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EVALUATION OF ZAPALOTE CHICO ACCESSIONS FOR CONSERVATION AND ENHANCEMENT

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ABSTRACT - Conserving traditional maize (*Zea mays* L.) landraces grown by farmers in Latin America can contribute to food security. Core subsets of such landraces that represents that diversity can be used for their enhancement. One such enhancement effort was conducted on a total of 81 accessions drawn from the CIMMYT maize collection of the race Zapalote Chico and newer accessions collected in 1999 from Istmo de Tehuantepec, Oaxaca, Mexico, where the race Zapalote Chico is predominantly grown. These accessions were evaluated at four locations in the Mexican states of Morelos, Guerrero, and Oaxaca for agronomic and morphological traits to assess the intraracial diversity for conservation and enhancement. Eleven agronomic and morphological traits were measured and used for a multivariate cluster analysis. The cluster analysis produced four non-overlapping clusters with 63 accessions, indicating the intraracial diversity of the Zapalote Chico. The analysis also formed two other clusters with four races, including Tepecintle and Olotillo, which have been introduced to the region of Istmo de Tehuantepec. Using a selection index that accounts for grain yield, grain quality, and standability, and an agronomic performance rating, a breeder core subset (the best 20%) was chosen to represent phenotypic diversity among the clusters. In the core subset, fifteen accessions comprise the core subset of the race Zapalote Chico and three accessions comprise the core subset for the other races. These core subset accessions, which represent the diversity of the regional landraces of maize, can be enhanced through introgression of improved lines or populations for yield potential, drought resistance, and ear rot resistance, while maintaining the desirable grain quality traits of the original races.

KEY WORDS: Zapalote Chico; Maize landrace; Core subset; Germplasm conservation.

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INTRODUCTION

Mexican farmers grow maize on 9.6 million ha, with an average yield of 2.2 t/ha. Commercial seed is only used for about 20% of maize production; for the rest, farmers use saved seed of traditional, open pollinated landraces or advanced generations of commercial maize varieties and hybrids (TABA, 1995, 1997; PINGALI, 2001). The traditional maize races are highly adapted to local production conditions and uses such as preferences for specific grain types for local dishes and they constitute a key component in household food security. On-farm in situ conservation of local maize races in Mexico requires proper assessment of intra-racial diversity and a breeding strategy for enhancement to increase productivity while maintaining genetic diversity on-farm (EYZAGUIRRE and IWANAGA, 1996; BRUSH, 1999; SPERLING *et al.*, 2001). Intra-racial diversity of agronomic and morphological traits in the races Tuxpeño, Conicó, and Bolita has been studied using the CIMMYT bank accessions, and core subsets (BROWN, 1989, 1995) were chosen to represent phenotypic diversity of each race (CROSSA *et al.*, 1994, 1995; TABA *et al.*, 1994, 1998a,b).

Core subsets of a germplasm collection cover the phenotypic spectrum of the collection (BROWN, 1995; BROWN and SPILLANE, 1999). As Latin American maize collections are composed of different maize races (GOODMAN and BROWN, 1988), core subsets can be constructed by racial core subsets. A racial core subset can be designated after evaluation of the accessions that are grouped by race or race groups, taking into account phenotypic diversity among them. Breeder core subset accessions of maize races in the CIMMYT maize collection can be useful sources of genes for yield, grain types, earliness, stress tolerance, and value added traits as they represent phenotypic diversity between and within maize races preserved at the CIMMYT maize bank (TABA *et al.*, 2003). In 1999, the CIMMYT maize genetic resources program initiated prebreeding using core subsets of the Carribean maize races (TABA *et al.*, 1998a) that were crossed with CIMMYT elite lines. To enhance CIMMYT gene pools, useful alleles can be identified in testcross evaluations of lines developed in breeding crosses involving the core subsets and subsequently introgressed into corresponding gene pools (TABA *et al.*, 2004). Similar maize germplasm enhancement work is reported in the project "Germplasm Enhancement of Maize" (GEM; POLLAK, 1997; POLLAK and SALHUANA, 2001), in which breeding crosses were made between elite tropical germplasm accessions and proprietary lines of private seed companies to obtain both enhanced accessions and elite germplasm lines for hybrid development.

Zapalote Chico, one of the prehistoric races of maize (WELLHAUSEN *et al.*, 1952), evolved with the indigenous Zapotec culture in the western coastal plains of the state of Oaxaca, Mexico. Genetic studies on chromosome knobs (KATO in collaboration with McCLINTOCK, 1981) indicated it belongs to the group of western coastal races of maize in Mexico that includes Zapalote Grande, Chapalote, Reventador, Pepitilla, and Harinoso de Ocho. Zapalote Chico is cultivated in the Istmo de Tehuantepec region (latitude 16-17:00 N and longitude 94-96:00 W) in Oaxaca at elevations from sea level to 800 m, on an estimated 80,000 ha over three growing seasons: October-January, where the crop is grown using residual moisture; February-May, when the crop is grown under irrigation; and June-October, during the rainy season. MUÑOZ *et al.* (1992) described the race's adaptation to local growing conditions and farmers' preferences, including early maturity, ease of shelling, good forage quality, and good corn chip and totopo making qualities. Zapalote Chico, an early maturity race, provides more reliable yields across years than higher yielding, late maturing varieties and hybrids in the Istmos de Tehuantepec, where strong winds can result in stalk and root lodging. Its kernel weight is low because of its floury texture. It is known to be relatively tolerant to long day lengths and to have active regulatory MuDR-like elements (GUTIERREZ-NAVA *et al.*, 1998), causing a high rate of mutation when crossed with non-Mutator lines. These genetic properties, together with farmers' selection practices, may be responsible for maintaining its relative purity as one of the traditional Mexican maize races.

