

ISOZYME VARIABILITY AMONG EUROPEAN MAIZE POPULATIONS AND THE INTRODUCTION OF MAIZE IN EUROPE

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ABSTRACT - Several theories have been advanced to explain the origin of European maize (*Zea mays* L.) based on history, yet neither the introduction of this crop, nor the variability among European populations, have been extensively studied until recently. The aim of the present investigation was to relate isozyme variation in European maize populations to the introduction of the species on this continent. Ten isozyme systems encoded by 19 loci were used to classify 404 maize open-pollinated populations from six European countries. The average number of alleles per locus in the collection was 3.1. Allele richness was quite homogeneous across countries. The distribution of rare alleles supports the historical conclusion that Spain was the main entrance of maize to Europe. Alleles shared by maize from Mediterranean countries and Mexico or Guatemala suggest that the source of Mediterranean maize could have been these areas of Central America. Some rare alleles absent in Spain suggest that maize was also introduced through other countries. The collection of European populations was classified into two main groups, the smaller containing those of Northern Europe and the larger divided into Mediterranean and miscellaneous subgroups. Several singularities were seen that might be explained by drift or particular introductions. The results suggest that maize from North America came to Europe via the European Atlantic coast, and from Central America to the Mediterranean region, though introduction through other ways cannot be discarded on the basis of the present isozyme data.

KEY WORDS: *Zea mays*; Allozyme; History; Numerical taxonomy.

INTRODUCTION

The Old World discovered maize (*Zea mays* L.) at the end of the 15th century (WEATHERWAX and

RANDOLPH, 1955). Probably, maize was first brought from the Bahamas by Columbus. According to BRANDOLINI (1970) it was planted in Seville (southern Spain) in 1494. The first description of corn made in Europe was due to Peter Martyr, but little attention was paid to this report (WEATHERWAX, 1945). Later, maize was introduced from the Guatemalan and Mexican highlands, and this may have been better adapted to the temperate conditions of Spain than was Caribbean maize. Spanish and Portuguese explorers arrived in South America at the beginning of the 16th century and had colonized the continent by the end of the same century. The first scientific expedition to study the natural setting of South America was organized in 1570. Maize may therefore have been brought from South America to Europe by the end of the 16th century. However, based on isozyme variation, REVILLA *et al.* (1998) concluded that South American maize had little impact on Spanish maize.

Due to a lack of adaptation or poor agronomic performance, maize did not become a popular crop in Europe until the 17th century, when introductions began from North America. However, European explorers went to that continent early in the 16th century, and could have introduced the 'Northern Flint' race into Europe. North American maize might also have been introduced into Europe several times in subsequent centuries. Further, Central American maize might have reached many European regions via the Mediterranean. According to MEYER (ILTIS, 2000), maize introduced from Asia, Turkey and Greece was cultivated in Germany in 1543. LENG *et al.* (1962) stated that maize was first introduced into the Balkans by the Turks. These authors summarize previous works showing that maize reached Hungary from Italy and was brought to Rumania by the Turks in 1611. The Turkish role in maize introduction is hard to prove nowadays because, though

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first introductions could have happened in 1600, large scale introductions were made in late 1920s (ILARSAN *et al.*, 2001). Planting of the species probably increased in most European countries during the 16th century. Merchants from Italy and Turkey might have played a significant role in the spread of the species through Europe. However, this crop was not successful in Britain, perhaps because the British did not like it (GREIG, 1996) or due to a lack of adaptation. Nowadays, the most widely accepted hypothesis is that maize was introduced into the Old World by Portuguese and Spanish traders who traveled around the world after Columbus brought the first American seeds into Spain.

BOUZA (1953) credited the governor of Florida, Méndez de Cancio, for the introduction of maize into the provinces of Corunna and Asturias (northwestern Spain) from North America in 1604. PÉREZ (1978) found that maize was first reported in the province of Pontevedra, northwestern Spain, around 1631, becoming popular in that region after 1640. The introduction of maize into northern Spain could have occurred at the same time as its introduction into the northwest, but it probably had different origins (LLAURADÓ *et al.*, 1993; REVILLA *et al.*, 1998). Merchants from northern Spain could have spread maize throughout Europe since this area played a significant role in the commercial relationship between Europe and America.

By the end of the 16th century there was industrial and commercial decline in northern Spain, but the population was not greatly reduced because maize allowed an increase in the food supply (PÉREZ, 1978). Its introduction into northwestern and northern Spain was so successful that it quickly replaced millet (*Setaria italica* L.) as the main crop.

Besides these major introductions, many single introductions into specific places could have been made by returning soldiers, explorers or emigrants during the time of the conquest of America and succeeding centuries. Several unique varieties could have been introduced (REVILLA *et al.*, 1998).

Some authors suggest that maize was cultivated in the Old World before Columbus (see MANGELSDORF, 1974), but the most accepted hypothesis postulates that Europeans introduced maize into Asia and Africa after Columbus. Maize was cultivated in China early in the 16th century (MANGELSDORF, 1974). LU *et al.* (2002) analyzed Chinese populations and found some alleles not previously described anywhere else. Introductions into Africa or Asia could, however, have been made directly from America.

SANOU *et al.* (1997) found that African maize germplasm was closer to Southern Dent than to European maize. BONAFOUS (1836) and JEFFREYS (1971) were the main supporters of the theory of maize being introduced into Europe via Asia. SAUER (1960) supported the pre-Columbian introduction of maize into Europe based on the early introductions of maize in Italy and the Balkans. Finally, based on morphology, EDWARDS and LENG (1965) supposed that most Spanish maize collections came from Central America or the West Indies, while southeastern European maize probably came from North America. LENG *et al.* (1962) reports that BROWN, in 1960, suggested that West Indian maize may have been the first maize introduced into Europe after Columbus trips. However, these authors found that southeastern European maize bears little resemblance to Caribbean maize, probably due to subsequent hybridization, introgression, and selection.

Few isozyme studies have been performed on European maize populations. SALANOUBAT and PERNES (1986) studied 21 European maize populations with little variability to look for inferences about their origin. They concluded that European maize probably derived from several introductions from America. GERIĆ *et al.* (1989) found quite high isozyme variability among the Yugoslav races, including some alleles never recorded in any previous characterization of maize variety isozymes. If these authors correctly scored isozyme variability, the origin of some of the alleles they describe cannot be explained by an American origin. LLAURADÓ *et al.* (1993) studied some populations from northern Spain and concluded that they may have originated from Latin American races, and that the populations of the north and northwest had different origins. They also found some alleles never reported in any other European or US maize genotype, but which were present in the Mexican maize collection.

REVILLA *et al.* (1998) characterized the Spanish maize races and hypothesized that the first introductions of maize from the West Indies into southern Spain met with little success. Guatemalan maize was more successful in the south of Spain and appears to have expanded towards the Mediterranean area. South American maize had less influence in Spain than North American maize, and the latter was well established in the north and northwest of the country during the 17th century, increasing its hold over the following centuries. These authors defined three main groups of Spanish maize: northern maize from North America, southeastern maize

from Central America, and a main group, produced by hybridizations arising from contact between the former two, with representation all over the country.

