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Population study of *Miscanthus floridulus* III. Population variation of *M. floridulus* in Green and Orchid Islets of Taiwan

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Abstract. *Miscanthus floridulus*, one of dominant grasses, is widely distributed in Taiwan as well as in the Green Islet and Orchid Islet. Along the coastal area of the islets, leaves of *Miscanthus floridulus* collected from 20 populations were analyzed for peroxidase and esterase by using polyacrylamide gel electrophoresis. Results of analyses showed that isozyme patterns are significantly different among 20 populations. The data of isozymes present are computed by the formula of Euclidean distance (hereafter called population distance) and the simple matching coefficients, resulting in dendrograms of populations. Three clusters were evidently found among the seven populations in the Green Islet, and four clusters in the populations of the Orchid Islet. Combining the populations of both islets, the populations could form four major clusters. The first cluster includes populations of Tapaisa, Kuomin hotel, Yeiin, Yungsing farm, and Orchird Islet airport. The second cluster includes populations Haishanpin, Tsaoshan, Green Islet airport, Yeiyou, and Fusing farm. The third cluster involve populations Chingshuishan, Chungsing farm, and Swanshuyen. And the fourth cluster include populations Rontao, Wannanjao, and Shuitaokao. In conclusion, the populations of *Miscanthus floridulus* in both islets have differentiated into four major clusters, resulting in ecotype formation. Further evidence showed that the populations of *Miscanthus floridulus* in both islets are discontinuous rather than continuous. The mechanism of discontinuity needs to be further studied.

Key words: Cluster; Esterase; *Miscanthus floridulus*; Population; Peroxidase.

Introduction

In recent years, isoenzyme study has received increasingly attention by geneticist to study the genes involved in enzyme synthesis, by biochemists and physiologists to look upon the different physico-chemical properties of isoenzymes as means to study the regulation of cell metabolism, and by evolutionary ecologists to study the existence of isoenzymes to enhance the biochemical adaptability of organism and to protect it against loss of function occasioned by mutation or environmental stresses (Shannon, 1968; Gottlieb, 1982; Schmitz and Kowallik, 1986). Isoenzyme studies in a variety of plants were carried out and tremendous growth of information appeared in literatures (Nei, 1965, 1973; Johnson, 1977; Kiang and Wu, 1979; Loukas *et al*., 1983; Silander, 1984). For example, Wu and Bradshaw (1972) studied the copper tolerance of *Agrostis tenuis* and *Festuca ovina* by using esterase isozyme as parameter and found that the isozyme patterns were significantly different among five grass populations where the soils were polluted by copper factory established in different year and from area without copper pollution. Chou *et al.* (1984a, 1984b, 1985, 1986) employed different isozymes to study the phylogenetic relationship among bamboo species, and Hsiao (1980) also used isozymes to study the chemotaxonomic relationship among taxa of *Chamecyparis*. They concluded that isozymes play a significant role in clarifying the taxanomic and phylogenetic position of plants studied.

Miscanthus floridulus (Labill.) Warb., a dominant endemic grass, is ubiquitously distributed in areas below 2000 m in elevation in Taiwan, and possesses an allelopathic potential forming a relatively pure stand in fields (Chou and Chung, 1974). On the other hand, *M. transmorrisonensis,* another dominant species, is generally distributed in mountainous area at the elevation above 2200 m. In the first year study, we concluded that four major clusters were found among 27 populations of *M. floridulus* in various habitats (Chou *et al.,* 1987). It was shown that the *Miscanthus* exhibited a wide heterogeneity, resulting in a wide adaptability to different environmental regimes, such as high salinity, polluted soil, and severe dry land. Furthermore, Chou and Chang (1988) also reported that the ecotypical variation of *Miscanthus* populations along the roadside of Tapingting from 1000 m to 1600 m in elevation. Three clusters of the *Miscanthus floridulus* were found and the species was mixed with *M. transmorrisonensis* at the elevation of 2200 m. Above 2200 meters there is also a pure stand of *M. transmorrisonensis.* The adaptation is due presumably to the regulation of isoenzyme polymorphism. The genetic study of polymorphism in *Miscanthus* is beyond present capacity. Hsu (1986) found that the wide distribution of *Miscanthus floridulus* is due in part to seed germination insensitive to temperature. Weng (1986, personal communication) showed that the compensation point for carbon dioxide of *M. floridulus* varied with habitats and their phenotypic characters among populations are hardly distinguished.

Moreover, *Miscanthus floridulus* is also widely distributed in two Islets of Orchid and Green, about 50 miles off the east coast of Taiwan proper. Based on the characteristic of islets, the heterogeneity and ecotypical variation of *Miscanthus floridulus* populations are thought to be different from the island proper. The findings of population study of *Miscanthus floridulus* in the two islets were therefore reported.

Materials and Methods

Materials

Leaves of *Miscanthus floridulus* were collected along the roadsides of seven populations of Green Islet and that of thirteen populations of Orchid Islet in May 1989. The locations of each sampling site in a radius of 100 m designated as a population are given in Fig. 1. The description of sampling locations of *Miscanthus* populations is also described in Table 1. The sampling techniques were the same as those described by Chou *et al.* (1987). The chmicals and solvents for electrophoresis were purchased either from the Sigma Corp. (USA) or from Merck Ltd. (West Germany).

