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A Method for Subgrouping the S-type of CMS Forms in Maize

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Summary. Simple correlations were calculated between nine different cms sources (cms-S, -R, -ML, -L, -CA, -EK, -C, -Rb, -T) on the basis of the weighted restoring reactions of 41 inbred lines. The Principal Component Analysis was applied to a 9×9 matrix which clearly grouped cytoplasms according to their similarities. The Principal Component I included S, R, ML, L, CA and EK cytoplasms; the Principal Component II contained C and Rb cytoplasms while T-cytoplasms was placed in Principal Component III. This corresponds to the main groupings indicated in the literature (Beckett 1971). However, after varimax rotation of the Principal Components, the S main group, including the 6 tested cytoplasms, fell into 3 subgroups: I.: S, R, ML; II.: L, CA; III.: EK.

These data indicate that the Principal Component Analysis can be used to select a limited number of cms sources from the S group, representing the variability of the cytoplasmic gene pool of that group.

Key words: Maize cytoplasmic male sterility – Fertility restoration patterns – Principal Component Analysis

Introduction

Although the epidemic of the T-race of *H. maydis* caused a considerable loss in yield only in the USA, the continuously growing exchange of basic materials and trade of seeds has lead experts in plant protection and breeding all over the world to exercise increased care. As a preventive measure, the use of cms analogues susceptible to T-race of *H. maydis* was forbidden in several countries. According to tests by Smith et al. (1971), cms-T (formerly used most frequently) Q, HA, and P cytoplasms could not be taken into consideration because they were susceptible to the T-race of *H. maydis,* and attention turned to non T-type resistant cytoplasmic male sterile sources. As more than eighty non T-type cytoplasmic male sterile sources were known by the middle of the 1960's (Duvick 1965), research could be started on a wide genetic basis in 1971.

On the basis of the fertility restoring ability of 5 to 18 inbred lines, Beckett (1971) classified the cms types into T, S, and C groups. Upon analysing fertility restoration by the major Rf genes, a negligible variability was found within the main groups (Beckett 1971). Gracen and Grogan (1974), however, in a more detailed study, observed variability within the groups.

Among the non T-type cytoplasmic male sterile sources, the members of C group are preferred in seed production because their male sterility is stable – male fertile revertants are not mentioned in any literature (Laughnan and Gabay 1978). Using a biochemical method – restriction endonuclease fragment analysis of mtDNAs – Levings and Pring pointed out significant differences among the N, T, S, and C cytoplasms, (Levings and Pring 1976, 1977; Pring and Levings 1978). On the basis of this method, representatives of the C group, also including recently discovered Bb, E and ES sources, were divided into three subgroups: (1) C, (2) Rb, Bb, E and (3) El Salvador (Pring et al. 1980). Some variability was also detected in the fragment patterns of mt DNA in normal cytoplasmic lines of different geographical origin (Levings and Pring 1977).

The S-group contains the majority of cms sources. They are more sensitive to environmental conditions than the T or C types of male steriles and show partial fertility on several inbred backgrounds. Cytoplasmic, as well as nuclear male fertile revertants, may also occur (Laughnan and Gabay 1973). In spite of this, their application is reasonable because only a minor proportion of inbred maize lines can be produced as male sterile analogues on cms-C sources. The Principal Component Analysis, as shown in this paper, may be of considerable help in choosing a limited number of sources from the great number of available cms-S sources.

Materials and Methods

Cms sources (cms-S, -R, -ML, -L, -CA, -EK, -C, -Rb, -T) were crossed as females with 41 inbred lines and then backcrossed to the same lines for at least five generations. Only one or two

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backcrosses were made in some cases since the recurrent parent proved to be an excellent restorer, and there was no segregation observed in the degree of pollen fertility. In 1979 and 1980 the degree of fertility was evaluated at silking phenophase by using a scale 1–4.

The criteria for rating fertility were the following:

1) male sterile, no anthers exserted;
2) sterile anthers exserted, no pollen shed;
3) partially fertile anthers exserted, some pollen shed; Proportion of anthers exserted highly variable;
4) slightly subnormal or normal anthers, fully fertile.

When heterogeneity could be observed in the degree of fertility within the plants of a cms analogue, restoration reaction of the given line was always characterized by the higher value (weighted fertility). Possible simple correlations between these 9 cytoplasms were calculated on the basis of the restoration reactions of these 41 lines. Data were estimated by the Principal Component Analysis. Calculations were done by Sváb's method (1979) with a computer R40 in the Institute of Cybernetics, University Attila József, Szeged.