Cultivation of Zapalote Grande has diminished in Istmo de Tehauntepec, where Zapalote Chico is now largely cultivated, while races Tepecintle and Olotillo, late white dent races, are largely cultivated in southern coastal regions in the state of Oaxaca. Both these latter races are late and tall, having long cylindrical ears with different kernel sizes and kernel row numbers that differentiate them. They can still be cultivated in the vicinity of Istmo de Tehuantepec, in areas where the seasonal winds do not affect stalk and root lodging of the plants.

Broadening the genetic base of local cultivars (EYZAGUIRRE and IWANAGA, 1996; BRUSH, 1999; SPERLING *et al.*, 2001) requires introgression of desirable alleles or traits from elite lines or populations and acceptance of enhanced cultivars by farmers. A study of intra-racial diversity and subsequent formation of core subsets of Zapalote Chico will assist in ex-situ conservation in the national maize bank (INIFAP) and CIMMYT gene bank, and in-situ maintenance in the Istmo de Tehuantepec and enhancement of the race in the Istmo de Tehuantepec and INIFAP and CIMMYT breeding stations. Superior core accessions can be enhanced through prebreeding using appropriate elite lines and populations, while maintaining the genetic background of the race through farmers' selection practices (LOUTTE *et al.*, 1997; LOUTTE and SMALE, 2000; BELLON *et al.*, 2002).

The objectives of this study were (i) to evaluate agronomic and morphological performance of the accessions collected in Istmo de Tehuauntepec, Oaxaca, Mexico, where the race Zapalote Chico is predominantly cultivated; (ii) to group them into non-overlapping clusters by cluster analysis; and (iii) choose breeder core subsets to represent phenotypic diversity among the clusters for use in on-farm in situ and ex situ conservation. We especially hope to use this information to enhance Zapalote Chico's yield potential and specific agronomic traits, while preserving grain quality and adaptation.

MATERIALS AND METHODS

Experimental data

We collected 69 new accessions in 21 villages in the region of Istmo de Tehuantepec, in May, 1999. Collection was undertaken from the stores of harvested ears (husks intact) of individual farmers. We consulted with representatives in the villages to collect as much local maize diversity as possible. Twenty-five to thirty-five ears were sampled from each farmer variety. After shelling, the bulked seed lot became an accession. Based on the ear types, most of the new accessions belonged to the race Zapalote Chico and several to other race classes such as Olotillo and Tepecintle, which were introduced to the regions by the farmers. The CIMMYT maize collection has 12 accessions of the race Zapalote Chico that were collected in the 1940-50s (WELLHAUSEN *et al.*, 1952). All new accessions and the race Zapalote Chico accessions of CIMMYT's maize collection were evaluated in a 9 x 9 simple lattice design with two replications. The trial was planted at four locations: (1) Tlaltizapán, Morelos State, 18:81N, 99:07W, at 940 masl; (2) Tepalcingo, Morelos State, 18:35N, 98:50W, at 1,200 masl; (3) Iguala, Guerrero State, 18:22N, 99:33W, at 731 masl; and (4) Sta. Gertrudis Miramar, Istmo de Tehuantepec, Oaxaca, 15:04N, 94:15W, at 50 masl. Plots consisted of two 5 m rows with approximately 53,000 plants/ha. Two seeds per hill were planted and later thinned to one plant per hill. Agronomic practices differed at each location but were the normal maize cropping practices of the experimental stations where each trial was planted. Planting dates varied from November 1999 through May 2000. In the trial at Tlaltizapán, CIMMYT Pool 16 (tropical, improved, lowland, early, white, dent germplasm) was planted as a border to have an idea of its performance compared to the accessions from Istmo de Tehuauntepec. The field data used for cluster analysis consisted of days from planting to anthesis (AN), plant height (cm) from soil to tassel top (PH), ear height (cm) from soil to ear node (EH), grain moisture (%) at harvest (MO), average ear length measured on 5 ears (cm) (EL), average ear diameter measured on 5 ears (cm) (ED), average kernel length of 25 kernels from 5 ears (cm) (KL), average kernel width of 25 kernels from 5 ears (cm) (KW), average kernel row number of 5 ears (KRN), shelling (%) (SH), and ears per plant (EP). In addition, measurements of the following characters, were recorded: grain yield (Mg ha^{-1}, adjusted to 15.0% H_2O) (YLD), ear rot (%) (ER), root lodging (%) (RL), stalk lodging (%) (SL), number of days from planting to silk emergence (SI), number of days after silk emergence to ear leaf senescence (SE), and agronomic performance rating at harvest (APR). APR was taken to indicate ear quality and uniformity, as well as plant aspects. Racial classification, days to silk emergence, and days to ear leaf senescence were taken at the trial at CIMMYT's Tlaltizapan station in the state of Morelos. Complete data were obtained from the trials at Tlaltizapán, Tepalcingo, and Sta. Gertrudis Miramar. Yield, ear, and kernel data of late flowering entries at Iguala were not available. The combined analysis was performed on the data available from at least three locations.