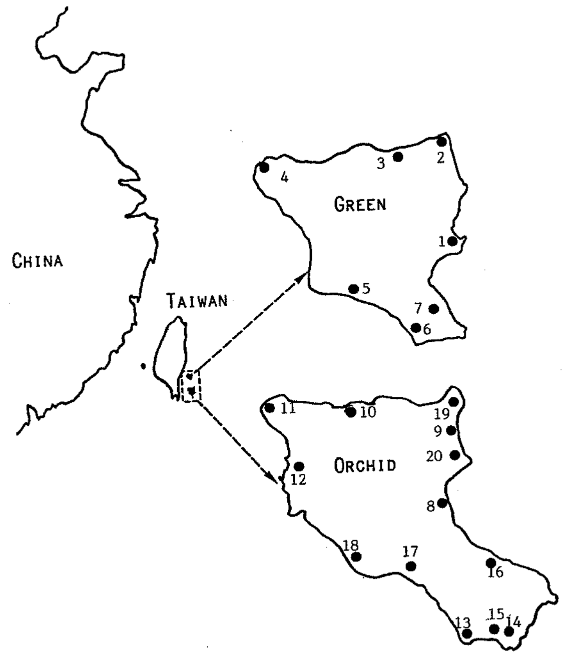

Fig. 1. The locations of sampling in Green Islet and Orchid Islet at the east of Taiwan proper.

Electrophoresis Analysis

A vertical gel electrophoresis (M & S Slab Electrophoresis, model SG-80) was employed and techniques for electrophoresis of *Miscanthus* leaves were described by Chou *et al.* (1984a). Peroxidase and esterase isozymes were selected for the study. After the analysis of electrophoresis, the gel was dried and a permanent sheet of zymogram was obtained. Accordingly, the R_f value of each band present in the zymograms was caculated and the frequency of each band in each population was obtained.

Statistical Analyses

In order to understand the ecotypic variation

Table 1. *Description of sampling site of 20 populations along the coastal highway of Green and Orchid Islet*

	Population number	Location	Sample repetition
Green Islet	1	Haishanpi (海 參 坪)	20
	2	Liumakao (流 麻 溝)	20
	3	Tsaoshan (草 山)	20
	4	Green islet airport (機 場)	5
	5	Kueiwanpi (龜 灣 鼻)	20
	6	Tapaisa (大 白 沙)	19
	7	Kuomin Hotel (國民旅社)	20
Orchild Islet	8	Yeiin (野 銀)	20
	9	Chingshuishan (清 水 山)	20
	10	Rontao (朗 島)	20
	11	Chingputzupi (親不知鼻)	20
	12	Yeiyou school (椰油國小)	20
	13	Fusing farm (復興農莊)	20
	14	Wannanjzo (望 南 角)	20
	15	Shuitaokao (四 道 溝)	20
	16	Yungsing farm (永興農莊)	20
	17	Chungsing farm (中興農莊)	20
	18	Orchid islet airport (機 場)	20
	19	Shwanshe yen (雙 獅 岩)	20
	20	Tungtsing villege (東 清 村)	20

among populations of *Miscanthus,* two mathematic equations, such as a simple matching coefficient (Ssm), and Euclidean distance (d_{jk}) hereafter we called *population distance*, was used (Sneath and Sokal, 1973). The formula of similarity coefficient, $Ssm = m/m+u$ was used. Where m is the number of matches, and u the number of mismatches. The data of Ssm were then set in a simple matrix table using each population as an operational populational unit (OPU's). The second formula called *population distance,* d_{jk} between OPU's j and k is

$$d_{jk} = [\sum_{i=1}^{n}(X_{ij}-X_{ik})^2]^{1/2}$$

Where X_{ij} is the average frequency of i band in the zymogram of j population and X_{ik} is the average frequency of i band in the zymogram of k population. By using these equations the data were computed to obtain a matrix for each isozyme and clustering analysis between populations was obtained by an unweighted pair-group method using simple arithmetic average described by Sneath and Sokal (1973); thus, a dendrogram was obtained.

Results

Distribution and Frequency of Peroxidase Isozymes in ***Miscanthus*** *Populations*

The zymogram of peroxidase present in 20 popula-

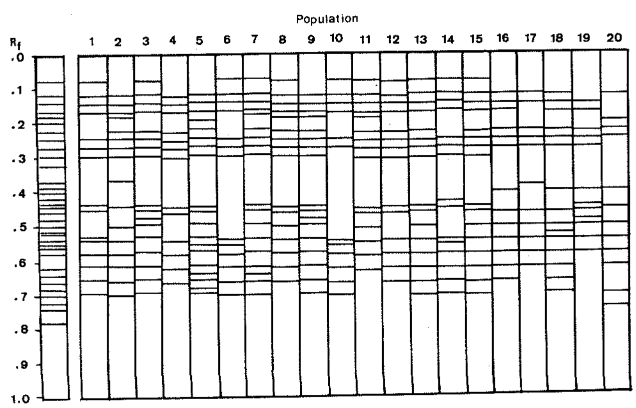

Fig. 2. The zymogram patterns of peroxidase in leaves of *Miscanthus* from 20 populations of Green and Orchid Islets. Description of population number see Table 1.

