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REGULAR ARTICLE

Sunflower (*Helianthus annuus* L.) germplasm evaluation for drought tolerance

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ABSTRACT

Future climate changes are expected to increase risks of drought, which already represent the most common stress factor for sunflower (*Helianthus annuus* L.) production throughout the world. It is important, therefore, to evaluate genotypes for this stress. Our objective was to study yield and yield-related traits under irrigated and drought conditions in 56 sunflower genotypes of different origin and growth habit. A wide range of intraspecific genetic variability was present in sunflower, which could be used to develop new genotypes, more adapted to drought conditions. The highest level of tolerance was present in local genotypes. Among restorers, the highest level of tolerance was present in RL-57 (Pakistan), whereas an exotic restorer F-Yu-82 (Spain) showed the highest yield, along with high drought susceptibility index. Inbred line ORI-9/B (Pakistan) was identified as the most tolerant line combined with low yield potential, whereas AMES-10107 and AMES-10103 (China) were found to be moderately drought-tolerant lines with highest yield. Selection among segregating progeny from hybridization among lines with good drought tolerance with lines of good yield potential may lead to the development of superior inbred lines.

Key Words: *sunflower; drought; yield; drought susceptibility index.*

INTRODUCTION

Drought stress is a worldwide production constraint of sunflower (Drgovic and Maksimovic, 1995). According to a report by USDA Agriculture Weather Facility (2005), oilseed production in 2005 was down 2% from 2004 due to drier than normal growing season. In Spain in particular, the sunflower crop suffered substantially from drought, decreasing production by 41%. Similarly in the Americas, drought was a key factor responsible for yield losses of up to 20% (Reddy et al., 2004). In Pakistan, sunflower acreage declined by 25% from 1998-99 to 2002-03, but the total sunflower production declined by 33% during the same period as a result of severe drought (GOP, 2003).

As most of the cultivated hybrids evolved under optimum conditions, breeding for drought tolerance is required. This indeed would depend on the presence of diverse germplasm so that potential sources of drought tolerance might be identified and subsequently used to assure high yield when drought occurs. High yield is the ultimate objective of any breeding program. However, high yield and drought tolerance are two different mechanisms that are often found to oppose each other. Traits, such as small plant size, reduced leaf area, and prolonged stomata closure, allow plant to limit water losses, but also lead to reduced dry matter production and, therefore, reduced final yield (Karamanos and Papatheohari, 1999; Fischer and Wood, 1979).

Traits related to drought tolerance and to high yield potential should be a priority in crop breeding programs for a target area and specific type of stress. Yield in low and high yielding environments is considered by some to be separate traits, which are not necessarily maximized by identical sets of alleles (Falconer, 1990). Consequently, breeding strategies should be different when targeting stress and non-stress environments (Ceccarelli et al., 1991; Ceccarelli et al., 1998). Other authors claim that selection under favorable conditions is required to select genotypes with good performance under both stress and non-stress conditions (Cattivelli et al., 1994; Sayre et al., 1995; Braun et al., 1997). There is some agreement that a high yield potential is advantageous under moderate stress, whereas cultivars with low yield potential and high drought tolerance may be useful when stress is severe, as would be the case if the precipitation were less than 300 mm (Voltas et al., 1999; Panthuwan et al., 2002).

Field selection is complicated by the high variability associated with multiple interactions contributing to drought tolerance of crops, as drought events occurr at different phases during the growing season or the spatial variability, which is amplified when water is limiting. This contributes to a large genotype × environment (GE) interaction, which may explain the slow progress in developing new cultivars of crops for drought conditions (Fukai et al., 1999). In most cases, no clear cause of the GE interaction has been identified because of lack of information about the environment (such as weather or soil) or the genotypes themselves (Voltas et al., 2002). Several indices have been proposed to describe the behavior of a given genotype under stress and non-stress conditions (Finlay and Wilkinson, 1963; Fischer and Maurer, 1978; Soika et al., 1981; Bidinger et al., 1987; Lin and Binns, 1988; Yadav and Bhatnagar, 2001).

In the present work, we evaluated yield and yield-related traits under irrigated and drought conditions in sunflower restorer and inbred lines. Drought was imposed during the reproductive stage, when a gradual rise in temperature is associated with a severe depletion of soil water resources.

