

ears is shown in Table 2. A wide range in stearic acid composition was observed among original sibbed kernels and  $S_1$  ears of P. I. 197503 (Ethiopia). P. I. 185619 (Egypt) and P. I. 177651 (Syria) had typical stearic acid composition in original kernels, but had high stearic acid in some  $S_1$  ears.

Stearic acid is a saturated fatty acid and a high percentage may not be desired in corn oil for commercial food use. However, the high composition found in certain plant introductions is of considerable interest for future genetic and biochemical studies. Some of these high stearic acid lines have been crossed with low stearic acid inbred lines for genetic studies. Of biochemical interest, lines which are high in stearic acid have also had a much higher than usual amount of arachidic acid. Selfing in this material is being continued to obtain homozygous lines with high stearic acid composition.

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1. Multiple character analysis and chromosome studies in the *Tripsacum lanceolatum-pilosum* complex.

The genus *Tripsacum* has nine recognized species all of which are native to the new world. The range of their distribution extends from northern South America through Central America and Mexico into the United States. There are four diploid ( $2n = 36$ ) species (*T. floridanum*, *T. australe*, *T. maizar* and *T. zopilotense*). *T. dactyloides* has both diploid and tetraploid ( $2n = 72$ ) forms while *T. laxum*, *T. latifolium*, *T. lanceolatum* and *T. pilosum* exist as tetraploids. While the diploid species of *Tripsacum* are morphologically very distinct the tetraploid "species" are not so sharply delimited. This is nowhere better illustrated than in the tetraploid populations in Mexico. Even though these tetraploids are referred to as *T. lanceolatum* or *T. pilosum*, there are a

Table 1

Mean values and standard deviations for eight morphological characters studied in four populations of Tripsacum.

Species		Plant Height (Meters)	Blade Length (Cms.)	Blade Width (Mms.)	Pilosity Score of Leaf Sheath	Condition of the Auricle	Number of Branches in the Terminal Spike	Venation Index	Pedicel Length of the Accessory Male Spikelet (Mms.)
<u>T. maizar</u>	Mean	3.545	182.494	69.939	2.250	1.875	22.535	4.738	3.204
	S.D.	0.328	21.156	6.609	0.463	0.835	5.014	0.598	0.466
<u>T. zopilotense</u>	Mean	1.179	59.490	9.161	0.083	0.583	1.233	10.250	0.208
	S.D.	0.100	7.413	1.163	0	0.515	0.287	1.072	0.108
<u>T. pilosum</u>	Mean	3.121	157.017	41.317	1.652	1.261	9.877	5.257	2.400
	S.D.	0.456	17.430	9.182	0.935	0.752	4.191	0.865	0.750
<u>T. lanceolatum</u>	Mean	2.506	126.838	18.913	1.091	0.818	2.495	8.218	0.601
	S.D.	0.299	23.019	6.644	0.701	0.603	0.738	1.715	0.152

Table 2

Pairwise comparison of "distances" and probable errors for eight morphological characters studied in four populations of Tripsacum.

Pairs of Populations		Plant Height	Blade Length	Blade Width	Pilosity Score of Leaf Sheath	Auricle Score	Number of Branches in Main Spike	Venation Index	Pedicel Length of Male Spikelet
<u>T. maizar</u>	Distance	11.048	8.612	15.638	9.722	1.914	8.036	6.602	10.424
vs.									
<u>T. zopilotense</u>	Prob. error	0.00200	0.00675	0.00200	0.00387	0.19199	0.00899	0.01842	0.00272
<u>T. maizar</u>	Distance	1.080	1.320	3.624	0.856	0.774	2.750	0.710	1.322
vs.									
<u>T. pilosum</u>	Prob. error	0.29128	0.25836	0.68163	0.32898	0.33950	0.12639	0.35061	0.25820
<u>T. maizar</u>	Distance	3.310	2.520	7.700	1.992	1.470	6.968	3.010	5.424
vs.									
<u>T. lanceolatum</u>	Prob. error	0.09555	0.14184	0.01064	0.18465	0.23972	0.01534	0.11095	0.00741
<u>T. zopilotense</u>	Distance	6.984	7.852	4.216	3.536	1.070	3.860	5.156	5.106
vs.									
<u>T. pilosum</u>	Prob. error	0.01522	0.00986	0.02233	0.08536	0.29287	0.07256	0.03798	0.03893
<u>T. zopilotense</u>	Distance	6.644	4.426	2.498	3.114	0.420	2.406	1.458	3.020
vs.									
<u>T. lanceolatum</u>	Prob. error	0.01805	0.05468	0.14338	0.10538	0.40536	0.04611	0.24118	0.11046
<u>T. pilosum</u>	Distance	1.628	1.492	2.832	0.686	0.654	2.992	2.296	3.990
vs.									
<u>T. lanceolatum</u>	Prob. error	0.22153	0.23710	0.12137	0.35474	0.36063	0.11182	0.18861	0.06800