tions of *Miscanthus* is given in Fig. 2, which shows 33 bands in 20 population studied. The Rf value and frequency of each band present in the peroxidase are also given in Table 2. Of 33 bands found in peroxidase, bands 2, 3, 4, 8, 9, 16, 17, 22, 25, 26, and 28 are predominant and ubiquitous distributed in most of populations. Indeed, in Orchid Islet, bands 2, 3, 4, 17, 19, 22 and 26 exhibited 100% presence (frequency is 1.00) in most of populations, particularly in population 17 (Chung Sing farm). The remaining bands of 5, 6, 11, 12, 13, 14, 20, 21, 23, 24, 27, 29, 31, 32, and 33 are distributed in low frequency and scattered in these populations. In addition, bands 21, and 34 are missing from the Green Islet, while bands 11, 14, 20, 27, 31, and 33 are missing from Orchid Islet. A few bands, no. 12, 13, 14, 20, 21, 24, 31, 32, 33, are only found in some populations, such as populations

Table 2. *The frequency of each band in peroxidase isozymes present in 20 populations along the coastal highway of Green and Orchid Islets*

Band No.	Rf.	Population																			
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
1	0.076	0.13		0.63			0.27	0.50	0.60		0.40	0.40	0.30	0.65	0.41	0.95					
2	0.118	0.62	0.40	0.84	0.71	0.45	0.70	0.50	0.40	0.90	0.65	0.60	0.70	0.60	0.59	0.90	0.60	1.00	0.65		0.35
3	0.142	0.48	0.61	0.79	0.86	1.00	0.75	0.65	0.95	0.85	0.95	0.95	0.75	1.00	0.59	1.00	0.50	1.00	0.80	1.00	
4	0.165	0.57	0.43	0.68	0.86	0.60	0.60	0.35	0.85	0.80	1.00	1.00	0.75	0.70	1.00	0.90	0.60		0.50	1.00	
5	0.181		0.13					0.25		0.45		0.20									
6	0.198		0.17			0.45															0.65
7	0.223	0.30	0.58	0.47	0.43	0.40	0.13	0.20	0.15	0.30		0.10	0.30	0.30		0.35	0.30	0.05	0.70	0.50	1.00
8	0.244	0.53	0.48	0.89	0.57	0.50	0.85	0.40	0.55	0.75	0.55	0.20	0.50	0.65	0.59	0.65	0.60	0.70	0.65	0.50	0.95
9	0.268	0.70	0.45	0.89	0.71	0.80	0.85	0.60	0.70	0.70	0.70	0.85	0.55	0.95	0.71	0.80	0.55	0.60	0.75	0.45	
10	0.292	0.43		0.84	1.00	0.75	0.35	0.90	0.75	0.75		0.70	0.90	0.75	0.82	0.50					
11	0.364	0.43	0.40																		
12	0.383	0.45					0.30											0.15			
13	0.396		0.34																0.40	0.05	0.40
14	0.414		0.13																		
15	0.428	0.43	0.23									0.35			0.24	0.40					
16	0.437	0.22	0.50	0.26	0.43	0.15	0.20	0.35	0.10	0.35			0.15	0.30	0.06					0.05	
17	0.452	0.65	0.50	0.26	0.29	0.20	0.50	0.15	0.10	0.30		0.10	0.15	0.25		0.30	1.00	1.00	1.00	1.00	1.00
18	0.478	0.10	0.08	0.11						0.20										0.05	
19	0.492	0.43	0.35	0.11		0.45	0.50	0.05	0.30	0.20				0.35		0.20	1.00	0.95	1.00	1.00	0.85
20	0.509		0.13																		
21	0.518																		0.30		
22	0.532	0.83	0.38	0.95	0.86	1.00	0.83	0.95	0.90	0.95										1.00	0.90
23	0.548	0.65					0.10				0.85	0.65	0.55	0.95	0.94	0.95	0.90	1.00	0.45		
24	0.558					0.60					0.30				0.18						
25	0.573	0.63	0.70	1.00	0.86	0.85	0.35	0.80	0.90	0.85	0.90				0.94					0.95	0.85
26	0.614	0.35	0.75	0.79	0.43	0.65	0.25	0.75	0.60	0.65		0.60	0.60	1.00		0.95	0.90	0.70	0.80		0.15
27	0.638		0.34					0.30													
28	0.652	0.50	0.30	0.32	0.29	0.55	0.30	0.60	0.20	0.35		0.45	0.20	0.85	0.94	0.75	0.05	0.50	0.55		
29	0.677		0.50			0.50															
30	0.692	0.50	0.23	0.11		0.45	0.15	0.35		0.10	0.35	0.30	0.20	0.65	0.41	0.55	0.20		0.90		1.00
31	0.718		0.24																		
32	0.732		0.24								0.10			0.25	0.06	0.25			0.70		0.90
33	0.774		0.02																		

Liumakuo, Yungsing farm, and Chungsing farm (Table 2).

Distribution and Frequency of Esterase Isozymes in ***Miscanthus*** *Populations*

The zymogram of esterase in populations of *Miscanthus* is given if Fig. 3, and the frequency of 33 bands respectively present in the zymogram is shown in Table 3. Of them, bands 6, 7, 8, 9, 11, 12, 21, 22, 23, 26, 27, 28, 29, 30, 31, 32, and 33 are commonly present in all populations and the frequency of each band in the population is exceedingly high, while the remaining bands are randomly distributed in populations studied. However, bands 18 and 19 are only present in population Rontao, while band 17 is only found in Shuitaukuo.

The Variation of Population Distance Based on Isozymes in ***Miscanthus***

The major objective of this study was ajmed to find out the population variation of *Miscanthus floridulus* in two islets, Green Islet and Ochird Islet. By means of population distance, d_{jk}, was used to answer the *population distance* among 20 populations of *Miscanthus* based on the peroxidase analysis (data of matrix are not present), from which a dendrogram was derived. The lower the values of d_{jk}, the shorter the population distance the closer between two populations.