MATERIALS AND METHODS

Field experiments were conducted during February 2006 at the sunflower experiment field of the Department of Plant Breeding & Genetics, University of Agriculture, Faisalabad, under irrigated and drought conditions. The soil was a sandy loam type, with low water retention capacity. The laboratory-measured field capacity (–25 KPa) and wilting point (–1800 KPa) of the soil averaged 35% and 18% by volume, respectively, pH 7.5, organic matter 0.91%, available phosphorous 28.6 ppm, and potassium 140 ppm. The plots were fertilized with 150 kg N ha^{-1} and 50 kg P ha^{-1}; no K was applied. Fifty-six genotypes of sunflower—20 restorer and 36 inbred lines—from different countries were used (Table 1). A split-plot restriction on randomization within a randomized block design (r =3) was used, where levels of water availability were assigned to main plots and genotypes to subplots.

Each subplot was 4.8 m wide and 6 m long, consisting of eight rows of a single genotype. The inter-row spacing was 60 cm and interplant spacing was 30 cm.

Table 1. A list of sunflower restorer and inbred lines and their countries of origin.

No.	Restorers	Country	No.	Inbred line	Country	No.	Inbred line	Country
1.	CM–621	Canada	1.	B–FS–88	Spain	21.	ORI–10/B	Pakistan
2.	CM–619	Canada	2.	CM–614	Canada	22.	ORI–25/B	Pakistan
3.	CM–815	Canada	3.	B–Sin–82	Spain	23.	ORI–26/B	Pakistan
4.	F–Yu–82	Spain	4.	PEM S–R88	Spain	24.	ORI–23/B	Pakistan
5.	R–Sin–82	Spain	5.	HA–350	USA	25.	ORI–24/B	Pakistan
6.	R–FSS–88	Spain	6.	HA–341	USA	26.	ORI–2/B	Pakistan
7.	CM–630	Canada	7.	HA–342	USA	27.	ORI–1/B	Pakistan
8.	CM–631	Canada	8.	Ames–10103	China	28.	ORI–19/B	Pakistan
9.	CM–632	Canada	9.	Ames–10107	China	29.	ORI–11/B	Pakistan
10.	RL–84	Pakistan	10.	CM–612	Canada	30.	ORI–9/B	Pakistan
11.	RL–64	Pakistan	11.	HA–G–P–13	USA	31.	ORI–27/B	Pakistan
12.	RL–37	Pakistan	12.	DM–2	USA	32.	ORI–28/B	Pakistan
13.	RL–46	Pakistan	13.	CM–628	Canada	33.	ORI–22/B	Pakistan
14.	RL–13	Pakistan	14.	HA–407	USA	34.	ORI–16/B	Pakistan
15.	RL–39	Pakistan	15.	HA–207	USA	35.	ORI–14/B	Pakistan
16.	RL–57	Pakistan	16.	HA–310	USA	36.	ORI–15/B	Pakistan
17.	RL–52	Pakistan	17.	ORI–13/B	Pakistan	–	–	–
18.	RHA 366	USA	18.	ORI–18/B	Pakistan	–	–	–
19.	RHA 375	USA	19.	ORI–21/B	Pakistan	–	–	–
20.	RHA 389	USA	20.	ORI–12/B	Pakistan	–	–	–

The most important aspect of screening for drought tolerance is the stage of plant growth at which stress occurs. Previous studies showed that greatest reduction in yield occurred when stress was applied at the flowering stage (Kirda et al., 1999; Unger, 1982). Similarly, moisture stress during the achene-filling stage resulted in a reduction of more than 70% in achene yield of sunflower, whereas the lowest reduction in achene yield was observed when moisture stress occurred during the vegetative stage (Jana et al., 1982).

Keeping the same in mind, the non-stress regime was irrigated during the entire growth cycle to maintain the soil water content close to field capacity (irrigated regime). In the stress regime, the plots were irrigated at the same time and with the same amount of supplemental water as in the non-stress regime during the vegetative phase; supplemental irrigation was completely withheld beginning with the button stage (R1) to achieve low soil moisture content during anthesis (drought regime). The soil moisture content was measured every 8-10 days (Figure 1). The total rainfall during crop growth cycle was only 75.6 mm, of which 51.6 mm fell during the vegetative phase; 20 mm fell very late during the reproductive phase (Figure 2). Leaf diseases were not present, and weeds were controlled manually.