Data analysis

We used a multivariate cluster analysis to study phenotypic diversity among the accessions. The adjusted means of AN, PH, EH, MO, EL, ED, KL, KW, KRN, SH, and EP from the combined analyses were used in the cluster analysis (FRANCO *et al.*, 1997) for grouping the accessions into non-overlapping homogeneous clusters. For numerical taxonomy and racial classification, GOODMAN and PATERNIANI (1969) and SANCHEZ *et al.* (1993) reported ear, kernel, and tassel traits were generally more stable than vegetative and agronomic traits in different environments. Breeders use these ear, kernel and agronomic traits for general selection, and

TABLE 1 - *The six non-overlapping clusters (CL) formed by the multivariate analysis* (WISHART, 1987). *The number of accessions, the numbers of core subsets (CS), the means of agronomic and morphological traits, and average Mahalanobis distances are shown for each cluster.*

			Agro-morphological traits[a]																			
CL	No. accs.	CS	AN day	PH cm	EH cm	YLD Mg/ha	MO %	SH %	EP	ER %	EL cm	ED cm	KRN	KL cm	KW cm	RL %	SL %	APR 1-5	EH/ PH	SE/ SI	SEL[b]	D²(i|j)[c]
a	12	2	94	217	134	2,4	27	82	0,8	16	14	4,4	12	1,1	0,9	18	6	2,8	0,62	0,47	4	181
b	29	7	71	141	71	2,8	12	88	1,0	18	11	4,3	10	1,1	1,0	10	13	2,8	0,50	0,66	77	98
c	6	1	86	177	95	3,2	24	80	0,9	18	14	4,7	13	1,1	0,9	8	5	2,5	0,53	0,53	44	105
d	21	4	70	137	66	2,3	11	87	1,0	21	11	4,2	11	1,1	0,9	7	11	3,0	0,48	0,67	74	112
e	11	3	76	156	80	2,8	14	86	1,0	16	12	4,4	12	1,1	0,9	8	12	2,8	0,51	0,58	74	67
f	2	1	73	140	68	2,5	12	90	1,0	23	10	4,8	14	1,2	0,8	8	9	3,3	0,48	0,63	74	131
Mean			76	156	82	2,6	15	86	1,0	18	12	4,4	11	1,1	0,9	10	11	2,9	0,52	0,61	63	
LSD5%			5	17	13	0,8	5	4	0,2	13	2	0,3	1	0,1	0,1	10	10	1,0				
C.V.			3	5	8	15,4	16	2	12,3	36	7	3,4	5	5,4	4,4	54	46	16,8				
[d]CIMMYT gene pool																						
Pool 16			85	144	81	6,1	15	83			17	4,6	14	1,2	0,9			1,5				

[a] AN = Days to anthesis, PH = Plant height, EH = Ear height, YLD = Yield ($Mgha^{-1}$), MO = Grain moisture at harvest (%), SH = Shelling (%), EP = Ears per plant, ER = Ear rot (%), EL = Ear length, ED = Ear diameter, KRN = Kernel row number, KL = Kernel length, KW = Kernel width, RL = Root lodging (%), SL = Stalk lodging (%), APR=Agronomic performance rating (1=good and 5=poor), EH/PH=ear/plant height ratio, and SE/SI=days to ear leaf senescence/days to silk ratio at Tlaltizapan.

[b] Selection index (SEL) = Yield*(1-(ear rot (%)/100))/4134*100+(100-(erect plants (%)))-4.45*grain moisture (%).

[c] D2(i|j) = Average Mahalanobis distance.

[d] Comparative performance of CIMMYT Pool 16, tropical early white dent at Tlaltizapan, Morelos, Mexico.

they are used for characterizing maize landrace accessions (WELLHAUSEN *et al.*, 1952). In the cluster analysis, the Ward method was used for obtaining the initial groups; the likelihood-based approach of Normix was utilized for improving those groups. The analyses were performed using the software CLUSTAN (WISHART, 1987). Canonical discriminant analysis was performed on the groups formed by the Ward-Normix method using SAS (SAS INSTITUTE INC., 1996), to display the variability between subgroups explained by the canonical variables. Mahalanobis distances were calculated to determine average distances among cluster centroids (MAHALANOBIS, 1930).

A breeder core subset of the CIMMYT maize collection was chosen with the use of a selection index based on yield (Mg ha-1), ear rot (%), erect plants (%), and moisture (%) calculated for each accession (Table 1). The selection index was used to account for grain yield, grain quality, and standability. In each of the non-overlapping clusters involving the race Zapalote Chico and other races such as Tepecintle and Ollotillo (according to the race classification), the best accessions (about 20%) with high selection index values and good agronomic performance ratings and/or representing specific variations of plant and ear traits were chosen to be included in the core subsets.

The Additive Main Effect and Multiplicative Interaction (AMMI) model (CROSSA, 1990) was performed on the adjusted mean yields of the selected core subsets of the trial (18) accessions and two typical accessions of Oaxaca 50 and 51 (WELLHAUSEN *et al.*, 1952), of the race Zapalote Chico that were collected in 1946 and have been preserved in CIMMYT's maize germplasm bank, to evaluate their adaptation at the 3 locations and to assess genotype x environment interaction.