Most authors assume that maize was introduced into Europe no more than five centuries ago, although the crop did not become popular until the 17th century, and maize breeding was not important in Europe until the 20th century. If this is so, variability among European maize populations should be related primarily to the American sources first introduced by the Spanish and Portuguese, and secondarily by Italian merchants. Maize introductions into other European countries could have occurred directly from America or through Spain, Portugal or Italy, but also via the Ottoman Empire which might have had an important role in the crop's diffusion throughout the Mediterranean coastal region, including the coast of Spain. Isozymes have been used to clarify historical relationships among maize populations because they allow fast and extensive characterizations using a fair number of alleles, loci, and individuals (GOODMAN and STUBER, 1983; SMITH, 1986; DOEBLEY *et al.*, 1983; 1985; 1988; REVILLA and TRACY, 1995; REVILLA *et al.*, 1998). The aim of the present investigation was to relate isozyme variation in European maize populations to the introduction of the species on this continent.

MATERIALS AND METHODS

A collection of 404 European maize populations were analyzed. These populations constitute the national maize collections of six countries: 99 populations from Italy (Istituto Sperimentale per la Cerealicoltura), 90 from Spain (Consejo Superior de Investigaciones Científicas and Instituto Nacional de Investigaciones Agrarias y Agroalimentarias), 80 from France (Institut National de la Recherche Agronomique), 70 from Portugal (Banco Portugues de Germoplasma Vegetal), 50 from Greece (National Agricultural Research Foundation), and 15 from Germany (Institute of Plant Genetics and Crop Plant Research, Gatersleben). These national collections were made in each country, based on morphological, agronomic, and molecular characterizations, as well as on previous knowledge of the germplasm gained by breeders or curators. All the populations analyzed are open-pollinated farmer varieties introduced in Europe along the last five centuries. Collection and conservation have depended on the current practices of each country until now. Isozyme data and previous observation suggest that genetic drift has played a significant role in some populations, particularly on those collected long time ago. Currently these populations are kept in the Institutions referred above following a common procedure that mainly consist on performing as many crosses as possible among 300 plants of each population using each plant once as male or female. Further information about characteristics and availability of

this collection can be obtained from each curator or from the web page <http://www.ensam.inra.fr/gap/resgen88/>

A minimum of 40 seedlings from each population were germinated under standard conditions, and 20 individuals were analyzed. Also several inbreds were used as checks to compare alleles (REVILLA *et al.*, 1998). Extraction methods, electrophoretic techniques, and scoring procedures were performed according to STUBER *et al.* (1988). The enzymatic systems were chosen from those reported by GOODMAN and STUBER (1983), SMITH (1986), DOEBLEY *et al.* (1983, 1985, 1988), REVILLA and TRACY (1995), and REVILLA *et al.* (1998), in order to compare data with those previously published. Each plant was analyzed for 10 systems encoded by the following 19 loci: *Idb1*, *Idb2*, *Adh1*, *Mdb1*, *Mdb2*, *Mdb3*, *Mdb4*, *Mdb5*, *Mmm*, *Glu1*, *Acp1*, *Phi1*, *Cat3*, *Pgm1*, *Pgm2*, *Enp1*, *Got1*, *Got2*, and *Got3*. The number of loci and individuals per population is the usual number for these kind of large studies. Banding patterns were used to calculate allelic frequencies. The standard error of each allele frequency (p) was obtained following FALCONER and MACKEY (1996) as the squared root of $p(1-p)/2n$. Null alleles were not considered and, therefore, some small bias in the estimation of allelic frequencies may have occurred. Allelic frequencies were analyzed by means of cluster analysis using the modified Rogers' distance (WRIGHT, 1978) and the UPGMA clustering method. Calculations were performed with the NTSYS-PC (1997) program.

RESULTS AND DISCUSSION

Allele richness

All loci, except *Pgm1*, *Got3*, and *Mmm*, were polymorphic for the 404 populations of the European collection. The average number of alleles per locus for the whole collection was 3.1. This is similar to other collections such as 2.78 recorded for the Northern Flint race (DOEBLEY *et al.*, 1988), 2.76 for the sweet corn collection (REVILLA and TRACY, 1995), 2.67 in the Corn Belt Dent racial complex (SMITH, 1986), and 3.3 alleles per locus reported in Southern Dent (DOEBLEY *et al.*, 1988). This figure is lower than 5.17 alleles in the Bolivian maize (GOODMAN and STUBER, 1983) and 7.09 for Mexican populations (DOEBLEY *et al.*, 1985). Given the wide range of ecological and geographical conditions of the different countries included, the present results could be biased by selection due to environmental adaptation. However, we can reasonably expect that selection or adaptation had a minor effect on allelic distribution because the selective value of isozymes is low according to most reports (NEVO *et al.*, 1988; GUSE *et al.*, 1988).

Allele richness was homogeneous across the national collections studied, the number of alleles per locus being almost proportional to the number of populations per country. The number of alleles per locus was similar in different countries, although highest for Spain (2.9), followed by France (2.6),

Portugal (2.5), Italy (2.5), Greece (2.4), and finally Germany (2.2). The number of rare alleles ($p < 0.01$ in the whole collection) was also higher in Spain (11), followed by Italy (9), France (8), Portugal (6), Greece (3), and finally Germany (2). These data show that Spain has the highest variability, and therefore the highest number of unique entries, probably because Spain was the main entrance for maize from America, as previously suggested based on historical data (WEATHERWAX and RANDOLPH, 1955).

All alleles found among European maize populations have been previously reported in American collections, supporting the American origin of European maize. Although this conclusion could seem obvious for current scientists, evidences supporting this explanations have been mainly historical or linguistic to date (FINAN, 1950). Furthermore, original Indian (MAURIA *et al.*, 2000) or Chinese (LI *et al.*, 2002) alleles, not present anywhere else, could be used to support the old theory of an Asian origin of maize, or at least to pretend that maize should have been introduced in Asia before than in Europe to allow the production of variability in Asia that has not happened in Europe.

The presence of rare alleles in a maize collection is helpful when considering the origin of germplasm. Some rare alleles are present in Spain, particularly *Mdb3-18* which was previously described in populations from the north of Spain (LLAUDÓ *et al.*, 1993), Yugoslavia (GERIĆ *et al.*, 1989), and in most sources of temperate and exotic American maize germplasm (Table 1). The allele *Mdb4-14.5* was present in maize from several Spanish regions (REVILLA *et al.*, 1998) and American sources (Table 1), *Glu1-9* was present in maize from even more Spanish regions than the previous alleles (REVILLA *et al.*, 1998) as well as in most American sources. The allele *Glu1-13*, present only in maize from Spain, Mexico (DOEBLEY *et al.*, 1985), and Guatemala (BRETTEING *et al.*, 1990) was absent elsewhere. The allele *Acp1-3.5* was previously described in Spanish maize (REVILLA *et al.*, 1998) and in several American sources (SMITH, 1984; DOEBLEY *et al.*, 1985; 1988; GOODMAN and STUBER, 1983). The presence of several rare alleles in Spain that are absent in the other European countries suggests that Spain was the entry for most of the variability of European maize, or that there were several singular introductions of maize into Spain.