wide range of morphological forms connecting the so called "typical" T. pilosum and T. lanceolatum.

It has been proposed by Randolph (Randolph and Hernandez-Xolocotzi, 1950; Randolph, 1955) that the Mexican and Central American tetraploids of Tripsacum originated as amphiploids of T. maizar and T. zopilotense or similar diploid species whose chromosomes are sufficiently similar to permit genetic exchange.

Recently, Tantravahi (1968) proposed that the tetraploid T. lanceolatum-pilosum complex in Mexico might have originated as segmental allopolyploids as a result of hybridization between T. maizar and T. zopilotense.

Even though the existence of polymorphic populations of Tripsacum in Mexico has been recognized by everyone that has seen them, so far a quantitative morphological study and detailed chromosome analysis of these populations has not been undertaken. In the present study, 55 plants belonging to the tetraploid T. lanceolatum-pilosum complex and its supposed diploid parents have been subjected to a detailed morphological study (1) to obtain information on the limits of morphological variations in the diploid species and in the tetraploids, (2) to see if a given specimen can be assigned with reasonable accuracy to a species to which it is closely related and finally (3) to see if the data would support the hypothesis stated above. Initially 15 morphological characters were chosen. Of these eight were finally selected (Table 1). The characters were not randomly selected but were chosen because of their value in distinguishing the parental diploid species. The same eight characters have been used for the tetraploids. The means and standard deviations have been computed for the eight characters. Single character "distances" have been obtained for a given pair of species by dividing the difference in the means of that pair by the average standard deviation (Table 2). The "distance" statistics would give the relative usefulness of a particular character in distinguishing a given pair of species. For example, blade width is of greatest importance in distinguishing T. maizar from T. zopilotense. The condition of the accessory male spikelet (Pedicellate or Sessile) and venation index are useful in distinguishing the so called T. pilosum from T. lanceolatum.

Table 3  
 Pairwise discriminant analysis of T. maizar, T. zopilotense,  
T. lanceolatum, and T. pilosum.

Morphological Character	Discriminant Coefficients					
	<u>T. maizar</u> vs. <u>T. zopilotense</u>	<u>T. maizar</u> vs. <u>T. pilosum</u>	<u>T. maizar</u> vs. <u>T. lanceolatum</u>	<u>T. zopilotense</u> vs. <u>T. pilosum</u>	<u>T. zopilotense</u> vs. <u>T. lanceolatum</u>	<u>T. pilosum</u> vs. <u>T. lanceolatum</u>
Plant height	4.009	-0.140	0.264	0.054	-0.869	-0.221
Blade length	0.081	0.006	0.007	-0.015	-0.013	0.005
Blade width	0.117	0.015	0.076	-0.005	0.062	0.006
Pilosity score of leaf sheath	0.182	0.044	0.256	-0.019	-0.328	0.028
Auricle score	0.449	-0.004	0.043	0.033	-0.021	0.014
No. of branches in main spike	0.081	0.024	-0.017	-0.031	-0.578	0.007
Venation index	0.033	0.079	0.099	0.057	0.091	-0.004
Pedicel length of male spikelet	1.292	0.005	1.600	-0.084	-2.200	0.183
Average discriminant function for						
<u>T. maizar</u>	44.434	2.736	13.435			
<u>T. zopilotense</u>	11.573			-0.343	-1.530	
<u>T. pilosum</u>		1.909		-2.637		0.823
<u>T. lanceolatum</u>			5.071		-5.085	0.269
D <sup>2</sup> (D)	591.6 (24.4)	23.9 (4.9)	142.1 (11.9)	75.7 (8.7)	74.6 (8.6)	17.7 (4.2)