DROUGHT SUSCEPTIBILITY INDEX

Drought susceptibility index (S) was calculated according to Fischer and Maurer (1978):

$$S = \frac{1 - Y/Y_p}{1 - X_d/X_p},$$

where Y is the achene yield per head of a given genotype under drought, Y_p is the achene yield per head of the same genotype under irrigation, X_d is the mean achene yield of all genotypes within group (restorer or inbred) under drought, and X_p is the achene yield per head of all genotypes within group under irrigation. The quantity $1–X_d/X_p$ is the drought intensity (Fischer and Maurer, 1978) with a range of 0 to 1.

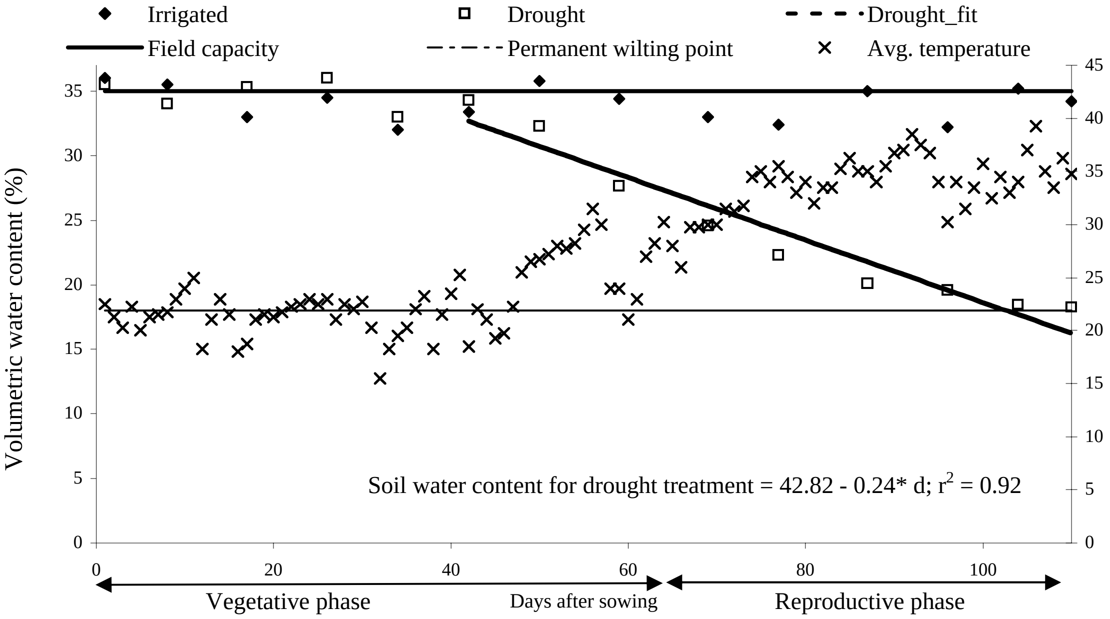

Figure 1. Volumetric water content for sunflower restorer and inbred lines irrigated for either the entire growing season (Irrigated) or only during vegetative development (Drought) plus average air temperature during the entire season. As a reference, field capacity (35% v/v) and permanent wilting point (18% v/v) are given as horizontal lines.