Spanish maize shares several rare alleles with other southern European countries (Table 1). *Idh1-6* was found in maize from Spain and Portugal, and

has been described in Yugoslavian germplasm (GERIĆ *et al.*, 1989) and most American collections; *Mdb1-10.5* was found in maize from Spain and Greece, and has been described in many previous reports; *Mdb4-9*, detected in Spain, France, Italy, and Greece has previously been reported in maize from Mexico (DOEBLEY *et al.*, 1985) and Guatemala (BRETTEING *et al.*, 1990), but is absent in Yugoslavia and the U.S.A. The rare alleles found in several Mediterranean countries suggest that the introduction of maize into this area differed from its entry to the Atlantic coast. The alleles *Glu1-13*, *Acp1-3.5*, and *Mdb4-9*, shared by maize from the Mediterranean countries (particularly Spain) and Guatemala, suggest that the source of this Mediterranean maize could have been the highlands of this Central American country.

Other rare alleles present in most European maize (Table 1) were *Mdb2-3.5* and *Glu1-1* (absent in Germany), *Glu1-10* and *Acp1-6* (absent in Greece and Germany), and *Phi1-3* (absent in Italy and Portugal). Finally, two rare alleles were absent in Spain: *Adb1-2*, present in maize from Italy and Germany, and previously reported in Yugoslavia (GERIĆ *et al.*, 1989), Guatemala (BRETTEING *et al.*, 1990) and Bolivia (GOODMAN and STUBER, 1983); and *Mdb2-3.8*, present only in France and previously found in Mexico (DOEBLEY *et al.*, 1985). The presence of rare alleles (*Adb1-2* and *Mdb2-3.8*) in some European countries which are absent in Spain (Table 1) suggests that there were maize introductions into Europe that were independent of Spain.

The European populations had high proportions of alleles (*Mdb5-15*, *Enp1-10* and *Enp1-12*) that are also found in high proportion in sweet corn populations related to the Golden Bantam cultivar (REVILLA and TRACY, 1995). Golden Bantam is closely related to Northern Flint, which has probably made a notable contribution to European Flint germplasm. Also, Central American germplasm seems to have made a singular contribution to European maize, since some alleles (*Mdb2-3.8*, *Mdb4-9*, and *Glu1-13*, exclusively from Guatemala or Mexico) are present in European maize – particularly Spain – but also in Italy, Greece and France. This was not detected for South American maize. On the contrary, alleles shared by Europe and Bolivia are always present in North America, though there are alleles shared by Europe and North America that are not present in Bolivia. This suggests that European maize is more likely to have come from North and Central America than from South America.

TABLE 1 - Mean frequency (\bar{p}) \pm standard error of rare alleles ($p < 0.01$ in the whole collection) in the maize collection of each European country and in previous reports.

Germplasm group	Locus and allele							
	<i>Idb1-6</i>	<i>Adb1-2</i>	<i>Mdb1-10.5</i>	<i>Mdb2-3.5</i>	<i>Mdb2-3.8</i>	<i>Mdb3-18</i>	<i>Mdb4-9</i>	<i>Mdb4-14.5</i>
Spain	0.005 \pm 0.005	0.000 \pm 0.0	0.0003 \pm 0.001	0.013 \pm 0.008	0.000 \pm 0.0	0.001 \pm 0.002	0.022 \pm 0.011	0.002 \pm 0.004
France	0.000 \pm 0.0	0.000 \pm 0.0	0.000 \pm 0.0	0.007 \pm 0.006	0.003 \pm 0.004	0.000 \pm 0.0	0.002 \pm 0.003	0.000 \pm 0.0
Italy	0.000 \pm 0.0	0.001 \pm 0.001	0.000 \pm 0.0	0.002 \pm 0.001	0.000 \pm 0.0	0.000 \pm 0.0	0.006 \pm 0.006	0.000 \pm 0.0
Portugal	0.001 \pm 0.005	0.000 \pm 0.0	0.000 \pm 0.0	0.001 \pm 0.002	0.000 \pm 0.0	0.000 \pm 0.0	0.000 \pm 0.0	0.000 \pm 0.0
Greece	0.000 \pm 0.0	0.000 \pm 0.0	0.012 \pm 0.011	0.001 \pm 0.003	0.000 \pm 0.0	0.000 \pm 0.0	0.103 \pm 0.03	0.000 \pm 0.0
Germany	0.000 \pm 0.0	0.014 \pm 0.021	0.000 \pm 0.0	0.000 \pm 0.0	0.000 \pm 0.0	0.000 \pm 0.0	0.000 \pm 0.0	0.000 \pm 0.0
Mean	0.002 \pm 0.001	0.001 \pm 0.001	0.001 \pm 0.001	0.005 \pm 0.003	0.001 \pm 0.001	0.000 \pm 0.0	0.019 \pm 0.005	0.001 \pm 0.001
Previous reports								
Other European ^a								
North of Spain ^a	0.211		0.002	0.027		0.018		
Spanish races ^a	0.012		0.001	0.024			0.076	0.016
Yugoslavia ^a	b	b	b	b		b		
U.S.A. ^c								
Inbreds ^c	0.030		0.039	0.227		0.086		0.005
Hybrids ^c	<0.01			0.37		0.04		<0.01
Northern flint ^c			0.140	0.008		0.010		
Southern dent ^c	0.044		0.002	0.135		0.122		0.009
South West ^c	b		b	b		b		b
Other American ^d								
West Indies ^d	b			b		b		b
Mexico ^d	0.158		0.024	0.14	0.004	0.094	0.007	0.026
Guatemala ^d	b	b		b		b	b	b
Bolivia ^d	0.005	0.001	0.006	0.010		0.013		
Germplasm group	Locus and allele							
	<i>Glu1-1</i>	<i>Glu1-9</i>	<i>Glu1-10</i>	<i>Glu1-13</i>	<i>Acp1-3.5</i>	<i>Acp1-6</i>	<i>Pbi1-3</i>	<i>Enp1-4</i>
Spain	0.009 \pm 0.007	0.002 \pm 0.003	0.025 \pm 0.012	0.0003 \pm 0.001	0.007 \pm 0.006	0.007 \pm 0.006	0.008 \pm 0.007	0.002 \pm 0.003
France	0.017 \pm 0.01	0.000 \pm 0.0	0.002 \pm 0.004	0.000 \pm 0.0	0.000 \pm 0.0	0.005 \pm 0.006	0.015 \pm 0.009	0.000 \pm 0.0
Italy	0.007 \pm 0.006	0.000 \pm 0.0	0.002 \pm 0.003	0.000 \pm 0.0	0.000 \pm 0.0	0.001 \pm 0.002	0.000 \pm 0.0	0.000 \pm 0.0
Portugal	0.0005 \pm 0.002	0.000 \pm 0.0	0.002 \pm 0.004	0.000 \pm 0.0	0.000 \pm 0.0	0.005 \pm 0.006	0.000 \pm 0.0	0.016 \pm 0.011
Greece	0.014 \pm 0.012	0.000 \pm 0.0	0.000 \pm 0.0	0.000 \pm 0.0	0.000 \pm 0.0	0.000 \pm 0.0	0.001 \pm 0.004	0.000 \pm 0.0
Germany	0.000 \pm 0.0	0.000 \pm 0.0	0.000 \pm 0.0	0.000 \pm 0.0	0.000 \pm 0.0	0.000 \pm 0.0	0.067 \pm 0.046	0.000 \pm 0.0
Mean	0.009 \pm 0.003	0.000 \pm 0.001	0.007 \pm 0.003	0.0001 \pm 0.0	0.002 \pm 0.1	0.004 \pm 0.002	0.007 \pm 0.003	0.003 \pm 0.002
Previous reports								
Other European ^a								
North of Spain ^a	0.002		0.003			<0.001		0.002
Spanish races ^a	0.002	<0.001			0.003	0.006	0.017	
Yugoslavia ^a	b		b			b		b
U.S.A. ^c								
Inbreds ^c	0.059	0.002	0.015				0.002	0.015
Hybrids ^c	0.03				<0.01		<0.01	<0.01
Northern flint ^c			0.027		0.007		0.005	0.004
Southern dent ^c	0.040		0.005				0.005	0.084
South West ^c	b	b	b				b	b
Other American ^d								
West Indies ^d		b	b				b	b
Mexico ^d	0.191		0.091	0.009	0.002	0.002	0.006	0.080
Guatemala ^d	b	b	b	b			b	b
Bolivia ^d	0.011	0.001	0.018		0.001		0.0003	0.003

