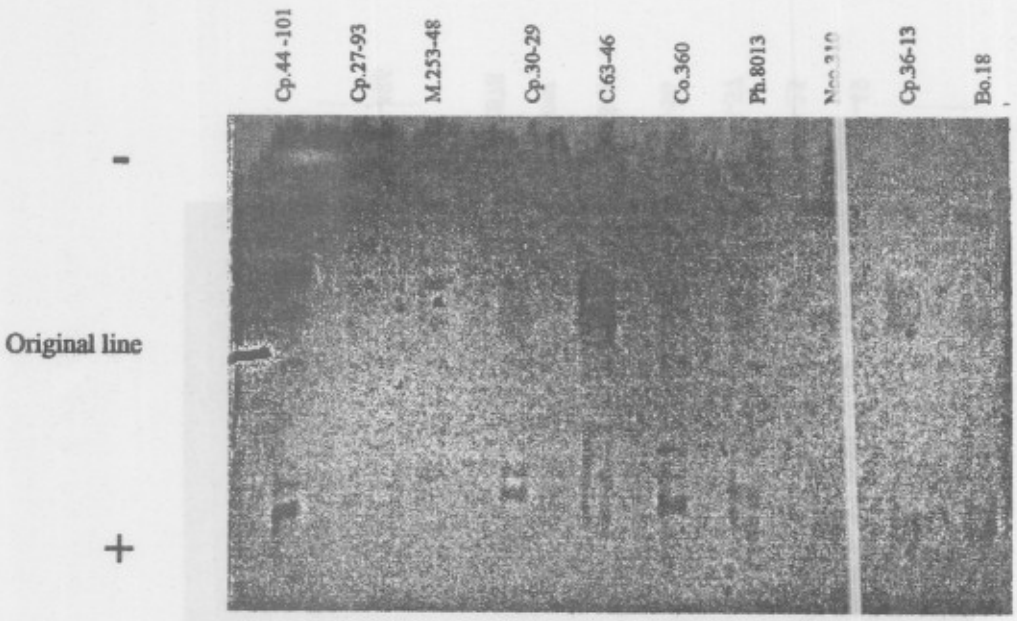

Figure (3): Electrophoretic patterns of peroxidase isozymes for ten sugar cane varieties (11-20).

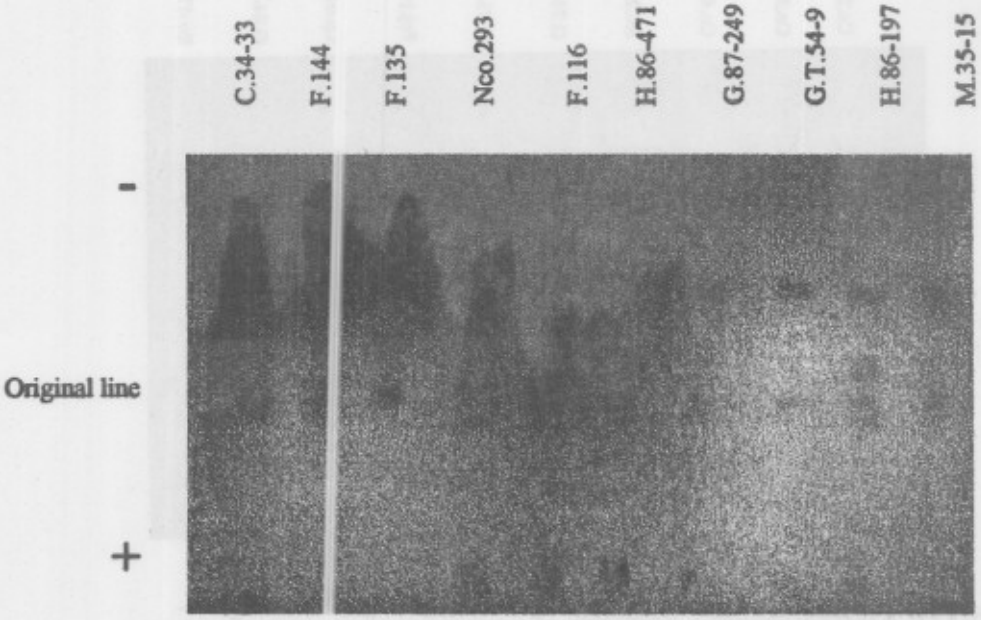

Figure (4): Electrophoretic patterns of peroxidase isozymes for ten sugar cane varieties (21-30).