Results

Correlations

Table 1 contains the weighted restoration reactions, while correlations between tested cytoplasms are given in Table 2. It may be seen from these data that T-cytoplasm has no correlation with any other cytoplasms. There was a close correlation between cms types S and R; S and CA; R and ML ($r < 0{,}7$) and medium correlations ($0.3 < r < 0.7$) could be revealed between cytoplasms C and Rb; S and ML; S and L; S and EK; R and CA; R and EK; ML and L; ML and CA; ML and EK; L and CA; L and EK; CA and EK.

Principal Component Analysis

The Principal Component Analysis provides additional information about which variables have high common variations and which ones belong together. As the data in Table 3 indicate, three out of five calculated Principal Components contain 75.3% of all variations of variables. There are 6 cytoplasms (S, R, ML, L, CA, EK) in the Principal Component I which binds 50.9% of all variations. The Principal Component II contains C and Rb cytoplasms – both with a negative sign. T-cytoplasm can be found in Principal Component III, where it forms a separate group, as could be expected on the basis of a simple correlation analysis. The situation of the variables before rotation is illustrated on the system of coordinates (Fig. 1). Cytoplasms S, R, ML, L, CA, EK, and C, Rb are settled close to the circle: that is to say, the major part of their variation is determined by the two

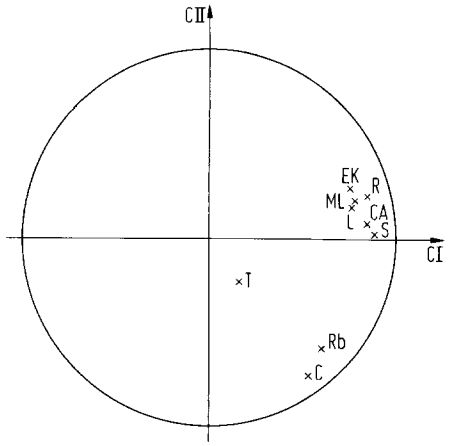

Fig. 1. Situation of the cms-types on the systems of coordinates without rotation

Table 1. Fertility ratings of 9 male sterile cytoplasms in 41 inbred backgrounds

	S	R	ML	L	CA	EK	C	Rb	T
A90	3	4	2	2	3	2	4	2	1
A654	1	1	1	1	2	3	1	1	1
A619	4	3	3	4	3	3	4	4	1
A632	4	4	4	2	3	4	3	3	1
A634	4	3	3	4	4	4	1	1	1
A635	4	4	3	3	3	3	1	1	1
A665	4	4	4	4	4	4	1	1	1
A636	4	4	4	2	2	3	3	3	1
A664	3	3	2	2	2	2	2	2	1
A641	3	2	1	2	2	2	1	2	1
A662	4	3	3	3	3	4	4	4	2
B14A	4	3	3	4	3	3	3	1	3
B37	2	2	1	2	2	2	1	1	1
B73	2	2	2	1	1	2	1	1	1
Mo17	3	2	2	2	1	1	1	1	1
CM7	2	2	4	1	1	3	1	2	1
CM105	2	2	2	2	3	2	1	2	1
CM109	3	2	2	1	3	2	3	2	1
GK3	2	2	2	1	1	3	4	2	1
GK13	4	4	4	4	4	4	4	4	1
GK71	4	3	2	1	3	3	1	2	1
GK71/L	3	2	2	2	2	3	1	2	1
GK72	2	2	2	2	2	3	1	2	1
Ia153	2	2	2	2	2	3	1	2	1
W153R	3	3	3	2	2	2	4	2	4
W153R/H	4	4	3	2	3	3	1	4	1
W153R/L	1	2	2	2	2	2	2	2	1
Bc5	2	1	1	1	1	2	1	1	4
Mt42	4	4	4	4	4	4	4	4	4
Oh43/301	4	2	2	2	4	4	4	4	1
Oh43/H	4	3	2	4	4	4	4	4	1
Oh43Rest3	4	4	3	4	4	4	4	4	1
Pa762	4	2	3	1	3	1	4	4	1
H98	1	1	2	2	2	2	1	1	1
W64A	2	1	1	2	2	1	4	4	1
W117	2	2	1	2	1	1	4	1	1
W401	1	1	2	1	2	2	2	1	1
WF9	1	1	1	2	1	2	1	1	1
W37A	3	3	2	2	3	2	1	1	1
Co125	4	2	2	2	3	4	3	3	1
W629A	2	1	2	1	2	2	1	4	1