^a LLURADÓ *et al.*, 1993; REVILLA *et al.*, 1998; and GERIĆ *et al.*, 1989, respectively^b The mean frequency of the collection was not available in the article, but is low in each variety^c Inbreds (STUBER and GOODMAN, 1983); hybrids (SMITH, 1984); Northern Flint and Southern Dent (DOEBLEY *et al.*, 1988); and South West (DOEBLEY *et al.*, 1983).^d BRETTING *et al.*, 1987; DOEBLEY *et al.*, 1985; BRETTING *et al.*, 1990; and GOODMAN and STUBER, 1983, respectively

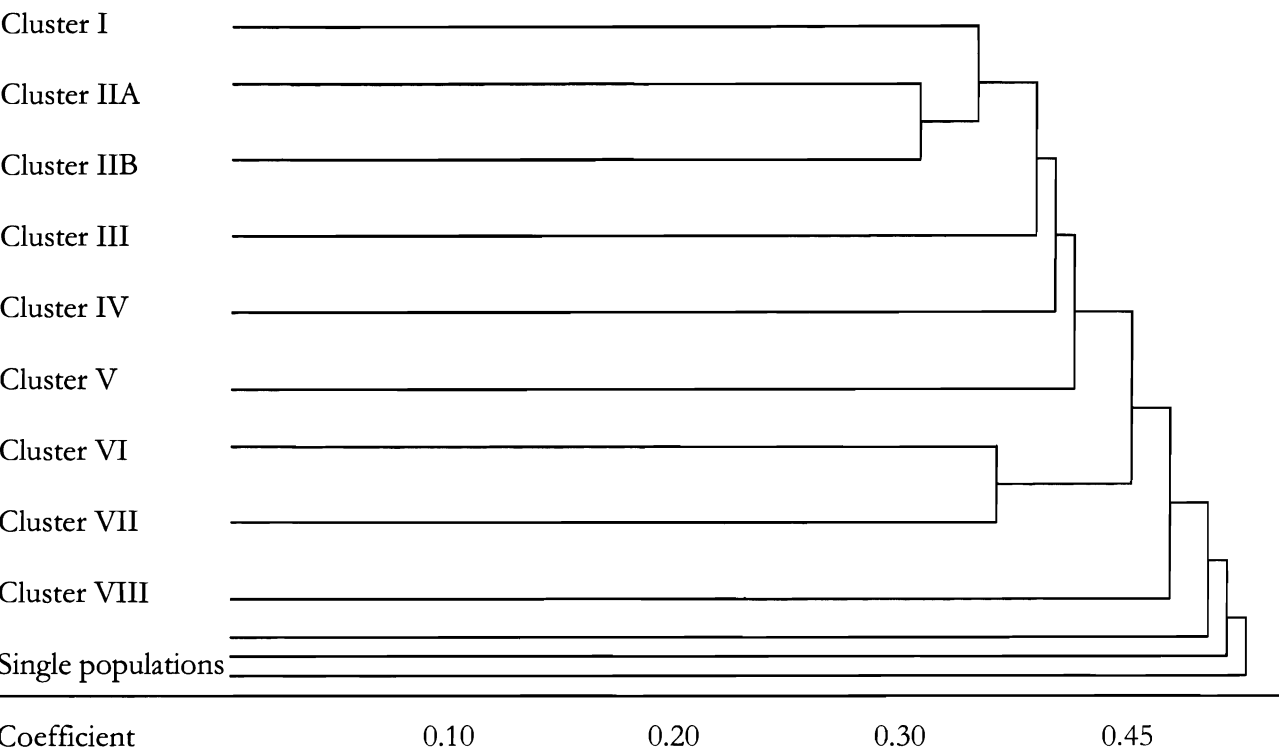


FIGURE 1 - Scheme of the average linkage cluster analysis of 404 open-pollinated populations using the modified Rogers' distance based on isozyme allele frequencies. See table 2 for cluster content.

Cluster analysis

The phenogram was based on the modified Rogers' distance and UPGMA clustering method, and had a low cophenetic correlation of 0.66 (NTSYS-PC, 1997). The poorly representation of the matrix distance by the phenogram, was due to the presence of ties, therefore we will not discuss the small clusters but only the most general clusters. Considering clusters at a coefficient of 0.3, two main clusters and several minor clusters and single populations can be distinguished (Figure 1 and Tables 2 and 3):

- I. A cluster of 39 populations consisting of 47% of the German populations, 25% of the French populations, and a few populations from all other countries. This could be considered a Northern European germplasm.
- II. A cluster comprising most European populations, with two subgroups:
 - II A - Mediterranean germplasm (98 populations consisting of 49% of the Greek populations, 31% of the Italian populations, 29% of the Portuguese populations, 19% of the Spanish populations, and 6% of the French populations).