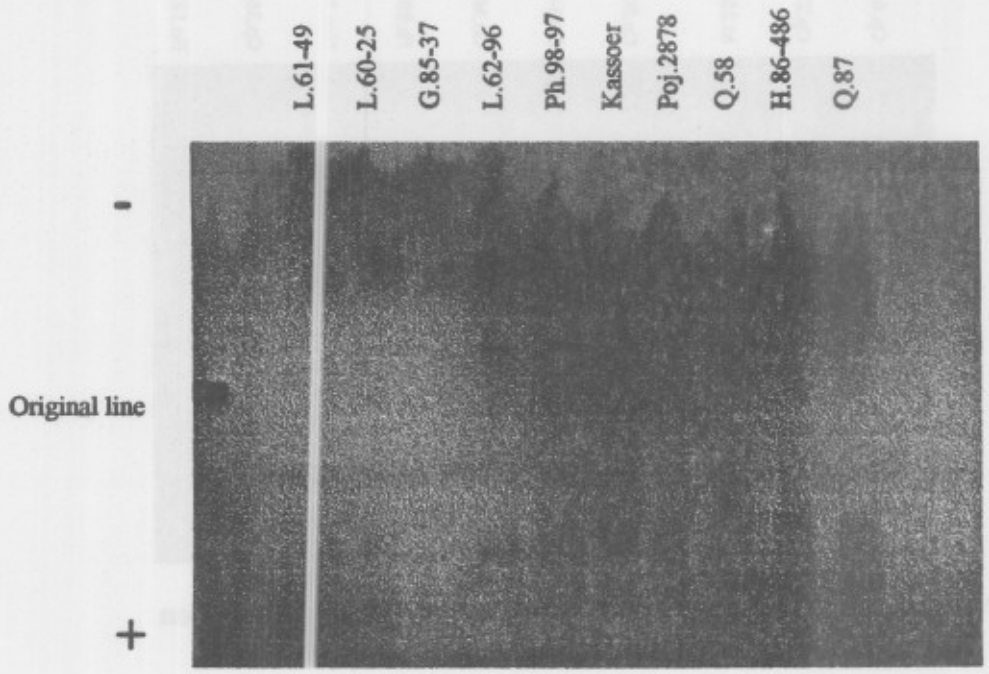

Figure (5): Electrophoretic patterns of peroxidase isozymes for ten sugar cane varieties (31-40).