The technique of linear discrimination as described by Kendall (1951) has been used. The computations were done on an IBM 7094 computer using the BIMD 05 program developed by the medical school of the University of California at Los Angeles. This technique finds the weighted sum of a number of characters that distinguish the populations best. The sum itself is the discriminant function. The weights determined by these computations are called discriminant coefficients (Table 3). The average value of the discriminant function for a given population can be obtained by multiplying the average value of each character by the corresponding coefficient and then summing. The discriminant coefficients have been computed for the eight characters used in a pairwise comparison for all four populations. These data together with  $D^2$  and  $D$  values are presented in Table 3. The last value is roughly the difference in the standard deviations between the mean value of the functions of a particular pair of populations. For example T. maizar differs from T. zopilotense by 24 standard deviations while T. lanceolatum and the so called T. pilosum differ from each other by only about four standard deviations.

The method of multiple character analysis as used in the present study has two advantages over simple pairwise comparison of single characters. This method takes all characters together in assigning weights to them. Characters that best distinguish a given pair of populations will have higher weights, strikingly bringing out the differences. This method in fact pulls the populations apart as far as possible making the evaluation of relationships more meaningful and discrete. Secondly, using the coefficients it is possible to assign any doubtful specimen, a posteriori, to one of the populations to which it is closest in its relationship. In the present study three plants (65-1236, 65-1237 and 65-1238) all collected at the same locality and assigned to the so called T. pilosum showed a diploid chromosome number of 36 instead of the expected 72 (Table 4). These plants have to be included under T. pilosum (originating as parthenogenetic diploids) or T. maizar, one of the diploid parents with which T. pilosum shows some morphological resemblance.

Table 4

Frequency distribution of various types of associations observed at diakinesis in plants belonging to the Tripsacum lanceolatum-pilosum complex.

Plant Number	Diploid Chromosome Number	Frequencies of the types of associations			
		Quadrivalents	Trivalents	Bivalents	Univalents
F.T.G 65-1221	70 (?)	20.7	-	77.3	2.0
65-1235	72	16.3	4.3	72.8	6.6
65-1236	36	-	-	90.6	9.4
65-1237	36	-	-	89.5	10.5
65-1238	36	-	-	93.4	6.6
65-1239	72	16.3	-	83.7	-
65-1240	72	19.3	2.8	73.3	4.6
65-1241	72	18.6	1.4	73.5	6.5
65-1242	72	13.4	0.8	74.0	11.8
65-1243	72	15.8	2.9	70.1	11.2
65-1243a	72	20.9	-	79.1	-
65-1246	36	-	-	97.7	2.3
65-1247	72	12.8	1.2	84.5	1.5
65-1248	72	23.1	1.1	67.6	8.2
65-1249	72	19.9	-	80.1	-
65-1250	72	15.4	5.5	71.5	7.6
65-1251	72	17.6	3.2	75.8	3.3
65-1252	72	13.8	2.2	79.7	4.3

The discriminant functions have been computed for these three plants and the values are 2.098, 1.945 and 2.060, respectively. All these values are lower than the average discriminant function of T. maizar vs. T. pilosum which is  $\frac{(2.736 + 1.909)}{2} = 2.323$  (Table 3). It is evident that these plants are more closely related to T. pilosum than to T. maizar. Again, specimens 65-1239 and 65-1243 were tentatively assigned to the so called T. pilosum even though these plants have narrower blades and showed a resemblance to T. lanceolatum in their growth habit, because of the presence of pedicellate accessory male spikelets, a character common to T. pilosum. The discriminant functions of these specimens are 0.736 and 0.727 respectively. These values are higher than the average discriminant function (0.546) for T. pilosum vs. T. lanceolatum suggesting that these specimens are "nearer" to the so called T. pilosum. Since the so called T. pilosum and T. lanceolatum differ from each other by only about four standard deviations, morphological types of one "species" should be expected to resemble the other and vice versa.