RESULTS AND DISCUSSION

Formation of non-overlapping clusters and their characterization

The Ward-Normix strategy analysis of Normix after Ward (WISHART, 1987) on the adjusted means of 11 agro-morphological traits revealed 6 non-overlapping clusters among the 81 accessions evaluated (Fig. 1). Canonical discriminant analysis (data not shown) shows the relative similarity among the accessions and the phenotypic diversity within and between the non-overlapping clusters. The first canonical variable is highly correlated with days to anthesis (r = 0.604), plant height (r = 0.520), ear height (r = 0.563), and seed moisture at harvest (r = 0.537). The second canonical variable is correlated with kernel row number (r = –0.692), and kernel width (r = 0.520). The first canonical variable explains 85% of the total variation between clusters and the second canonical variable explains 8% of the total variation. Both canonical variables together explained 93% of the variation.

Clusters a and c combined consist of 18 accessions, including accessions of Mexican late white dent maize races of Olotillo, Tepecintle, Tuxpeño,

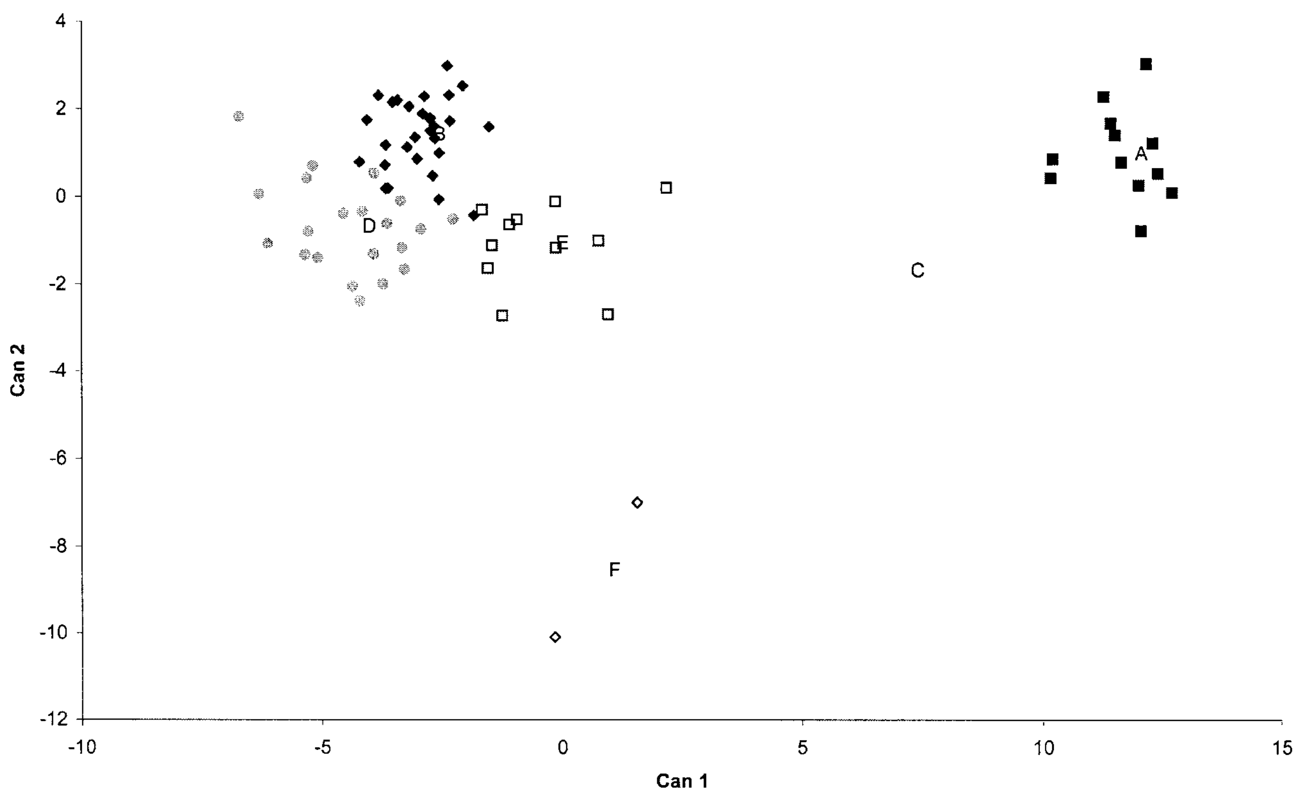


FIGURE 1 - A plot of the centroids of six clusters (a-f) based on the multivariate cluster analysis (WISHART, 1987) for 81 accessions from Istmo de Tehuantepec, drawn by the first and second cannonical variables. Each cluster is denoted by the same symbol.