- II B - General European germplasm (229 populations including 67% of the Portuguese populations, 66% of the Spanish populations, 59% of the Italian populations, 59% of the French populations, 32% of the Greek populations, and 13% of the German populations).
- III. Three northern Italian plus one French population coming from the border with Italy.
- IV. Two Greek populations.
- V. One Greek and one Spanish population.
- VI. A miscellaneous group from Mediterranean counties, France, Italy, Spain and Greece.
- VII. One Spanish popcorn and one flint French population.
- VIII. Two Italian populations.
- S. Several single populations separated from the rest.

Allelic compositions are not particularly rare even in the singular populations. These populations separated from the rest because they include several fixed alleles, most of them are probably caused by drift due to the small number of specimens used for multiplication in the past. Drift cannot be discarded in any cultivated populations because tradi-

TABLE 2 - Accession number and name (when available) of 404 European maize populations^a, and the cluster into which they are grouped in an average linkage cluster based on isozyme allele frequencies using Rogers' distance.

Number	Name	Cluster	Number	Name	Cluster
France (FRA041-)			Italy (ITA037-)		
0003	Weckelsheim	I	0005	Nostrano dell'isola	IIB
0005	Wantzenau	I	0009	Melia del dent, melia del bec	IIB
0006	Bade	I	0019	Rostrata o dentedicane piem	IIA
0010	Estarvielle	IIA	0021	Pignoletto ibrido	IIA
0011	Sost	IIB	0022	Quarantino	IIA
0012	Portet	IIB	0024	Caragua bianco	IIB
0014	Massat	I	0025	Rostrato giallo	IIA
0015	Lacaune	I	0026	Melgun bianco	IIB
0016	Berry	IIB	0027	Agostano giallo	IIB
0017	Barisis	IIB	0029	Bergamasco	IIB
0019	Liesse	IIA	0030	Quarantino bianco	IIB
0020	Campan Galade	IIB	0031	Quarantino giallo 100 gg	IIA
0022	Esterre	IIB	0034	Bani	IIB
0023	Laruns	IIB	0035	Quarantino	IIB
0028	Colmar	IIB	0037	Quarantino bianco	IIB
0031	Wagnonville	IIB	0043	Brianzolo	IIB
0032	Laval_LUzege	IIB	0044	Taiolone cremonese	VIII
0036	Ruffec	IIB	0045	Ottofile mantovano	IIA
0090	St_Maximin	IIA	0050	Locale	IIA
0098	Landes	IIB	0055	Melgonin	IIA
0121	Beaumont	I	0057	Nostrano dell'isola	IIA
0193	Aturignac	I	0058	Scagliolo	IIA
0194	Grand Roux Basque	IIB	0060	Giallo agostano	IIA
0198	Marmande.36	I	0061	Ottofile	IIB
0199	Parleboscq	I	0063	Nostrano locale	IIB
0241	Millette de Finham.AG	IIB	0067	Locale	IIA
0243	Floirac Ccamboni B	I	0071	Agostanello	IIA
0361	Cassagnabere	I	0076	Nostran di sterch	IIA
0363	Landes	IIA	0080	Zamengo	III
0473	Thurey	IIB	0088	Scagliolo frassine	IIB
0474	Ain	IIB	0091	Bianco Perla	III
0478	Labastide de Boussign	IIB	0092	Dentone	I
0484	Gironde	IIB	0100	Bianco perla	III
0488	Estaut	IIB	0108	Otesa (tipica)	IIB
0492	Juif	IIB	0112	Pignolino nostrano	IIB
0494	Chateaufrenaud	I	0114	Cinquantino Bianchi	IIB
0496	Louhans	IIB	0116	Wisconsin locale bianco	IIB
0497	Bantanges	S	0118	Bianco perla locale	IIB
0499	Rovon	III	0121	Pignoletto d'oro	IIA
0502	Lautrec	IIB	0122	Pignolo giallo	IIA
0565	Correze.123	I	0123	Sorgo prestarolo (bonorivo)	IIB
0567	Boutx B	IIB	0124	Pignolo nostrano	IIA
0570	Moncassin	IIB	0128	Scagliolo cinquantino	IIB
0571	Seltz	I	0133	Denton	IIA
0572	Strasgourg Bades	I	0142	Bastardo	IIA
0599	Burg	IIB	0143	Spin	VIII
0600	Serre Rustaing	IIB	0148	Paesan	I
0602	St Laurent de Neste	IIB	0150	Colleoni	IIB
0606	Passins	IIB	0152	Scagliolo Marne	IIB
0607	Dore de Bordes	IIB	0154	Marano Vicentino	IIB
0610	Grand Roux Basque	IIB	0155	Scagliolo locale	IIB
0613	Millette la Cassignol	IIB	0163	Rossa precoce o Ungherese	IIB
0617	St Parthem.B	IIB	0166	SilverMine	S
0624	Landes	VI	0167	Rostrato (bianco)	IIB
0625	Landes	IIB	0168	Wisconsin 7 o 14	VI
0628	Dubos	IIB	0171	Giallo Tosoratti	IIB