A clear view of the degree of separation of the populations is achieved by plotting the discriminant functions using T. maizar-pilosum as the abscissa and T. pilosum-lanceolatum as the ordinate (Fig. 1). This graph shows that there is no overlapping between T. maizar and T. zopilotense, the two parental diploid species. This shows that the two species are very distinct. They occupy opposite corners of the graph. Members of the T. lanceolatum-pilosum complex overlap with each other as expected and lie in the center of the graph. The fact that the members of T. lanceolatum-pilosum complex are intermediate in the eight morphological characters that distinguish its supposed diploid parents and that they are distributed in the center of the graph is strong evidence that the T. lanceolatum-pilosum complex could have originated as a result of hybridization between T. maizar and T. zopilotense followed by chromosome doubling.

Detailed chromosome analysis has been made on 21 plants belonging to the T. lanceolatum-pilosum complex and the two diploid parental species. The diploid species showed regular pachytene pairing resulting in 18 bivalents at diakinesis and Metaphase I. The tetraploids showed



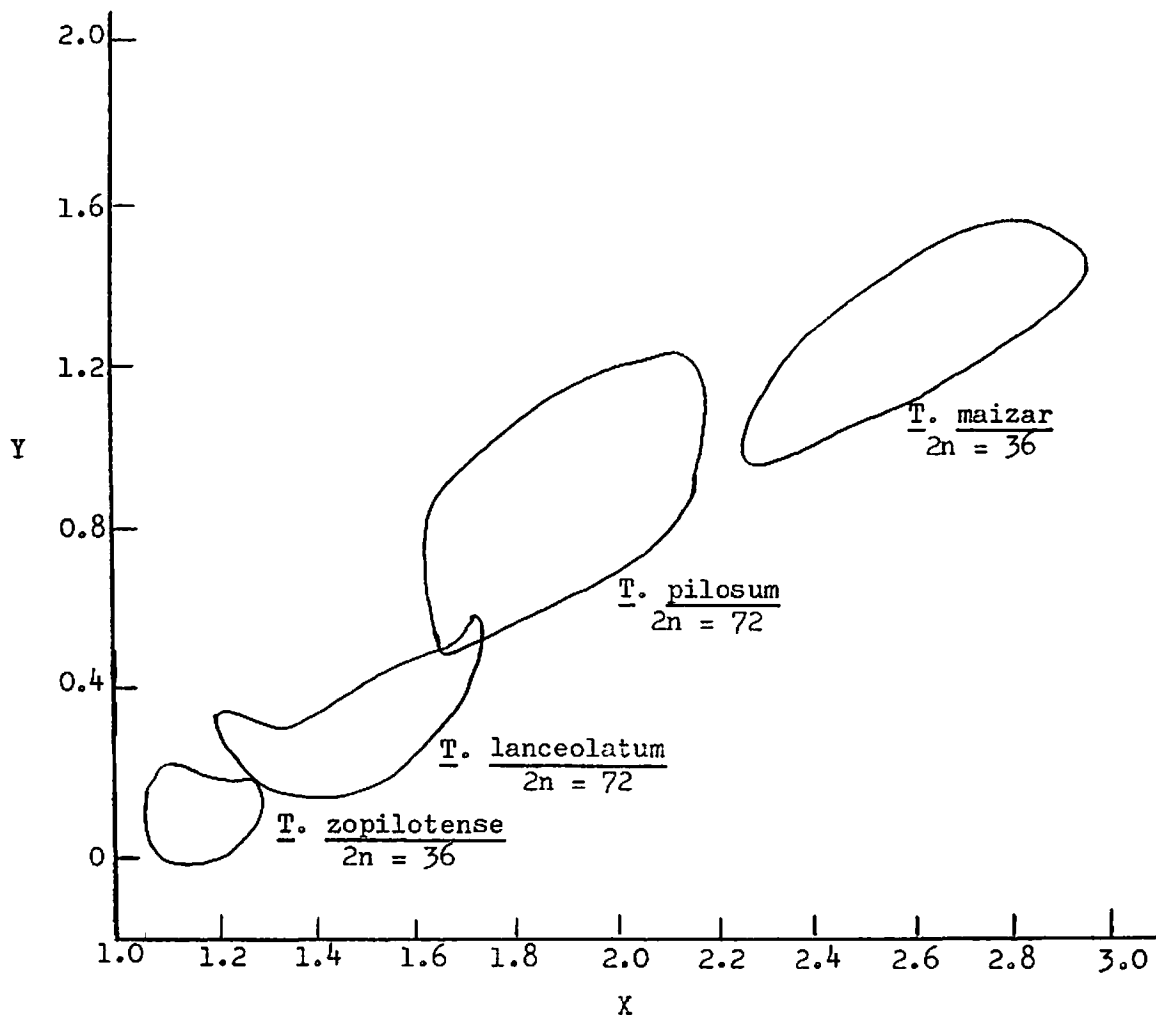


Figure 1. Graph showing the relative disposition of four populations of Tripsacum. The discriminant functions of T. maizar-pilosum are used as the abscissa while those of T. pilosum-lanceolatum are used as the ordinate.

"associations of four" chromosomes followed by exchange of partners at pachytene. At diakinesis varying numbers of multivalents, bivalents and univalents are observed (Table 4).

Of the ten possible types of quadrivalents (Darlington, 1931), those requiring multiple chiasma formation were observed in low frequencies. Types 11 and 17 were found in much higher frequencies (Table 5). Since exchange of partners takes place between "homeologous" chromosomes leading to the heterogenetic association of quadrivalents, there is

Table 5

Frequency distribution of the various types of quadrivalents observed at diakinesis in the T. lanceolatum-pilosum complex.

Plant number	Frequencies of quadrivalent types									