TABLE 2 - *Agronomic performance of core subsets and typical race Zapalote Chico accessions in the states of Oaxaca, Morelos, and Guerrero in Mexico in 2000. The collection data and primary race class of the accessions are shown.*

CL[a]	Entry	Collection Name		Collection site and state province	Lat. N	Long. W	Alt. m	Race primary	AN[b] day	YLD Mg/ha^{-1}	APR 1 to 5	SEL[c]
a[1]	2	OAXA	841	La Reforma, Santa Maria Ecatepec	16:23	95:46	900	TEPECI6	93	2,6	3	25
a[1]	9	OAXA	848	Magdalena Tequisistlan,Tehuantepec	16:22	95:35	325	OLOTIL8	84	4,5	2	85
b[2]	27	OAXA	866	Cerro Iguana, Niltepec	16:29	94:43	100	ZAPCHI	69	3,0	3	89
b[2]	34	OAXA	873	Union Hidalgo, Juchitan	16:28	94:50	50	ZAPCHI	71	2,8	3	86
b[2]	46	OAXA	885	Santa Maria Xadani, Juchitan	16:21	95:01	50	ZAPCHI	73	2,9	3	82
b[2]	50	OAXA	889	Coronia Santa Rosita, Juchitan	16:26	95:01	50	ZAPCHI	69	3,0	3	79
b[2]	57	OAXA	896	El Crucero, Santa Maria Mixtequill	16:20	95:14	50	ZAPCHI	71	3,0	3	85
b[2]	59	OAXA	898	San Blas Atempa, Tehuantepec	16:16	95:10	50	ZAPCHI	73	3,0	3	77
b[2]	60	OAXA	899	Morro Mazatan, Tehuantepec	16:06	95:23	50	ZAPCHI	72	3,2	3	92
c[1]	29	OAXA	868	San Dionisio del Mar, Juchitepec	16:19	94:46	50	ZAPCHI	85	4,3	2	64
d[3]	80	OAXA	50	Niltepec	16:29	94:43	100	ZAPCHI9	70	1,7	4	65
d[3]	79	OAXA	51	Niltepec	16:29	94:43	100	ZAPCHI9	69	1,9	4	76
d[2]	24	OAXA	863	San Francisco Ixhuatan, Juchitepec	16:22	94:29	50	ZAPCHI	70	2,5	3	84
d[2]	49	OAXA	888	Jalapa del Marquez, Tehuantepec	16:26	95:56	50	ZAPCHI	68	2,4	3	84
d[2]	67	OAXA	906	Santiago Astala, Tehuantepec	15:59	95:40	800	ZAPCHI	71	2,8	3	91
d[2]	68	OAXA	907	El Camaron, Nejapade Madero	16:33	96:02	800	ZAPCHI	65	2,5	3	74
e[2]	17	OAXA	856	Chahuites, Juchitan	16:17	94:11	80	ZAPGRA	81	3,4	3	66
e[2]	61	OAXA	900	Morro Mazatan, Tehuantepec	16:06	95:23	50	ZAPCHI	76	2,9	3	84
e[2]	66	OAXA	905	Santiago Astala, Tehuantepec	15:59	95:40	50	ZAPCHI	75	2,8	3	90
f[2]	30	OAXA	869	San Dionisio del Mar, Juchitepec	16:19	94:46	50	ZAPCHI	74	2,4	3	81
						Trial mean (81 accesions)			76	2,6	3	63
								LSD 5%	5	0,8	1	
								C.V.	3	15	17	

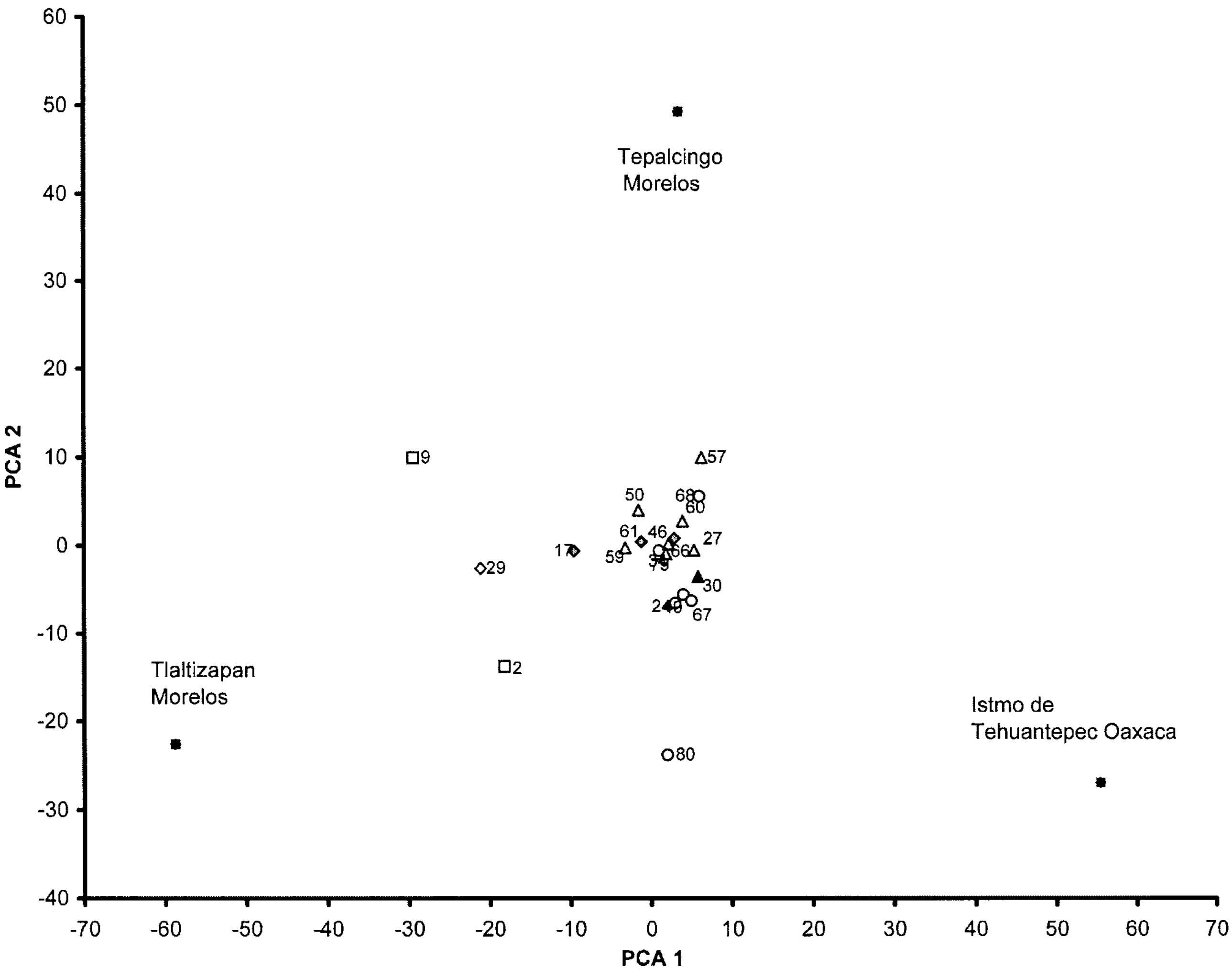
[a] CL = Cluster defined by Normix After Ward's (Wishart, 1987).
[b] AN = Days to anthesis, YLD = Yield Mg/ha. APR = Agronomic performance rating (1=good and 5=poor) at Tlaltizapan.
[c] Selection index (SEL) = Yield*(1-(ear rot (%)/100))/4134*100+(100-(erect plants (%)))-4.45*grain moisture (%).
[1] Core subsets of Tepecintle (TEPECI) and Olotillo (OLOTIL), late white dent clusters.
[2] Core subsets of Zapalote Chico (ZAPCHI) and Zapalote grande (ZAPGRA).
[3] Typical Zapalote Chico accessions from CIMMYT Maize Germplasm Bank.