Based on the peroxidase analysis of the *Miscanthus* populations, the populations can therefore be grouped into several clusters (Fig. 4). Populations 3 (Tasoshan), 13 (Fusing farm), 8 (Yeiin) and 7 (Kuomin hotel) are one cluster; populations 4, 9, 11, and 12 form another cluster. These two clusters acturally are very close at population distance of 1.054. Another clusters include populations 16 (Yungsing), 17 (Chungsing), 18 (Orchid airport), and 19 (Shwansheyen). Two sets of populations 10 and 14, and 1 and 6 are obviously close together at d_{jk} below 1.335. Never-

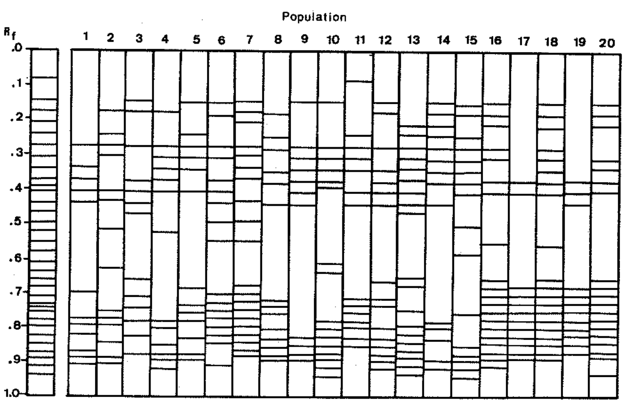

Fig. 3. The zymogram patterns of esterase in leaves of 20 populations of *Miscanthus* in Green and Orchid Islets at the eastern Taiwan. The designation of population number see Table 1.

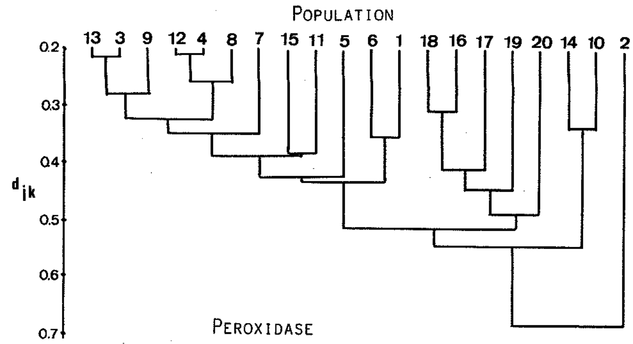

Fig. 4. The dendrogram of 20 populations of *Miscanthus* in both Islets of Green and Orchid, Taiwan based on the frequency of peroxidase isozyme present and derived from the formula of *population distance* (d_{jk}).

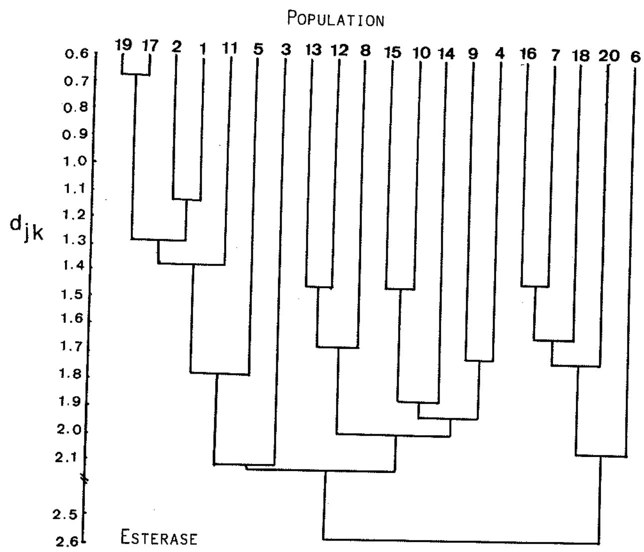

Fig. 5. The dendrogram of 20 populations of *Miscanthus* in both Islets of Green and Orchid, Taiwan based on the frequency of esterase isozyme present and computed by the formula of *population distance.*

theless, population 20 seems to be far from the remaining populations.

Regarding the data of esterase analysis, three major clusters were derived from the data of population distance. The first cluster involves population 1 (Haishanpin), 2 (Liumakuo), 11 (Chingbutzupi), 17 (Chungsing), and 19 (Swansheyen); the second cluster consists of two subclusters, namely, populations 8 (Yeiin), 12 (Yeiyou), 13 (Fusing), which are also linked closely with another subcluster of population 4 (Green island airport), 9 (Chingshue shan), 10 (Rontao), 14 (Wannanchao), and 15 (Shutaokuo); the third cluster therefore consists of populations 6 (Tapaisa), 7 (Komin hotel), 16, 18, and 20 (Tung ching villege). The third cluster is ecologically far from the other two clusters shown (Fig. 5).

Moreover, the data combined from the peroxidase and esterase were also run for statistical analysis using the formula, *population distance* from which a dendrogram was derived (Fig. 6). It was noticeable that three major clusters of *Miscanthus floridulus* populations were found. For example, populations 1, 2, 5, 9, 11, 17 and 19 form a cluster; populations 6, 7, 8, 16, and 18 form the second cluster; and populations 3, 4, 10, 12, 13, 14, and 15 result in a third cluster. It is interesting to note that population 20 (Tungching villege) is far from the above mentioned populations.