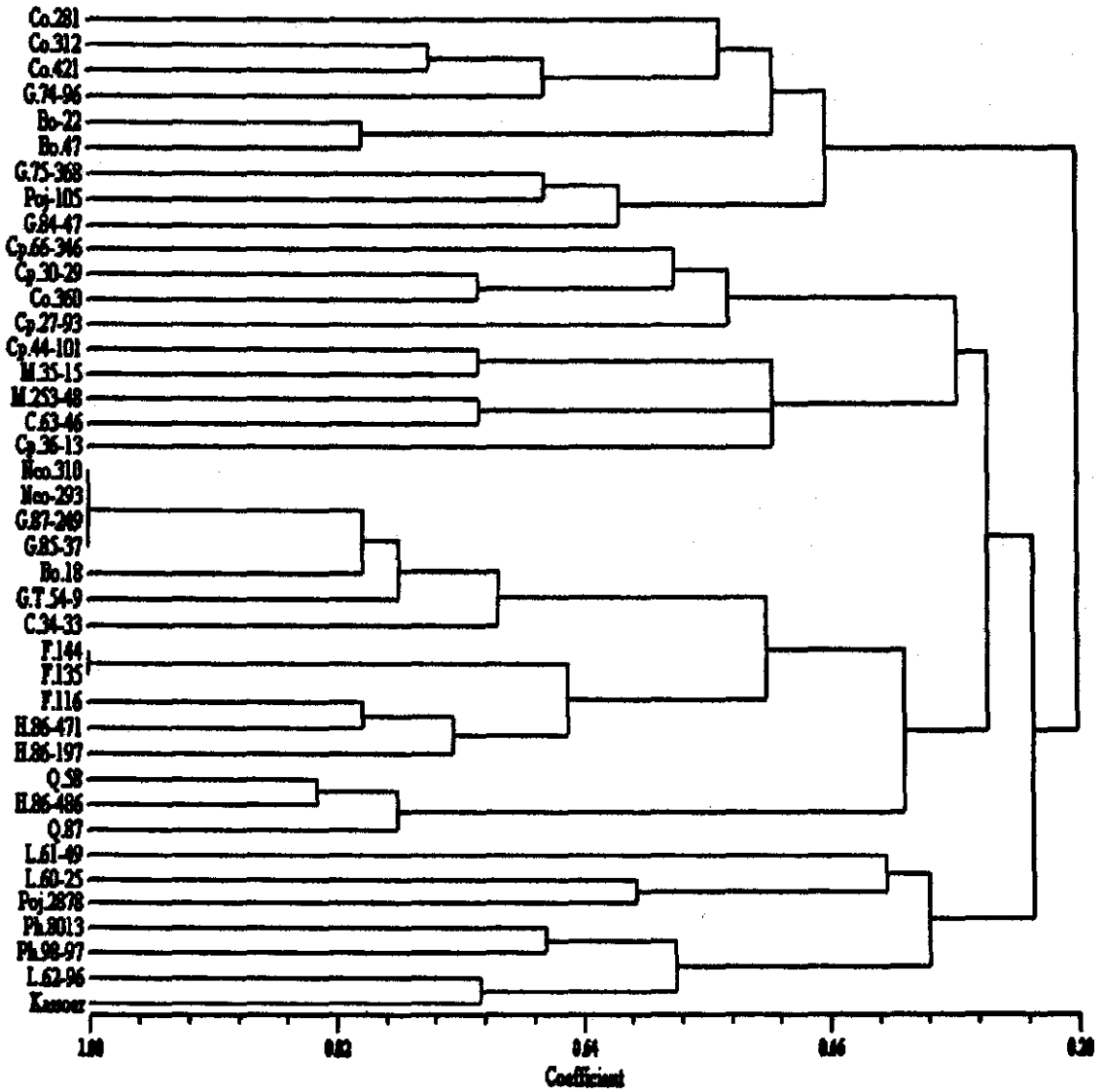

Figure (6): Dendrogram of forty sugar cane varieties based on (0-1) data of isoperoxidase activity.

parameters were found to be different among the forty studied varieties. Band No, 5, in the cathodal side, existed in all studied varieties. Every variety contained five to three bands in the cathode. These data are in good agreement with those reported by Glaszmann *et al* (1989). Table (6) shows (0 and 1) data, whereas (0) refers to absence of band, while (1) refers to band presence.

Figure (6) shows the dendrogram of cluster analysis based on (0 and 1) data employing the NTSYS-pc ver. 2.1 software.

The figure shows that there were four clusters in the dendrogram tree. The analysis was capable to classify the studied forty varieties into four main clusters; cluster No.1 contained nine varieties (Co.281, Co.312, Co.421, G.74-96, Bo.22, Bo.47, G.75-368, Poj.105 and G.84-47). Cluster No.2 contained nine varieties in two sub-clusters (Cp.66-346, Cp.30-29, Co.360, Cp.27-93, Cp.44-101, M.35-15, M.253-48, C.63-46 and Cp.36-13). While, cluster No.3 contained fifteen varieties in three sub-clusters (Nco.310, Nco.293, G.87-249, G.85-37, Bo.18, G.T.54-9, C.34-33, F.144, F.135, F.116, H.86-471, H.86-197, Q.58, H.86-486 and Q.87). Seven varieties were found in the last cluster. They were L.16-49, L.60-25, Poj.2878, Ph.8013, Ph.98-97, L.62-96 and Kassoer.

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الملخص العربي

أستخدام بعض الصفات الكمية و المشابهات الأنزيمية لأنزيم البيروكسيديز في تحديد التباعد الوراثي لأربعين صنفا من قصب السكر

مجدي سعد صالح و نبويه صالح عبده غره و محمود أحمد المنحلي

قسم التربية والوراثة – معهد بحوث المحاصيل السكرية

أجريت هذه التجربة في محطه البحوث الزراعية بالصبحية بالأسكندرية. والغرض الأساسي لاجراء هذا البحث هو دراسة التباعد الوراثي لأربعين صنفا من أصناف قصب السكر باستخدام المشابهات الأنزيمية لأنزيم البيروكسيدز، بالأضافة إلى أستخدام سبع صفات كمية من الصفات المورفولوجية والمحصولية والصناعية لقصب السكر، وهي:-

١- وزن العود.

٢- طول العود.

٣- سمك العود.

٤- عدد السلاميات.

٥- نسبه المواد الصلبه الذائبة الكلية.

٦- نسبه السكروز.

٧- درجه النقاوة.

وقد تم اجراء هذه التجربة بمزرعة محطة البحوث الزراعية بالصبحية في عامي ٢٠٠٢و٢٠٠٣. وقد أظهرت النتائج المتحصل عليها وجود فروق معنوية عالية بين جميع الأصناف في جميع الصفات تحت الدراسة. وفي تحليل الشجرة – سواء المشابهات الأنزيمية أو السبع صفات الكمية قيد الدراسة – فقد وضع هذا التحليل الأربعين صنفا تحت الدراسة في مجاميع مختلفة، مما يعني أنه عند أختيار الآباء لأجراء التهجينات في برامج التربية يمكن أختيار الأباء التي تحمل الصفات الجيدة سواء كانت صفات محصولية أو تصنيعية والتي تنتمي لمجاميع مختلفة في الشجرة وذلك للحصول علي قوة أعلي للهجين .