and Comiteco in the primary race classifications (Table 2 and Fig. 1). They had 86-94 days to anthesis and were separated from the rest of the clusters.

Clusters b, d, and f flowered in 70-76 days, displayed early maturity, and the Zapalote Chico ear and kernel type. Clusters b, d, and f combined contained 52 accessions and were homogeneous for Zapalote Chico as the primary race. Cluster b was characterized by early maturity (71 days to anthesis) and relatively good yield (2.8 t/ha). Cluster d was the earliest to flower (70 days to anthesis) among the homogeneous race clusters, but also had the lowest yield (2.3 t/ha). Two typical collections of Zapalote Chico (Oaxaca 50 and Oaxaca 51) belong to cluster d. Cluster f had a high number of kernel rows (14) and a high shelling percentage (90%) among the clusters.

Cluster e with 11 accessions was also homogeneous for Zapalote Chico, except for 2 accessions with Zapalote Grande as the primary race. Cluster e included a late flowering accession of Oaxaca 856 that had the local name Zapalote Grande and yielded 3.4 t/ha (Table 2). It had 76 days to anthesis and 2.8 t/ha average yield.

The characteristics of clusters of b, d, e, and f are indicative of the phenotypic variations within the race Zapalote Chico, except Oaxaca 856 (classified as Zapalote Grande as the primary race and Olotillo as the secondary race), which is grouped in cluster e. There were clear variations between and



Note in the biplot: core subsets of Zapalote Chico: 17, 24, 27, 30, 34, 46, 49, 50, 57, 59, 60, 61, 66, 67, 68; typical Zapalote Chico accessions: 79, 80; core subsets of Tepecintle and Olotillo: 2, 9, 29.

FIGURE 2 - Biplot for yield performance of core subsets of the trial and Zapalote Chico typical accessions by principal component 1 and 2 from the AMMI analysis.

within the race clusters b, d, e, and f with respect to agro-morphological traits (Table 1 and 2) and average Mahalanobis distances seemed to confirm the relative distances among the centroids of the six clusters (Table 1 and Fig. 1).

Development of breeder core subsets and their characterization

The mean performance among the Zapalote Chico race clusters indicated that the race should have genetic variation for maturity and yield, although the differences among the clusters were small (Table 1). According to the characterization and race classification of the accessions, clusters b, d, e, and f were race clusters of Zapalote Chico and clusters a and c involved other races from the same region. A breeder core subset of Zapalote Chico, therefore, can be designated from the four, non-overlapping clusters b, d, e, and f to represent their phenotypic diversity as well as possible. To ensure genetic diversity is represented in the breeder core subset, 10 to 20% of the accessions from each of the non-overlapping clusters from the cluster analysis can be included. In the theoretical study by Brown (1989), about 10% of the representative samples should retain more than 70% of the alleles present in the original collection. The accessions in the different clusters are considered to be genetically diverse, and sampling 20% of accessions from each cluster should represent most of genetic diversity within it. Another criterion used for selecting individual accessions within a cluster for a breeder core is to designate agronomically superior accessions using a selection index based on yield, ear rot (%), standability (%), and seed moisture (%) (Tables 1 and 2). As a breeder core subset is expected to represent genetic diversity of a set of the accessions evaluated in the trial, it also includes unique characteristics such as plant and kernel color, kernel row number, and kernel type, in addition to the traits that have been accounted for using the selection index. Fifteen accessions in the trial were designated as a breeder core subset representing the race Zapalote Chico and chosen from clusters b, d, e, and f. Oaxaca 856 accession was classified as Zapalote Grande (primary race) and Olotillo as a secondary race; it seemed to be a racial mixture, having more kernel row numbers, and was grouped in cluster e by the cluster analysis. Zapalote Grande and Zapalote Chico are closely related races (WELLHAUSEN *et al.*, 1952) grown in adjacent regions.