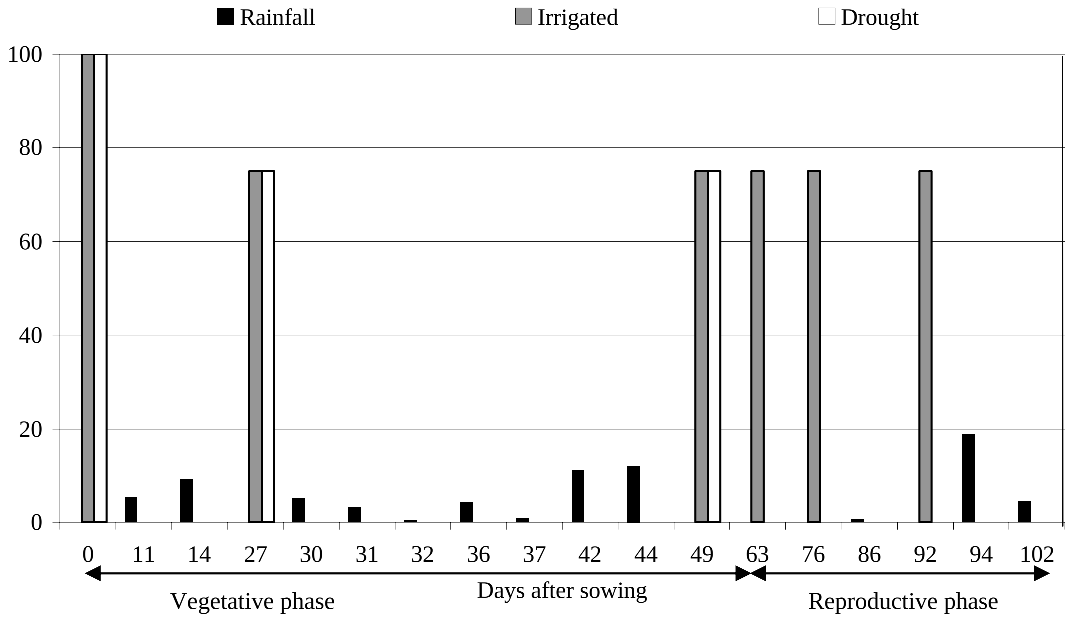

Figure 2. Natural rainfall and water applied through supplemental irrigation for sunflower restorer and inbred lines for either the entire growing season (Irrigated) or only during vegetative development (Drought).

EVALUATION OF YIELD AND YIELD-RELATED PARAMETERS

At maturity, the following characters were measured: head diameter, achene yield per head, mass of 100 randomly selected achenes (henceforth referred to as average achene mass), and number of achenes per head on middle four rows; the measurements were made eliminating two rows at each side, within single plot and on eight competitive, i.e., surrounded by intact plants spaced at specified plant population density, plants per genotype and replicate within a single row, thereby eliminating plants at each end. Heads were harvested separately from each plant per genotype and replicate and achenes removed manually from dried receptacles and measured. The number of achenes per head was calculated from total achene weight per head and average achene mass.

STATISTICAL ANALYSIS

Data were analyzed using analysis of variance (ANOVA). Water availability was a treatment factor (W) with the two levels, irrigation and drought. Genotype was a treatment factor (G) with 56 levels, and consisted of 20 restorer (R) and 36 inbred lines (L). The sum of squares for genotypes and water availability × genotype were partitioned into sum of squares due to restorers, sum of squares due to inbred lines, and the linear contrasts between restorers and inbred lines. Associations of yield and its related traits with susceptibility index were examined using phenotypic correlations with a microcomputer statistical program (MSTAT-C Development Team, 1989)

RESULTS

Highly significant differences ($P \leq 0.01$) for genotypes overall, among restorers, and among inbred lines were observed for all measured traits (Table 2). The contrast between restorers and inbred lines was also highly significant for the same traits. Genotypes, restorers and inbred lines showed highly significant ($P \leq 0.01$) interactions with water levels for all traits, except average achene mass for restorers. Similar to main effects, the interaction contrasts were also highly significant.

Table 2. Mean squares from the analysis of variance for 20 restorer and 36 inbred sunflower lines evaluated for yield-related traits under two water levels, either irrigated for the entire growing season or only during vegetative development.

Source	df	Head diameter	Achene		
			Yield per head	Mass of 100	No. per head
Replication	2	0.3	2.0	0.01	3029
Water level (W)	1	456.9**	5193.1**	6.00**	1977658**
Error$_a$	2	0.9	0.3	0.11	329
Genotypes (G)	55	44.4**	2051.9**	38.32**	362908**
Restorers (R)	19	43.0**	351.4**	9.77**	438348**
Lines (L)	35	46.3**	2901.7**	53.84**	322361**
R vs. L	1	7.4*	4617.8**	37.95**	348681**
W*G	55	5.3**	120.1**	0.26**	32647**
W*R	19	2.7**	42.6**	0.02	30858**
W*L	35	6.5**	159.2**	0.37**	33170**
W*R vs. W*L	1	9.4**	222.8*	0.96**	48366**
Error$_b$	220	0.4	2.0	0.03	1488

** Significant at 0.01 probability level.

Achene yield per head for restorer lines was plotted against the drought susceptibility index (Figure 3a). Segment I comprises the most desirable group of genotypes characterized by low drought susceptibility index and a high achene yield per head. Restorer RL-57 (No. 16) was the most drought-tolerant restorer, but restorer RL-52 (17) gave the highest yield in this segment. Segment III includes genotypes with a high drought susceptibility index and high achene yield per head. In this segment, RHA-375 (19) was the restorer with the highest drought susceptibility index and restorer F-Yu-82 (4) had the highest achene per head.