	11	12	13	14	15	16	17	18	19	20
F.T.G 65-1221	21.0	5.0	-	-	-	-	67.0	7.0	-	-
65-1235	38.6	9.5	-	-	6.9	-	37.4	7.6	-	-
65-1239	40.0	-	-	-	-	-	53.3	6.7	-	-
65-1240	16.4	9.1	3.6	-	12.7	-	52.7	5.5	-	-
65-1241	24.1	-	-	-	-	-	69.0	6.9	-	-
65-1242	26.9	19.2	20.5	-	-	-	32.1	1.3	-	-
65-1243	22.4	6.6	1.3	-	-	-	64.5	5.2	-	-
65-1243a	19.4	3.2	-	-	5.3	-	65.6	6.5	-	-
65-1247	24.3	-	3.0	-	-	-	66.7	6.0	-	-
65-1248	23.1	2.9	-	-	1.9	-	62.5	9.6	-	-
65-1249	17.4	-	-	-	-	-	67.4	15.2	-	-
65-1250	30.7	5.3	4.7	-	-	-	55.7	3.6	-	-
65-1251	26.2	10.2	-	-	3.4	-	52.3	7.9	-	-

segregation in these tetraploid "species" for the morphological characters that distinguish the diploid parents. It is the hybrid origin of these populations that makes them so variable.

Even though the names T. pilosum and T. lanceolatum may be used for the purposes of classifying a given specimen, in view of the presence of a wide range of morphological characters and the small D value between the so called T. pilosum and T. lanceolatum it seems better to regard the whole group of tetraploids as a T. lanceolatum complex which includes a wide range of morphological forms.

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1. Electron microscopy of the chlamydospores of corn smut (Ustilago maydis (DC.) Corda).\*

This report is based on our preliminary observations with carbon replicas of the chlamydospores of corn smut at the ultrastructural level. The replication method employed here is a methyl-methacrylate heat-pressure technique. The technique has been slightly improved by us in order to use this method in fungal palynology.

The chlamydospores suspended in water are evenly spread on the flat surface of a prepolymerized, methacrylate plastic plate (about 15 by 20 mm in size). The preparations are then allowed to air-dry overnight in a clean chamber. Further the plate, with a thin layer of dried spores, is sandwiched between glass plates, held tightly with clamps and heated to about 105 degrees C for ca. 15 to 20 minutes. After cooling to room

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\*This work was partly done at the Biology Department, Yale University, New Haven, Conn.