In cluster a, Oaxaca 841 and Oaxaca 848 are chosen as a core subset of the cluster and classified as race Tepecintle and Olotillo, respectively. Oaxaca 868 is classified as race Tepecintle to represent the phenotypic diversity of the cluster c. These accessions of Tepecintle and Olotillo were collected in 1999 and are grown in the lowland of the states of Oaxaca and Chiapas near Istmo de Tehuantepec.

In the CIMMYT maize germplasm collection, race Tepecintle and Olotillo include 218 and 149 accessions, respectively. In addition, these accessions have been evaluated for agronomic and morphological traits (TABA *et al.*, 2003) to assess phenotypic variability within the race.

Evaluation of Latin American maize landrace accessions, for agronomic and morphological traits, has been carried out at CIMMYT's maize germplasm bank since the late 1980s. It has been reasonable to group the accessions by race for further evaluation, as maize accessions from the same race confer similar ear and plant morphology (ANDERSON and CUTLER, 1942). New accessions to be evaluated in CIMMYT's maize gene bank are grouped by race name, collection site, and country of origin. Breeder core subsets (about 20% of total) are then designated, following cluster analysis, to represent phenotypic diversity among the clusters formed in the evaluation trials. To date, we have evaluated some 7,700 accessions and have designated preliminary core subsets (TABA *et al.*, 2003). Confirmation of the breeder core subsets of CIMMYT's maize collection will be based on results from performance trials. After all accessions in the collection have been evaluated for their agronomic and morphological traits, it is hoped that about 20% of the collection will be designated to breeder core subsets, to represent each race, sub-race, and geographic variations.

In Table 2, collection sites, collection names, race classifications, days to anthesis, yields, agronomic scale ratings, and selection index values are shown for the designated core accessions as well as for Oaxaca 50 and Oaxaca 51, typical racial accessions of Zapalote Chico (WELLHAUSEN *et al.*, 1952). Among the race Zapalote Chico core subset, Oaxaca 907 was earliest to anthesis (65 days). From the race Zapalote Chico breeding core, Oaxaca 873, in cluster b, and Oaxaca 900, in cluster e, had purple culm color and Oaxaca 906, in cluster d, had a flintier and rounder grain type than the other accessions.

AMMI analysis

The AMMI bi-plot of Fig. 2 shows the analysis of yields of the 20 accessions in Table 2. Mean squares of variance for the accessions and accessions x locations were highly significant (data not shown). The bi-plot of the core subsets by principal components 1 and 2 indicated that all race Zapalote Chico core accessions were well adapted in Tepalcingo, Tlaltizapán, and Istmo de Tehuantepec. Among them, Oaxaca 866 (entry 27), Oaxaca 889 (entry 50), Oaxaca 899 (entry 60), and Oaxaca 885 (entry 46), from cluster b, yielded well in the range of 3.0-3.2 t/ha (Fig. 2, Table 2). In contrast, Tepecintle and Olotillo accessions (entry numbers 2, 9, and 29 that correspond to Oaxaca 841, 848, and 868 in Table 2 and Fig. 2) tended to perform better at Tlaltizapán and Tepalcingo than at Istmo de Tehuantepec. Oaxaca 50, entry 80, the typical accession of Zapalote Chico, had an average yield of 1.7 t/ha and performed poorly at three locations.

Fig. 3 indicates the collection sites for the 20 accessions of the core subsets and the typical accessions of Zapalote Chico that were analyzed using AMMI. All core subsets of the race Zapalote Chico were collected in Istmo de Tehuantepec (Fig. 3 and Table 2). The core subsets represented the geographical area where Zapalote Chico is predominantly cultivated. Oaxaca 50 and 51, the typical accessions of Zapalote Chico, came from Niltepec vil-