The Variation of Similarity Among Populations of ***Miscanthus***

By using a simple matching coefficient, a matrix of similarity based on peroxidase of isozymes among 20 populations was obtained (matrix data are not present here) and derived into a dendrogram shown in Fig. 7. In overall, the similarity coefficient between populations studied is generally above Ssm 0.566, indicating that these populations are quite similar. If we take the similarity coefficient above 0.824, the populations can be grouped into three clusters. The populations 3, 4, 7, 8, 9, and 13 form a cluster; populations 1, 5, 6, 11, and 15 form the second cluster; and populations 10, 14, 16, 17, 18, and 19 form the third cluster.

Regarding the similarity coefficient based on the esterase analysis of 20 populations mentioned, the overall Ssm is above 0.530, indicating that the aforementioned populations exhibited high similarity. If we take the Ssm above 0.660, these populations could be

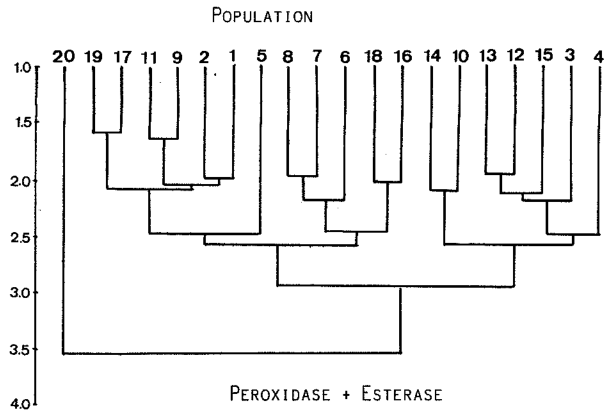

Fig. 6. The dendrogram of 20 populations of *Miscanthus* in both Islets of Green and Orchid, Taiwan based on the frequency of peroxidase and esterase present and computed by the formula of *population distance.*

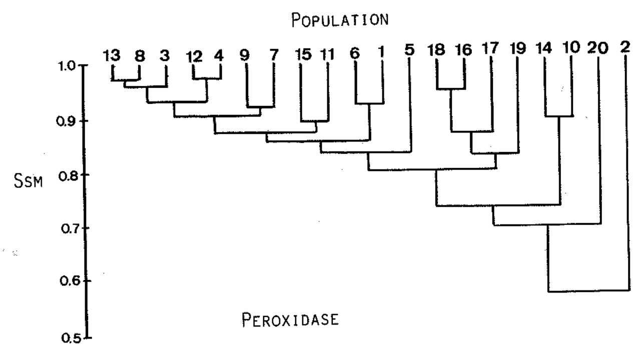

Fig. 7. The dendrogram of 20 populations of *Miscanthus* in both Islets of Green and Orchid, Taiwan based on data of presence of peroxidase and computed by the formula of simple matching coefficient, Ssm.

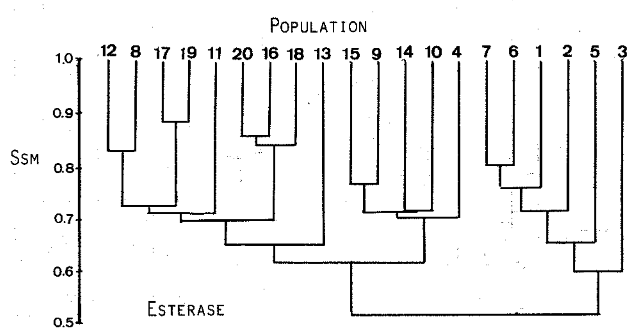

Fig. 8. The dendrogram of 20 population of *Miscanthus* in both Islets of Green and Orchid, Taiwan based on the data of presence of esterase and computed by the formula of Ssm.

Table 3. *The frequency of each band in esterase isozymes present in 20 populations along the coastal highway of Green and Orchid Islets*