0629	Feillens	IIIB	0172	Early Orange	IIIB
0630	Quarantaun	IIIB	0173	Giallo Nostrano	IIIB
0631	St Pantaleon	I	0177	Nostrano dell'Isola	IIIB
0636	Marmande.19	I	0185	Rostrato tardivo	IIIB
0639	Millette du Lauragais	IIIB	0192	Rostrato x Dentato	IIIB
0641	Grand Roux Basque	VI	0194	Gran. d. S.Colombano 16 file	IIIB
0651	Auga	IIIB	0195	Granoturco di En	IIIB
0667	Dijon	IIIB	0200	Locale	IIIB
0668	Millette Montagne Noire	I	0205	Maggengo	IIIB
0710	Frasema	I	0240	Nostrale locale	IIIB
0820	Blanche Htes. Pyr.C1	S	0277	Ignoto varietà R	IIIB
0964	Argeles	IIIB	0284	Locale	IIA
0967	Pontacq	IIA	0292	Cinquantino locale	IIA
0969	Monein	IIIB	0298	Nostrale	IIIB
1012	Ratenelle	IIIB	0317	Cutaccione	IIA
1019	Pontailier	VI	0336	Locale	IIA
1023	Alsace.3	IIIB	0344	Nostrano incrociato	IIA
1038	Saint Creac.C	IIIB	0346	Ideale	IIA
1039	Sarrancolin	IIIB	0352	Quarantino bianco	VI
1044	Pierrefitte	IIIB	0362	Zeppetello	IIIB
1047	Saint Engrace	IIIB	0373	Maglianese	IIIB
1050	Tarbes	I	0375	Primaticcio	IIIB
1051	Lourdes	IIIB	0394	Agostino o agostinello	IIIB
1052	Bagnères de Bigorre	VII	0403	Locale	IIA
Portugal (PRT0010-)			0408	Locale	IIIB
0008	M. ^b Pega	IIIB	0433	Bianco Montoro	IIA
0016	M. Branco	IIIB	0440	Quarantino bianco	IIIB
0019	M. Amarelo	IIIB	0441	Primitivo	IIIB
0020	M. Amarelo Laranja	IIIB	0464	Granoturco rosso	IIA
0029	M. Folhao	IIIB	0466	Giallo locale	IIIB
0032	M. Branco	IIA	0475	Bianco precoce	IIA
0045	M. Amarelo	IIIB	0477	Vinato	IIIB
0049	M. verdial	IIIB	0479	Bianco precoce	IIIB
0056	M. Boca de Sapo	IIIB	0488	Rodindia	IIIB
0072	M. Amarelo	IIA	0489	Nostrale o Calabrese	IIIB
0086	M. Pego	IIA	0493	Frumento Amuri Nostrale	IIIB
0088	M. Amarelo	IIA	0496	Bianco comune	IIIB
0107	M. Branco	IIIB	0509	Quarantino	IIA
0120	M. Amarelo	IIIB	0517	Locale irriguo	IIIB
0121	M. Branco	IIIB	0528	Bianco perla	IIIB
0133	M. Branco	IIA	0536	Quarantino bianco	IIIB
0136	M. da Batata	IIA	0545	Bastardello	IIIB
0143	M. Branco	IIA	0548	Poliota o Cortolese	IIA
0163	M. Branco Amarelado	IIIB	Spain, Pontevedra (ESP009-)		
0168	M. Branco	IIA	0020	Rebordanes	I
0186	M. Amarelo	IIIB	0025	Enano levantino/Hembrilla	IIA
0219	M. Amarelo Escuro	IIA	0028	Fino	IIA
0228	M. Amarelo Escuro	IIIB	0032	Rastrojero	IIA
0291	M. MORENO	IIA	0033	Tremesino	IIA
0304	M. Branco MORENO	IIIB	0055	Moeche	IIIB
0308	M. Amarelo	IIIB	0067	Norteño largo	IIIB
0310	M. Amarelo	IIIB	0069	Hembrilla/Queixalet	IIA
0317	M. Amarelo Laranja	IIIB	0089	Gomesende	IIIB
0318	M. Branco Amarelado	IIA	0200	La Estrada	IIIB
0319	M. Branco	IIA	0202	Pontevedra	IIIB
0321	M. Branco Perola	IIA	0203	Salcedo	IIIB
0324	M. Amarelo	I	0204	Canicouva	IIIB
0330	M. Amarelo	IIA	0205	Tuy	IIIB
0335	M. Branco Amarelado	IIA	0206	Guillarey	IIIB
0339	M. Amarelo Escuro	IIIB	0214	Viana	IIIB
0348	M. de Mirandela	IIIB	0217	Foz	IIIB
0358	M. Branco	IIIB	0218	Recaré	IIIB

0361	M. Milhao	IIIB	0238	Ribadumia	IIIB
0362	M. Unha de Porco	IIIB	0241	Coristanco	I
0366	M. Amarelo Miudo	IIIB	0242	Padrón	IIIB
0392	M. Branco Perola	IIIB	0248	Oroso	IIIB
0394	M. Branco	IIIB	0262	Hembrilla	IIA
0406	M. Amarelo	IIIB	0267	Maceda	IIIB
0415	M. Amarelo	IIIB	0270	Lalín	IIIB
0431	M. de Pias	IIA	0309	Berguedá	IIIB
0435	M. Branco	IIIB	0310	Andaluz	IIIB
0453	M. Branco	IIA	0311	Andaluz/Tremesino	IIIB
0457	M. do Rego	IIIB	0315	Basto/Blanco	V
0462	M. Verdial	IIIB	0322	Blanco	S
0530	M. Pichorro	IIIB	0323	Daxa	IIA
0566	M. Amarelo	I	0328	Queixalet	IIA
0569	M. Amarelo Gigante	IIIB	0330	Rosero	IIIB
0617	M. Rajadinho	IIIB	0334	Medina	IIIB
0667	M. Fasciado	IIIB	0336	Vejer	IIIB
0677	M. Amarelo	IIIB	0338	Basto/Rastrojero	S
0685	M. de Sto. Antoninho	IIIB	0343	Fino/Tremesino	IIIB
0813	M. Amarelo de Sequeiro	IIIB	0348	Grano de trigo/Rosero	VII
0815	M. de Cunha	IIIB	0349	Perla	S
0828	M. Amarelo	IIIB	Spain, Zaragoza (ESP007-)		
0867	M. Miudo Charnequinha	IIA	0032	Dobres	I
0916	M. Sequeiro, Miudo ou Rati	IIA	0109	Queirós	IIIB
0966	M. Amarelo Sequeiro Miudin	IIIB	0127	Haza de Sobas	IIA
1216	M. Branco	IIIB	0217	Arredondo	I
1526	M. Amarelo	IIIB	0218	Castro Urdiales	IIIB
2047	M. Branco Perola Verdial	IIIB	0220	Anero	IIA
2678	M. Amarelo	S	0235	Guadix	IIA
2704	M. Branco	IIA	0280	Cangas de Onís	IIIB
2764	M. Branco	IIIB	0298	Azcoitia	IIIB
2898	M. Amarelo	IIIB	0300	Sajambre	IIIB
3702	M. Branco	IIIB	0302	Valdeón	IIIB
Greece (GRC001-)			0339	Cangas de Narcea	IIA
0012		IIIB	0350	Riomolín	IIIB
0015		IIA	0436	Úbeda	IIIB
0016		IIIB	0441	Villanueva del arzobispo	VI
0017		IIA	0447	Nieves del Caso	IIIB
0019		IIA	0647	Lisa de Vall	IIIB
0021		IIA	0725	Andoain	IIIB
0030		IIIB	0784	Guetaria	IIA
0037		VI	0810	Berastegui	IIIB
0041		S	0892	Lazcano	IIIB
0050		IIA	0937	Soraluce	IIIB
0051		IIIB	0943	Castellote	IIA
0058		S	Spain, Coronna (ESP119-)		
0062		IIIB	73C03	Aranga 1	IIIB
0064		IV	78020	Boimorto 1	IIIB
0066		IIA	78057	Sobrado dos Monxes	S
0067		IV	78061	Camariñas	IIIB
0068		I	81006	Negreira	IIIB
0077		IIIB	81023	San Sadurniño	IIA
0083		IIA	81033	Arzúa	IIIB
0084		IIIB	81040	Teo	IIIB
0085		S	81047	Carnota	IIIB
0090		IIA	81054	Aranga 2	I
0091		IIIB	81061	Tinco	IIIB
0099		IIA	81063	Irixoa	IIIB
0103		IIIB	81064	Begonte	IIIB
0104		IIA	81068	Sarria	IIIB
0109		IIA	82001	Ayala	IIIB
0113		IIA	82002	Amurrio	IIA