Achene yield per head for inbred lines was plotted against the drought susceptibility index (Figure 3b).

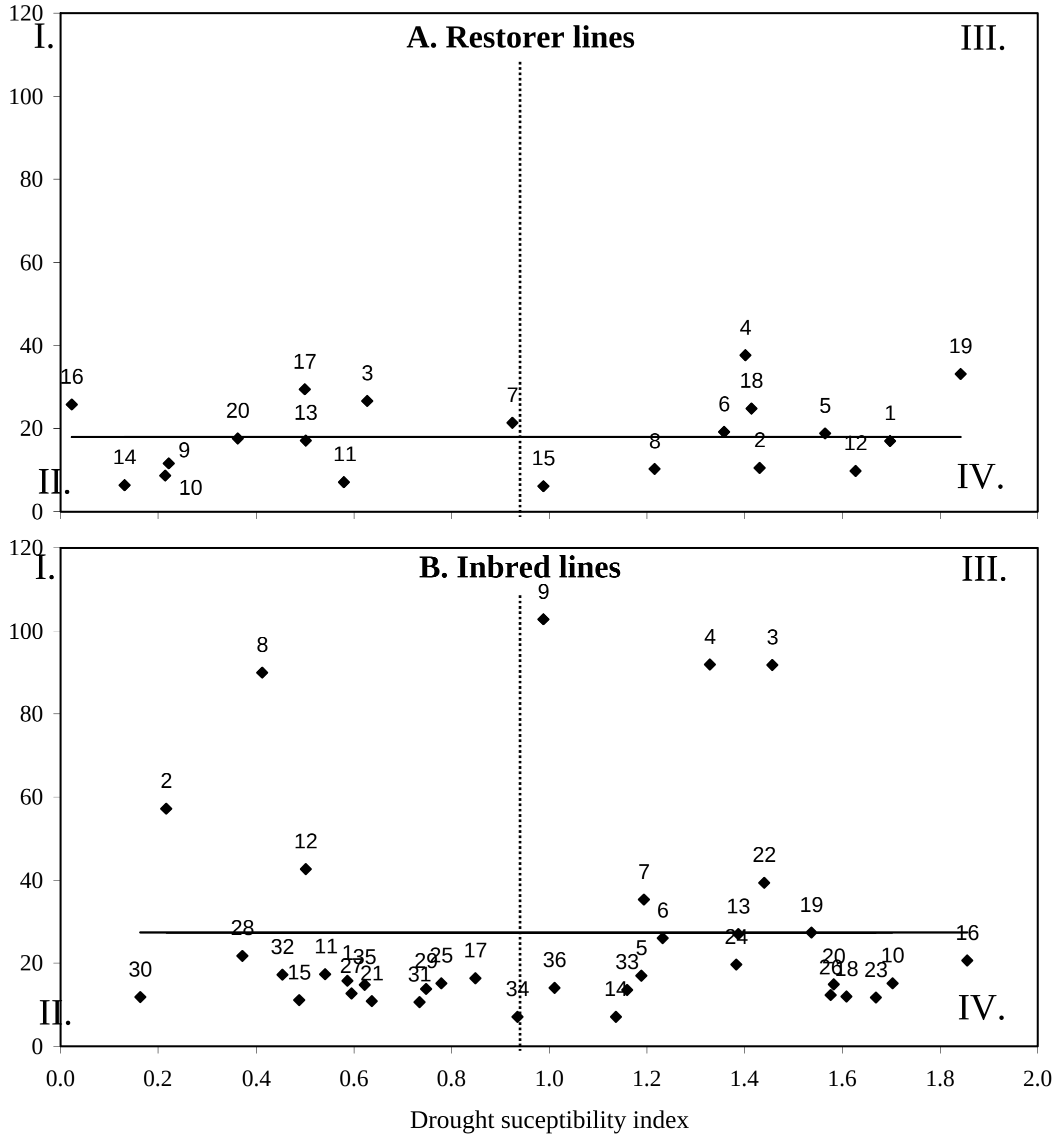

Figure. 3. Relationship between achene yield per head and the drought susceptibility index for restorer lines (top panel) and inbred lines (bottom panel. The dotted vertical line represents the average drought susceptibility and the solid horizontal lines the average achene yield per head within either restorer or inbred lines. The numbers provide the link to a particular line as listed in Table 1.