Band No.	Rf.	Population 1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
1	0.085											0.25									
2	0.148			0.25		0.30	1.00	0.85		0.40	0.40		0.45		0.40	0.35	0.85		1.00		1.00
3	0.179		0.21	0.30			1.00	0.85	0.20				0.25		0.40	0.65	0.85		0.65		1.00
4	0.213				0.40			0.85						0.11	0.20		0.85		0.45		1.00
5	0.245		0.13			0.30			0.70			0.45		0.26	0.20	0.45					
6	0.278	0.33	0.18	0.40	0.60	0.80	1.00	1.00	0.75	0.65	0.70	0.75	0.15	0.74	0.60	0.55	0.60		0.40		
7	0.310	0.08	0.25		0.60	0.50	0.90	0.75		0.40	0.75	0.45		0.26	0.50	0.65	0.60		0.40		0.35
8	0.342	0.30	0.02		1.00	0.60		0.50	0.25	0.80	0.55	0.30	0.20	0.16	0.90	0.15			0.40		0.55
9	0.372	0.30		0.50	0.60		0.10	0.70	0.30	0.25	0.30		0.60	0.16	0.70	0.05	0.40	0.15	0.45	0.10	0.90
10	0.392										0.15							0.15	0.40		
11	0.406	0.35	0.37	0.75	0.60	0.30	1.00			0.25		0.10	0.85	0.68		0.10	0.70			0.20	0.95
12	0.436	0.20	0.32	0.65			0.90	0.40	0.80	0.85		0.40	0.45	0.79	0.60					0.10	
13	0.464		0.02	0.20										0.26							
14	0.494		0.02				0.50	0.70								0.05					
15	0.521		0.50		0.40																
16	0.549		0.05				0.35	0.45									0.75		0.25		
17	0.577															0.05					
18	0.608										0.45										
19	0.632		0.13								0.45										
20	0.654	0.23	0.10	0.40									0.35	0.26			0.20		0.35		0.50
21	0.675	0.43	0.10			0.50		0.50						0.32			0.45	0.10	0.30	0.30	0.60
22	0.703	0.50	0.35	0.70			0.30	0.50	0.20			0.40	0.40				0.65	0.75	0.60	0.65	0.80
23	0.725	0.48	0.35			0.70	0.50	0.60	0.25			0.45	0.40				0.70	0.75	0.70	0.75	0.95
24	0.735			0.90																	
25	0.747	0.50	0.70			0.60	0.60	0.70	0.45			0.85		0.47			1.00	1.00	0.95	0.85	0.95
26	0.772	0.85	0.85	0.40	1.00	0.60	1.00	1.00		0.60	0.05	0.95			0.70		0.95	0.80	0.90	1.00	1.00
27	0.791	0.85	0.85		0.80		0.85	1.00	0.75		0.35	0.85	0.25	0.47	0.90		0.95	0.70		0.85	0.95
28	0.821	0.73	0.35	0.45		0.90	0.75	0.75	0.85		0.70	0.70	0.65	0.58	0.80		0.80	0.60	0.75	0.75	0.90
29	0.842	0.35	0.80	0.30	0.60		0.40	0.55	0.95	0.70	0.65	0.30	0.75	0.89		0.30	0.75	0.30	0.50	0.75	0.40
30	0.867	0.60			0.80	0.60		0.35	0.80	0.85	0.65		0.75	0.89		0.30	0.25	0.25	0.25	0.55	0.40
31	0.882	0.58	0.27		0.40	0.30		0.05	0.65	0.85	0.90		1.00	0.95	0.80	0.70	0.25	0.05	0.10		0.15
32	0.906	0.60	0.50		0.60		0.10				1.00		0.60	1.00	0.90	0.55	0.20				
33	0.930										0.40			0.21		0.20					0.25

grouped into three major clusters. Thus, populations 8, 11, 12, 13, 16, 17, 18, 19, and 20 belong to one cluster; populations 4, 9, 10, 14, and 15 are the second cluster; and populations 1, 2, 3, 5, 6, and 7 are the third clusters, indicated that the populations in the Green Islet are evidently different from that of the Orchid Islet (Fig. 8).

Furthermore, when the data of peroxidase and esterase zymogram patterns were combined for similarity analysis, results of analysis showed that the patterns of population clusters are similar to those described above (Fig. 9). Furthermore, the patterns of similarity of the *Miscanthus* population agreed mostly with those of population distance. The closer the populations distance the higher similarity within the populations. Nevertheless, the similarity or population dis-

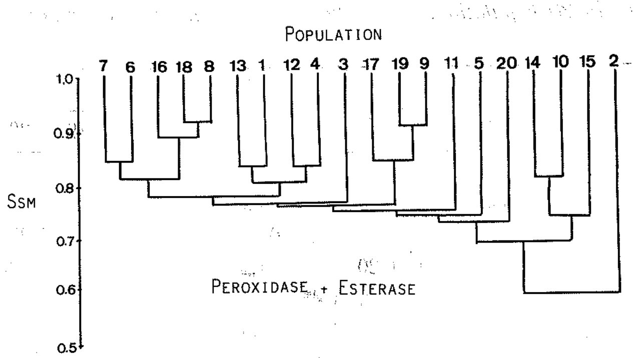

Fig. 9. The dendrogram of 20 populations of *Miscanthus* in both Islets of Green and Orchid, Taiwan based on the data of presence of peroxidase and esterase and computed by the formula of Ssm.

tance among populations was unlikely continuous that evidence would be described in the next paragraph.

Population Discontinuity of ***Miscanthus floridulus***

If we examine two populations within one cluster, for example, populations 10 and 14, it can obviously be seen that these two populations are not adjacent to each other but rather separate in quite distance. In other words, the populations within a cluster are often discontinuous instead of continuous (Fig. 10). If two populations forming a cluster are considered a population pair based on the similarity and population distance (Ssm > 0.75 for both peroxidase and esterase; $d_{jk} < 0.4$ for peroxidase and $d_{jk} < 1.8$ for esterase), the population pairs are thus as follows: populations 10 and

Table 4. *Population pairs of* ***Miscanthus floridulus*** *in Green and Orchid Islets based on similarity and population distance of esterase (E) and peroxidase (P) analyses*

Population Pair	Similarity (Ssm)			Population distance (d_{jk})			Sum of presence of (+)
	E	P	P+E	E	P	P+E	
19,17	+		+	+	+	+	5
20,16,18	+	+				+	3
15, 9	+						1
14,10	+	+	+	+	+	+	6
7, 6, 1	+	+	+				3
13, 8		+		+	+		3
12, 4		+	+		+		3
9, 7		+					1
15,11		+					1
18, 8,16			+	+	+		3
13, 1			+				1
2, 1				+		+	2
12,13				+		+	2
15,10				+			1
9, 4				+	+		2
16, 7				+			1
13, 3					+		1
6, 1					+		1
19,16					+		1
8, 7						+	1