0115	IIA	82004	Llodio	IIIB
0119	V	82012	Ataún	IIIB
0128	IIA	82019	Fika	IIIB
0132	IIA	82024	Azpeitia	IIA
0134	IIA	82031	Guernica	I
0147	IIA	82036	Markina	IIIB
0157	IIA	83002	Dumbria	IIIB
0160	IIIB	84020	Boimorto 2	IIIB
0161	IIIB	85020	Ponteareas	IIIB
0162	IIA	85022	Forcarei	IIA
0165	IIA	85025	Mondariz	IIIB
0169	IIA	85034	Covelo	IIIB
0172	I	Germany (DEU146-)		
0174	IIIB	0003	Gelber Badischer Landmais	S
0179	IIA	0010	Janetzki Gloria	I
0183	IIIB	0013	Caspersmayer Silozahnmais	S
0185	IIA	0014	Janetzki Astra	I
0187	IIIB	0015	Caspersmayer II	S
0190	IIIB	0023	Mecklenburger	IIIB
0191	IIIB	0024	Dippes Zuckermais	S
0250	IIIB	0026	Rimpaus Früher Binder	S
0328	IIA	0158	Mahndorfer Mais	I
		0239	Strenzfelder	I
		0242		I
		0312	Petkuser	I
		0707		I
		0730		IIIB
		1005		S

^a 80 French populations from the INRA, Diversité et Genomes des plantes cultivées, Montpellier, France. 99 Italian populations from the Instituto Sperimentale per la Cerealicoltura, Sezione di Bergamo, Italy. 70 Portuguese populations from the Banco Portugues de Germoplasma Vegetal, Braga, Portugal. 90 Spanish populations, 39 from the Misión Biológica de Galicia, Spanish Council for Scientific Research, Pontevedra, Spain; 23 from the Estación Experimental de Aula Dei, Spanish Council for Scientific Research, Saragossa, Spain, and 28 from the Centro de Investigaciones Agrarias de Mabegondo, A Coruña, Spain. 50 Greek populations from NAGREF, Cereal Institute of Thessaloniki, Greece. 15 German populations from Institute of Plant Genetics and Crop Plant Research (IPK, Gatersleben, Germany).

^b M. stands for Milho, the Portuguese name of maize.

tional farmers used to keep few ears for next planting and some of the first germplasm collectors probably were not aware of the adequate sample sizes required to keep variability.

Clusters I, III, IV, VIII, and some single populations are probably independent of any Spanish ori-

gin (Tables 2 and 3). Cluster I could represent direct introductions of North American maize into northern Europe. Clusters IIA, III, IV, V, VI, VII, VIII, and several single populations are Mediterranean groups some of which could be related to southeastern Spanish maize (REVILLA *et al.*, 1998). Apparently there

TABLE 3 - Distribution (%) of populations from each country among the clusters defined from the average linkage cluster of 404 European maize populations based on isozyme allele frequencies using Rogers' distance.

II. Main group			
I. North European	IIA. Mediterranean	IIIB. Miscellaneous	Other
25% French	6% French	59% French	10% French
47% German	0% German	13% German	40% German
4% Greek	49% Greek	33% Greek	14% Greek
2% Italian	31% Italian	59% Italian	8% Italian
3% Portuguese	29% Portuguese	67% Portuguese	1% Portuguese
7% Spanish	19% Spanish	66% Spanish	8% Spanish

is a large variability among Mediterranean maize. REVILLA *et al.* (1998) hypothesized that this germplasm might be related to maize of the first introductions from Central America, and in turn to that of the West Indies (BRETTEING *et al.*, 1987). The large variability of Mediterranean maize could be a sample of the variability found in Central America. The Ottoman empire could have spread this maize throughout the Mediterranean area. The East of Spain (the former kingdom of Aragon) was not allowed to trade directly with the Americas. Thus, maize should have been introduced from the south of Spain or by Mediterranean merchants, as this clusters suggests. Cluster IIB, representing most European maize germplasm, is related to the main group of Spanish maize as defined by REVILLA *et al.* (1998). This cluster suggests that later and successful introductions of North American maize via the Atlantic coast, quickly extended across Europe and probably gave rise to hybrids through contact with representatives of previous introductions. LLURADÓ *et al.* (1993) and REVILLA *et al.* (1998) suggest that there were at least two different introductions into northern and northwestern Spain. However, North American maize may have come into Europe an indeterminate number of times via the Atlantic coast of Europe over the last four centuries. This germplasm should have been well adapted to European conditions, and could have produced quite wide but homogeneous variability in most European maize, rendering it difficult to distinguish the original introductions.

Lastly, inbred production started in 1922 in the northwest of Spain and hybrid maize seed was commercialized since 1930 by GALLASTEGUI (1958). U.S.A. hybrids have been subsequently introduced in Europe, therefore, further hybridizations with original introductions and hybrids could have taken place. Single populations come from all countries, showing that singular introductions have probably happened everywhere; another cause of these singularities can be drift due to poor conservation could have caused many of these singularities. Particularly diverse are some of the German populations.

European breeders commonly use the term European Flint as part of the usual heterotic pattern European Flint x U.S. Dent. However, none of these clusters can be considered a true European Flint race since there is semident maize in most clusters, and the distances between clusters are not clear enough to allow the definition of races. Cluster I, however, might be the closest to a hypothetical Eu-

ropean Flint race. Isozyme variability among European maize is quite continuous, making difficult the distinction of discrete races. Within European maize, Mediterranean germplasm (IIA) maybe related to Central American, and Northern European germplasm (I) to the U.S. race Northern Flint. General European germplasm (IIB) is closer to the Mediterranean (IIA) than to Northern Flint (I) germplasm (Fig. 1).

The moderate variability of European maize, the short history of the crop in Europe (five centuries at the most) and the active exchange of crops between European countries and between Europe and America, does not allow a clear classification of European maize into definite races, either based on variability or origin. The isozyme data presented here suggest that maize from North America came to Europe through the middle and northern Atlantic coast, and from Central America to the southern Atlantic coast and the Mediterranean region. However, introduction through other ways cannot be discarded on the basis of the present isozyme data.