Two lines in segment I with high achene yield per head were AMES-10107 (9) and AMES-10103 (8), although the latter had a lower drought susceptibility index. Other lines included in this group were CIM-614 (2) and DM-2 (12). Line ORI 9/B (30), located in segment II, had the lowest drought susceptibility index of all inbred lines evaluated, but also had one of the lowest achene yields per head. Line HA-310 (16) in segment IV showed the highest drought susceptibility index, but was low yielding.

Yield and yield-related traits under irrigated conditions were not significantly correlated ($P > 0.05$) with the drought susceptibility index, while showing significant negative correlation ($P \leq 0.05$) under drought during the reproductive phase, except for average achene mass (Table 3). The correlations among yield-related traits were similar in both water regimes, but their magnitude was smaller under drought. All significant correlations ($P \leq 0.05$) between yield and yield-related traits were positive.

Table 3. Phenotypic correlations among traits for genotypes under irrigation (above diagonal) and under drought conditions during the reproductive phase of development (below diagonal).

	Head diameter	Achene			Drought susceptibility index
		Yield per head	Mass of 100	No. per head	
Head diameter		0.73**	0.56**	0.62**	–0.13
Achene yield per head	0.42*		0.77**	0.70**	0.16
Mass of 100 achenes	0.32	0.73**		–0.18	0.12
Achene no. per head	0.50**	0.74**	–0.16		0.20
Drought susc. index	–0.58**	–0.39*	0.14	–0.57**	

*, ** Significant at 0.05 and 0.01 probability level, respectively.

DISCUSSION

Diverse germplasm from several countries has been screened for drought tolerance in many crop species, such as common bean (Phaseolus vulgaris L.), for which several hundred germplasm accessions, breeding lines, and cultivars of diverse origins were systematically screened for drought tolerance (Laing et al., 1983; Singh and Terán, 1995). However, as reported in this study, the highest level of drought tolerance in those studies was found in locally adapted genotypes. For restorers, the highest level of drought tolerance was present in genotype RL-57 (Pakistan). Because of their evolution under harsh conditions characterized by low precipitation and high temperature, it was expected that local genotypes might possess some degree of drought tolerance.

Similarly, local line ORI-9/B (Pakistan) was identified as the most drought-tolerant genotype, but it was very low yielding. Plant breeders are mainly interested in lines that combine drought tolerance with high yield, which might suggest wide adaptation possibilities (Gomez et al., 1998). Genotypes AMES-10107 and 10103, both originating from China, had the highest yield with a moderate level of drought tolerance. Crossing ORI-9/B and AMES 10107, for example, could combine drought tolerance of the former with high yield of the latter. The line HA-310 (USA) proved to be a highly drought-susceptible line. It is important to understand the physiological mechanism of drought tolerance and susceptibility. Therefore, a detailed study of physiological parameters must be carried out in highly susceptible and tolerant lines.

Yield under irrigation did not show any correlation with drought susceptibility index. This indicated that yield under irrigated conditions could not be considered a reflection of its yield under drought condition and that yield under irrigated conditions should not be used as a selection criterion to improve yield under drought (Baldini et al., 1992). Therefore, yield in low and high yielding environments should be considered as two separate traits that are not necessarily maximized by identical sets of alleles (Falconer, 1990). Consequently, plant breeding strategies should be different when targeting stress and non-stress environments (Ceccarelli et al., 1991; Ceccarelli et al., 1998). However, there was significant negative correlation between achene yield per head and the drought susceptibility under drought conditions, indicating that the drought susceptibility index might be a very useful selection criterion for drought-tolerance breeding in sunflower (Fereres et al. 1986; Baldini et al. 1992), as suggested by Fisher and Maurer (1978) for wheat.

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