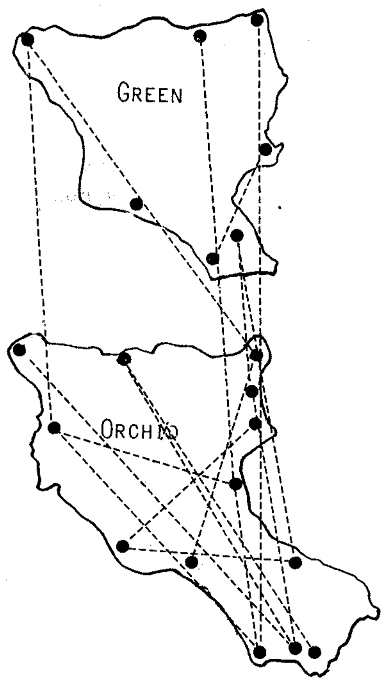

Fig. 10. A correlation of population similarity and population distance of *Miscanthus floridulus* in both Islets of Green and Orchid. The broken line expresses the highest similarity and the lowest population distance between two populations that form a cluster.

14, populations 12 and 8, populations 16 and 18, populations 19 and 17, and many other pairs (Table 4), clearly demonstrated their population discontinuity. These two populations are not located adjacently but rather located across the islets, or located in an opposite direction (Fig. 10). The phenomenon of population discontinuity is more pronounced in the Orchid Islet than in Green Islet. As we know, both islets were formed by volcanic deposition and their geological and edaphic conditions are not significantly different. In other words, the habitats of both islets for *Miscanthus floridulus* have not much difference. The elucidation of this population discontinuity is thus needed to be further investigated.

Discussion

In a series population study of *Miscanthus floridulus,* we reported that the ecotypes of *Miscanthus* were found in various industrial area in Taoyuan county, west part of Taiwan plain, and in mountainous area along the Shenmonlingtao of Nantou county (Chou *et al.*, 1987; Chou and Chang, 1988). The coastal area of Taoyuan county has been nortorious for the industrial pollution for nearly twenty years, and vegetations, such as *Casuaria glauca, Pandanus odoratissimus,* and *Ipomoea* spp., which are salt tolerant species, were severely injuried by air pollutants (Chang and Tang, 1975). Nevertheless, *Miscanthus floridulus* exists healthily, indicating that adaptive mechanism has been developed. Our present data show that the isozyme patterns based on peroxidase and esterase present in leaves of *Miscanthus* could explain the reason for such adaptation. Secondly, the geographic distribution of *Miscanthus floridulus* may not be so important as far as the ecotypic variation is concerned, because we found populations Linco and Taitung L are forming a cluster based on population distance of peroxidase (in Fig. 3 of Chou *et al.*, 1987). These two populations are situated in the east and west coast of the Islet. However, 3 populations of Tapinting and 2 populations of Yushan were found close intraspecifically.

Chou (1989) reported that the population divergence of *Miscanthus floridulus* was significantly different among *Miscanthus* populations in island Taiwan and Islets of Pescardero, Green, and Orchid. This probably attributes to the islet isolation without intraspecific hybridization. However, the geographic effect on *Miscanthus floridulus* differentiation was also obvious, resulting in preservation of genetic characters. For example, the zymogram pattern of Yushan population (2750 m in elevation) was quite similar to that of Green Islet (Chou, 1989). In the present study, we found that the overall similarity coefficient of 20 populations based on the peroxidase and esterase analyses was quite high ($Ssm > 0.60$), indicating that the high similarity was due primarily to the long isolation of islets. Thus, we proposed that the origin of *Miscanthus floridulus* in populations of Yushan and Islets of Green and Orchid could possibly be the same. Molecular basis of polymorphism in *Miscanthus* populations should therefore be further studied.

One may also question on the effect of climate on the isozyme patterns of *Miscanthus.* We found that results of seasonal variation of isozyme patterns were insignificant (Chou and Chang, 1988). However, the effect of altitude on the isozyme variation is significant and is presumably related to the effect of temperature. We have conducted experiments on the effects of temperatures on the variation of isozymes present in *Mis-*

canthus floridulus under phytotron and field conditions. Preliminary results of the study showed that the zymogram patterns are significantly different between temperature treatments and the experiment is being repeated (Chou *et al.,* 1989 unpublished data).

Based on the individualistic concept of plant community (Gleason, 1926), McIntosh (1967) pointed out that plant communities are mostly continuous. Whittaker (1967) also concluded that the vegetation change is always according to environmental gradient, resulting in a gradient distribution of plant communities. Whittaker's hypothesis was indeed elucidated from Gleason's hypothesis. According to our present findings based on the two isozyme analyses, it is obvious that the discontinuity of *Miscanthus floridulus* populations in both Islets of Green and Orchid is contradictory to continuum hypothesis. However, the present findings can not clarify the point of population discontinuity based on the phenotypic variation of *Miscanthus.* Nevertheless, our findings could strongly favor Gleason's individualistism.

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Literature Cited

Chang, C. H. and C. R. Tang. 1975. Effects of air pollution on the growth of forest vegetation. National Chung Hsing University Monograph 136, Taiwan.

Chou, C. H. 1989. Plant molecular ecology: a population study of *Miscanthus floridulus* and *M. transmorrisonensis. In* Chen, C. S. and C. Tu, (eds.), Plant Molecular Biology, Institute of Botany, Academia Sinica Monograph Series No. 8, Taipei, Taiwan. pp. 103-113.