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REFERENCES

- BONAFOUS M., 1836 Histoire naturelle agricole et économique du maïs. Paris, France.
- BOUZA F., 1953 Noticias históricas sobre la introducción del cultivo del maíz en Galicia. Bol. Real Acad. Hist. **132**: 35-72.
- BRANDOLINI A., 1970 Razze Europee di maïs. Maydica **15**: 5-27
- BRETTEING P.K., M.M. GOODMAN, C.W. STUBER, 1987 Karyological and isozyme variation in West Indian and allied American mainland races of maize. Amer. J. Bot. **74**: 1601-1613.
- BRETTEING P.K., M.M. GOODMAN, C.W. STUBER, 1990 Isozymatic variation in Guatemalan races of maize. Amer. J. Bot. **77**: 211-225.
- DOEBLEY J.F., M.M. GOODMAN, C.W. STUBER, 1983 Isozyme variation in maize from the southwestern United States: taxonomic and anthropological implications. Maydica **28**: 97-120.
- DOEBLEY J.F., M.M. GOODMAN, C.W. STUBER, 1985 Isozyme variation in the races of maize from Mexico. Amer. J. Bot. **72**: 629-639.
- DOEBLEY J., J.D. WENDEL, J.S.C. SMITH, C.W. STUBER, M.M. GOODMAN, 1988 The origin of Cornbelt maize: the isozyme evidence. Econ. Bot. **42**: 120-131.
- EDWARDS R.J., E.R. LENG, 1965 Classification of some indigenous maize collections from southern and southeastern Europe. Euphytica **14**: 161-169.

- FALCONER D.S., T.E.C. MACKEY, 1996 Introduction to quantitative genetics, p. 51. Ed. Longman, Essex, U.K.
- FINAN J.J., 1950 Maize in the great herbals. Chronica Botanica Company, Waltham, Mass., U.S.A.
- GALLASTEGUI C., 1958 El campo gallego. Ed. Citania, Buenos Aires, Argentina.
- GERIĆ I., M. ZLOKOLICA, C. GERIĆ, C.W. STUBER, 1989 Races and populations of maize in Yugoslavia. Isozyme variation and genetic diversity. Systematic and Ecogeographic studies on crop gene pools 3. International Board for Plant Genetic Resources, Rome.
- GOODMAN M.M., C.W. STUBER, 1983 Races of maize. VI Isozyme variation among races of maize in Bolivia. Maydica **28**: 169-187.
- GREIG J., 1996 Archaeobotanical and historical records compared – a new look at the taphonomy of edible and other useful plants from the 11th to the 18th centuries A.D. Circaea **12**: 211-247.
- GUSE R.A., J.G. COORS, P.N. DROLSOM, W.F. TRACY, 1988 Isozyme marker loci associated with cold tolerance and maturity in maize. Theor. Appl. Genet. **76**: 398-404.
- ILARSAN R., Z. KAYA, A.A. TOLUN, P.K. BRETTING, 2001 Genetic variability among Turkish pop, flint, and dent corn (*Zea mays* L. spp. *mays*) races: enzyme polymorphism. Euphytica **122**: 171-179.
- ILITIS H.H., 2000 Homeotic sexual translocation and the origin of maize (*Zea mays*, Poaceae): A new look at an old problem. Econ. Bot. **54**: 7-42.
- JEFFREYS M.D.W., 1971 Pre-Columbian maize in Asia. pp. 376-400. In: Riley C.L., Kellwy J.C., Pennington C.W., Rands R.L. (Eds.), Man across the sea. Univ. of Texas Press, Austin and London, Texas.
- LENG E.R., A. TAVCAR, V. TRIFUNOVIC, 1962 Maize of southeastern Europe and its potential value in breeding programs elsewhere. Euphytica **11**: 263-272.
- LLAURADÓ M., J. MORENO-GONZÁLEZ, P. ARIÚS, 1993 Classification of Northern Spanish populations of maize by methods of numerical taxonomy. II. Isozyme variation. Maydica **38**: 249-258.
- LIU H., J.S. LI, J.L. LIU, R. BERNARDO, 2002 Allozyme polymorphisms of maize populations from southwestern China. Theor. Appl. Genet. **104**: 119-126.
- MANGELSDORF P.C., 1974 Corn. Its origin, evolution and improvement. Harvard Univ. Press, Cambridge.
- MAURIA S., N.N. SINGH, A.K. MUKHERJEE, K.V. BHAT, 2000 Isozyme characterization of Indian maize inbreds. Euphytica **112**: 253-259.
- NEVO E., A. BEILES, T. KRUGMAN, 1988 Natural selection of allozyme polymorphisms: a microgeographical differentiation by edaphic, topographical, and temporal factors in wild emmer wheat (*Triticum dicoccoides*). Theor. Appl. Genet. **76**: 737-752.
- NTSYS-PC, 1997 NTSYS-PC numerical taxonomy system. Version 2.02c. Exeter Software, Setauket, New York.
- PÉREZ J.M., 1978 Aproximación al estudio de la penetración del maíz en Galicia. In: E. Roel *et al.* (Eds.), La historia social de Galicia en sus fuentes de protocolos. Univ. Santiago de Compostela, Spain.
- REVILLA P., W.F. TRACY, 1995 Isozyme variation and phylogenetic relationships among open-pollinated sweet corn cultivars. Crop Sci. **35**: 219-227.
- REVILLA P., P. SOENGAS, R.A. MALVAR, M.E. CARTEA, A. ORDÁS, 1998 Isozyme variation and historical relationships among the maize races of Spain. Maydica **43**: 175-182.
- SALANOUBAT M., J. PERNES, 1986 Enzyme polymorphisms within and between European maize populations. Maydica **31**: 269-278.
- SANOU J., B. GOUENARD, A. CHARRIER, 1997 Isozyme variability in West African maize cultivars (*Zea mays* L.). Maydica **42**: 1-11.
- SAUER C.O., 1960 Maize into Europe. Vienna, Acts Int. Americanist Congress **34**: 777-787.
- SMITH J.S.C., 1984 Genetic variability within U.S. hybrid maize: Multivariate analysis of isozyme data. Crop Sci. **24**: 1041-1046.
- SMITH J.S.C., 1986 Genetic diversity within the Corn Belt Dent racial complex of maize (*Zea mays* L.). Maydica **31**: 349-367.
- STUBER C.W., M.M. GOODMAN, 1983 Allozyme genotypes for popular and historically important inbred lines of corn, *Zea mays* L. Agric. Res. Service, Southern Series No. 16. North Carolina Sta. Univ. Raleigh, North Carolina.
- STUBER C.W., J.F. WENDEL, M.M. GOODMAN, J.S.C. SMITH, 1988 Techniques and scoring procedures for starch gel electrophoresis of enzymes from maize (*Zea mays* L.). North Carolina Agric. Res. Ser. Tech. Bull. 286. North Carolina Sta. Univ. Raleigh, North Carolina.
- WEATHERWAX P., 1945 Early contacts of European science with the Indian corn plant. Proc. Indiana Acad. Sci. **54**: 169-178.
- WEATHERWAX P., L.F. RANDOLPH, 1955 History and origin of maize. pp. 1-61. In: Sprague G.F. (Ed.), Corn and corn improvement. Academic Press Inc., Publishers, New York.
- WRIGHT S., 1978 Evolution and the genetics of populations. Vol. 4. Variability within and among natural populations. University of Chicago Press, Chicago, Illinois.