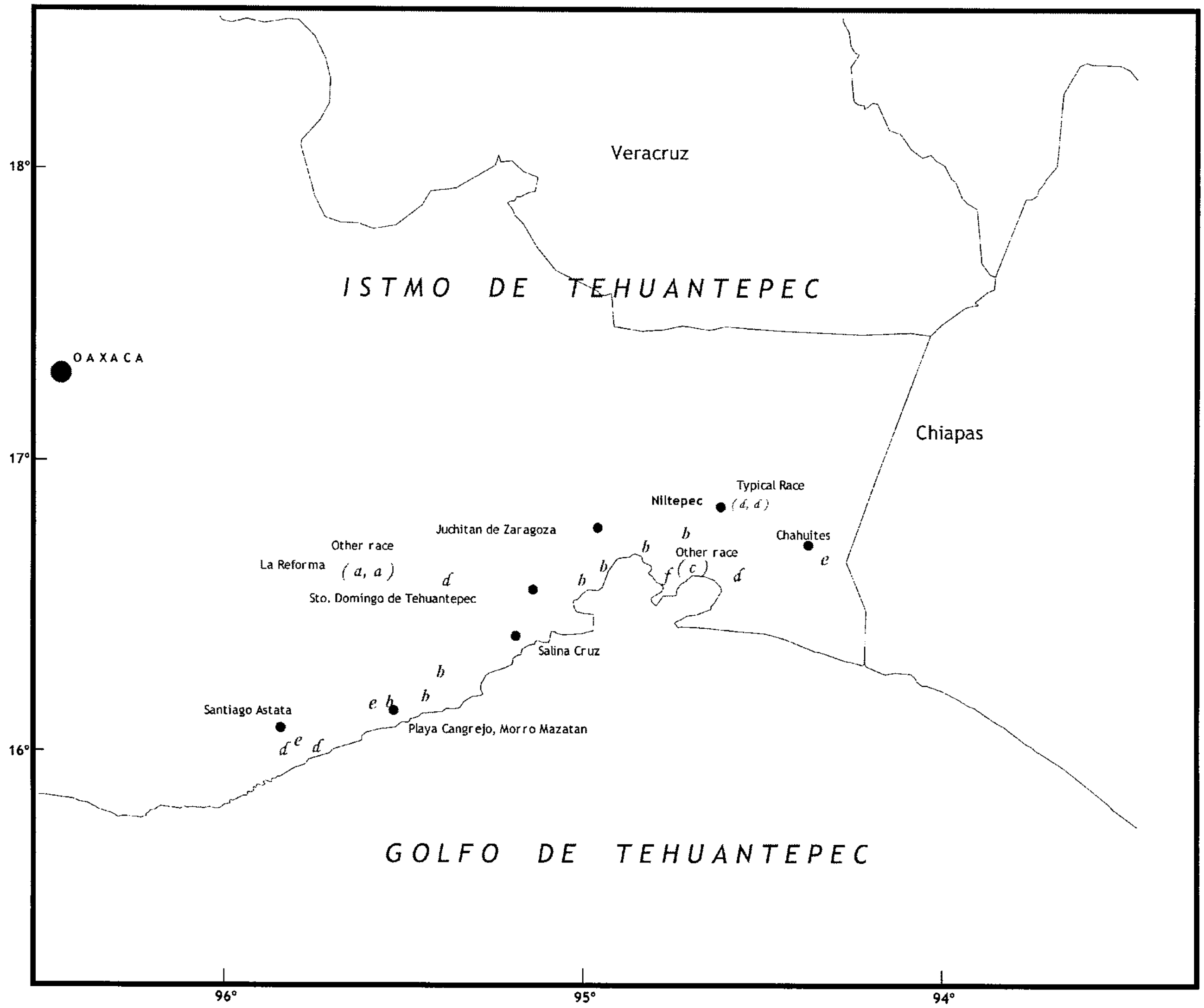


FIGURE 3 - Collection sites of core subsets of the trial and Zapalote Chico typical accessions in Istmo de Tehuantepec, Oaxaca, Mexico. Each accession is denoted by the cluster a-f to which it belonged (Table 2). Typical and other race (Tepecintle and Olotillo) accessions are in parenthesis.

lage in the central region. Oaxaca 868, race Tepecintle in cluster c, was collected in a village near the coast. Oaxaca 841 and 848, race Tepecintle and Olotillo, respectively, were collected in northwestern villages of Istmo de Tehuantepec (Fig. 3).

Use of core subsets

Maize races currently under cultivation in the region are open-pollinated, and the seed for planting the next crop is harvested from farmers' fields. Seed lots are often exchanged between the farmers (LOUETTE *et al.*, 1997; LOUETTE, 1999; LOUETTE and SMALE, 2000). Participatory plant breeding that employs mass selection from only local germplasm, without gene flow from other sources, can reduce on-farm genetic diversity (BROWN and YOUNG, 2000). It is desirable to enhance local maize races by introgression of new useful genetic traits.

Core subsets of Zapalote Chico can be crossed with advanced lines or gene pools of known heterotic patterns. Breeders can then evaluate them as sources of new genetic diversity, such as earliness and ear rot resistance, during prebreeding. The same crosses can be evaluated in situ (in this case at Istmo de Tehuantepec) after one or two backcrosses to the core subsets to recover the desirable traits of the race. Further recombination and selection in the respective breeding populations, involving the race core accessions, can be done on-farm to broaden the genetic base of the race.

CIMMYT Pool 16, a tropical, early, white dent planted in the border rows of the Tlaltizapán trial, had an estimated yield of 6.1 t/ha - almost twice the yield of the race core subsets - and took 80-85 days to reach anthesis (Table 1). To broaden the genetic base of the race Zapalote Chico, lines from CIMMYT Pool 16 and Pool 15 (flint counterpart to Pool 16) or advanced cycles of the pools can be introgressed to the core accessions to improve their yield potential, drought tolerance, standability, and resistance to foliar diseases, as well as avoiding the negative effects of MuDR-like elements. Ten core accessions from this study have been crossed with pools 15 and 16 and backcrossed to both the pools and the accessions. The backcrosses with the pools performed better than those to the accessions in the CIMMYT breeding nursery. The breeding populations (BC_1), improved for grain types, will be tested in Istmos de Tehuantepec using a participatory approach with farmers in the area.

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