Chou, C. H. and F. C. Chang. 1988. Population study of *Miscanthus floridulus* II. Ecotypic variation of *M. floridulus* and *M. transmorrisonensis* as affected by altitude in Nantou, Taiwan. Bot. Bull. Academia Sinica **29:** 301-314,

Chou, C. H. and Y. T. Chung. 1974. Allelopathic potentical of *Miscanthus floridulus.* Bot. Bull. Academia Sinica **15:** 14-27.

Chou, C. H. and Y. H. Hwang. 1985. A biochemical aspect of phylogentic study of Bambusaceae in Taiwan III. The genera *Arthrostylidium, Chimonobamtusa,* and *Dendrocalamus.* Bot. Bull. Academia Sinica **26:** 155-170.

Chou, C. H., S. Y. Hwang, and F. C. Chang. 1987. Population study of *Miscanthus floridulus* (Labill.) Warb. I. Variation of peroxidase and esterase in 27 populations in Taiwan. Bot. Bull. Academia Sinica **28:** 247-281.

Chou, C. H., Y. H. Hwang, and S. Y. Hwang. 1986. A biochemical aspect of phylogenetic study of Bambusaceae in Taiwan. IV. The genera *Arundinaria, Pseudosasa, Semiarundinaria, Shibataea, Sinobambusa* and *Yushania.* Bot. Bull. Academia Sinica **27:** 117-130.

Chou, C. H., S. S. Sheen, and Y. H. Hwang. 1984a. A biochemical aspect of phylogenetic study of Bambusaceae in Taiwan II. The genus *Bambusa.* Bot. Bull. Academia Sinica **25:** 177-189.

Chou, C. H., C. M. Yang, and S. S. Sheen. 1984b. A biochemical aspect of phylogenetic study of Bambusaceae in Taiwan I. The genus *Phylostachys.* Proc. Nat. Sci. Counc., ROC (B) **8:** 89-98.

Gleason, H. A. 1926. The individualistic concept of the plant association. Bull. Torrey Bot. Club **53:** 7-26.

Gottlieb, J. Y. 1982. Conservation and duplication of isozymes in plants. Science **216:** 373-380.

Hsiao, J. Y. 1980. A biochemical systematic study of the genus *Chanaecyparis* in Taiwan. Proc. Nat. Sci. Counc., ROC **4(1):** 69-77.

Hsu, F. S. 1986. Germination of native forage species with salt, drought, and cold to tolerance in Taiwan. (manuscript in preparation).

Johnson, G. B. 1977. Isozyme, allozyme, and enzyme polymorphisms: structural constraints on polymorphic variation. Current Topics in Biological and Medical Research **2:** 1-19.

Kiang, Y. T. and L. Wu. 1979. Genetic studies of esterase on the Taiwan wild rice population. Bot. Bull. Academia Sinica **20:** 103-116.

Loukas, M., Y. Vergini, and C.B. Krimbas. 1983. Isozyme variation and heterozygosity in *Pinus halepenis* L. Biochem. Genet. **21:** 497-509.

McIntosh, R. P. 1967. Continuun concept of vegetation. Bot. Rev. **33:** 130-187.

Nei, M. 1965. Variation and covariation of gene frequencies in subdivided populations. Evolution **19:** 256-258.

Nei, M. 1973. Analysis of gene diversity in subdivided populations. Pro. Nat. Acad, Sci., USA **70:** 3321-3323.

Schmitz, U. K. and K. V. Kowallik. 1986. Polymorphism and gene arrangement among plastomes of ten *Epilobium* species. Plant Mol. Biol. **7:** 115-127.

Shannon, L. M. 1968. Isozymes. Ann. Rev. Plant Physiol. **19:** 187-210.

Silander, J. A. Jr. 1984. The genetic basis of ecological amplitude of spartina patterns. III. Allozyme variation. Bot. Gaz. **145(4):** 569-577.

Sneath, P. H. and R. R. Sokal. 1973. Numerical Taxonomy. W. H. Ferman and Company, San Francisco, 573 pp.

Whittaker, R. H. 1967. Gradient analysis of vegetation. Biol. Rev. **42:** 207-267.

Wu, L. and A. D. Bradshaw. 1972. Aerial pollution and the rapid evolution of copper tolerance. Nature (London) **238(5360):** 167-169.

五節芒之族群研究
III. 台灣離島中綠島及蘭嶼之五節芒族群變異

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五節芒(*Miscanthus floridulus*)除分布於台灣本島外，亦遍布於台灣離島中的綠島及蘭嶼。沿著該兩島的海岸公路，作者在綠島上選七個族群，在蘭嶼島上選十三個族群，並採其五節芒葉子以聚丙烯醯胺電泳法(PAGE)分析芒草中同功酶如過氧化酶及酯酶的分布。再以族群距離及相似度之公式分別將同功酶之資料計算出其族群間之關係。上述分析結果得知，綠島的五節芒有三個群叢，蘭嶼有四個群叢。若將兩島之族群合併分析得知四個群叢，並顯現生態種之形成。族群分析的結果亦進一步指出族群間並不連續，而成非連續性，且族群的相似配對成跳躍性，此結果與 McIntosh 的植被連續說(continuum concept of vegetation)相違。但卻支持 Gleason 的「植被個體觀」(individualistic concept of vegetation)之理論。本研究之族群非連續性結果有待更深入的探究。