Table 2. Correlations calculated on the fertility ratings of the cms-types

	S	R	ML	L	CA	EK	C	Rb	T
S	1.0000								
R	0.7793	1.0000							
ML	0.6195	0.7303	1.0000						
L	0.5735	0.6164	0.4715	1.0000					
CA	0.7254	0.6280	0.4951	0.6726	1.0000				
EK	0.5664	0.5407	0.5671	0.5697	0.6482	1.0000			
C	0.4054	0.3153	0.2593	0.3185	0.3502	0.1816	1.0000		
Rb	0.4960	0.2955	0.3593	0.2590	0.4804	0.3564	0.6108	1.0000	
T	0.1132	0.0907	0.1548	0.1627	0.0044	0.0557	0.2129	−0.0054	1.0000

Table 3. Principal component weights (a_{ij}) of the 9 cms types

cms-types	Without rotation					After varimax rotation				
	I	II	III	IV	V	I	II	III	IV	V
S	0.876	0.025	−0.028	−0.125	0.144	0.482	0.019	−0.348	−0.589	−0.157
R	0.840	0.239	0.079	−0.268	0.277	0.472	0.014	−0.137	−0.806	−0.010
ML	0.765	0.187	0.135	−0.480	−0.141	0.183	0.101	−0.160	−0.778	−0.258
L	0.762	0.191	0.140	0.433	0.218	0.769	0.117	−0.128	−0.299	−0.125
CA	0.843	0.089	−0.186	0.290	0.024	0.666	−0.074	−0.310	−0.345	−0.287
EK	0.750	0.277	−0.072	0.184	−0.482	0.485	0.013	−0.104	−0.353	−0.650
C	0.529	−0.747	0.037	0.036	0.196	0.215	0.170	−0.771	−0.130	0.124
Rb	0.601	−0.602	−0.337	−0.068	−0.255	0.095	−0.079	−0.830	−0.177	−0.281
T	0.158	−0.232	0.899	0.072	−0.170	0.041	0.971	−0.059	−0.061	−0.005
Eigenvalue	4.577	1.189	1.009	0.634	0.529	1.814	1.008	1.574	1.988	0.721
Cum. eigenvalue %	50.86	64.07	75.28	82.32	88.20	20.16	31.36	48.85	70.94	78.94

Principal Components. Cytoplasm T is, however, near the origin. This is to be expected as its variation can be explained by the Principal Component III. After varimax rotation of Principal Components, cytoplasms were grouped – according to their connections – on the basis of Principal Component weights (a_{ij}):

Principal Component I: L, CA (S group)
Principal Component II: T (T group)
Principal Component III: C, Rb (C group)
Principal Component IV: S, R, ML (S group)
Principal Component V: EK (S group)

Cytoplasm T, C, and Rb fell into the Principal Component II and III whereas the six cytoplasms, belonging to the S main group, were classified, after rotation, into three different Principal Components (Table 3).

Discussion

From the correlations between the tested cytoplasms, it is evident that T-cytoplasm has no connection with any other cytoplasms. We obtained similar results in the case of C- and Rb-cytoplasms although the correlation between them was moderate. After applying Principal Component Analysis to the 9×9 matrix, cytoplasms were clearly grouped according their similarities (Fig. 1). The results agree with the main groups known from literature (Beckett 1971; Gracen and Grogan 1974; Koncz et al. 1980). After varimax rotation of the Principal Components, however, cytoplasms belonging to the S group were further classified into three subgroups (Table 3). On the basis of these results, Principal Component Analysis appears to be suitable not for separating main groups with high authenticity but also for subgrouping within one main group. Subgrouping cytoplasms EK and L separately is also supported by Gracen et al.'s tests (1979). The authors designate these two cytoplasms as tpyes showing no typical S restoration pattern. The principle of choosing cms sources emerges from the knowledge of subgroups, the correct procedure using fewer cms types from one subgroup and more from different subgroups. In this way, the danger of gene vulnerability can be reduced.

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