Approaches to Breeding Programs and Genomics Studies Aiming Tropical Maize Characterization for Water Stress and Drought Tolerance in Brazil.

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Introduction

Phenotyping, or the characterization of genetic resources for diverse attributes, has been an essential component of breeding programs and now is also becoming a key complement for genotyping in molecular breeding investigation. The plant response for drought stress is associated with its internal content of water. Although the effects of those stresses are known, the significant practical results are a few. The adaptation of plants to adverse environments, or situations under sub-optimum environment factors, involves the adaptation to multiple stresses, with direct and indirect interactions. The elucidation of physiological, biochemical and molecular mechanisms might boost the process of generation of new genetic materials, besides contributing for the development of selection techniques that could reduce the time and the work for evaluation of genetic sources for tolerance to abiotic stresses. The present work is related with tropical germplasm characterization for water stress and it summarizes the effort in the development of promising genetic material for maize improvement for drought tolerance in Brazil.

Material and Methods

The methodologies involve the knowledge of strategies in the definition of appropriate genetic degree, related to identification and description of secondary morph-physiological characteristics and grain yield (GY) as useful parameters for the phenotyping, and as potential use in a breeding program with MAS techniques. The preliminary phenotyping strategy starts with the identification of selfed maize progenies with multiple stresses tolerance through recurrent selection breeding scheme. The strategy is the direct selection for drought stress tolerance and indirectly for increasing P and N efficiency, aiming the development of heterotic and productive lines with multiple stresses characteristics, for hybrid production and genomics studies. Also, from a synthetic elite flint composed based mainly in the parameters anthesis-silking interval (ASI) and GY, under water stress environments. Complementary, an intermediate phenotyping strategy has studied the parameters: prolificacy, yield and theirs components, leaf rolling, and leaf senescence. These parameters will be useful to amplify the contrasting characterization of elite lines, and to identify possible mechanisms involved in drought tolerance. In this phase is demonstrated as the efforts concentrated in a flint maize synthetic and in the production of inbred lines, hybrids and segregating families (RIL's) from selected contrasting lines for the traits ASI and GY, evaluated under water stress. In the advanced phenotyping strategy it is included improved genotypes identified in the previous phases and from the research collaborative

programs. Also, in this step, are evaluated other traits to confirm the contrasting genotypes and theirs performance under drought, e.g., lipids peroxidation and antioxidant stress enzymes, as well as, chlorophyll fluorescence, looking for the conventional improvement and genomics approaches. Site-Specific Experimental: Sete Lagoas and Janaúba, MG, Brazil.

Results and Discussion

Since the middle of 1990, maize breeders at the Embrapa Maize and Sorghum Research Center have started a drought tolerance breeding program using adequated source populations like BR 105 as a tropical, early cycle and flint type maize variety. Hundreds of selfed progenies were evaluate each winter for drought tolerance; selection was mainly based on an efficient phenotypic index, the anthesis-silking interval (ASI), for water stress tolerance. After several cycles of inbreeding selections using the ASI parameter, two groups of lines were selected with no tolerance and tolerance to water stress conditions, using Preliminary Phenotyping Protocols (**Figure 1** and **Table 1**).

For one study, six maize inbred lines were chosen to represent a group of tolerant and non-tolerant lines to drought stress (Durães et al., 1999; 2000a; 2000b; 2002). The aim of this study was to evaluate the genetic potential for grain yield of six maize inbred lines derived from a flint variety BR 105, by comparing the F_1´s and reciprocal effects, produced form a diallel set of crosses under water stress condition. The results and conclusions were: 1) Some of the single crosses were as productive as the check hybrids in the stress environment indicating the good potential of the selected lines for hybrid development; 2) The line L6.1.1 had the highest GCA effect, while lines L1147 and L10.1.1 had low GCA effects for grain yield in both experiments; 3) SCA mean square showed no significant difference for the hybrids in the experiment with stress; 4) Hybrids L1147 x L10.1.1 and L1170 x L8.3.1 presented the highest SCA effects in the non-stress and stress environment, respectively; 5) The checks grain yield means were greater than the means of the hybrids in both environments.

The state of art of the maize improvement program for drought (Durães et al. 2005):
1. Lines selected for ASI (Anthesis-Silking Interval) and GY were originated from the flint maize Synthetic, using Preliminary Phenotyping Protocols;
2. After 3 generations of recombination, selfed progenies were extracted from this Synthetic with selection for ASI in the irrigated area of Embrapa.
3. The flint maize Synthetic was formed with lines in S_4 and S_5 that presented low ASI, and to be released shortly.
4. Lines: It is been worked a group of 80 originated lines of this improved Synthetic.
5. Intermediate and Advanced Phenotyping Protocols: 12 inbred lines S_8 were characterized and segregating families from 2 contrasting lines to ASI, GY and others traits were used for QTL studies (**Figures 2** and **3**).
6. Hybrids (Field tests to ASI x Grain Yield under different water regimes): It were included SC, TWC and DC hybrids with 50% ASI and 100% ASI and varied sources of lines. Also, it were included some hybrid without ASI (commercial tester) and promising hybrid of the breeding program. The results showed that the characteristic ASI has manifested in a more accentuated way in the hybrid selected for ASI and GY.

Obtention of maize selfed progenies with tolerance to multiple stresses through recurrent selection: Schematic design (Gama et al. 2002)

Synthetic (P and N) => Self progenies S1 => Evaluation S1 under drought => Recombination of selected progenies (low P and N) => C1 => Self progenies S1 => Evaluation S1 => Recombination => C2 => and so forth ...

The synthetics were made by using inbred lines introduced from collaborative and Embrapa breeding program, using a recurrent selection with selfing progenies. The future step will test the tolerant lines to multiple stresses in combination with other heterotic lines for hybrid production and to release the synthetic for being use in the research program and in small-scale farming.

Conclusions

The Embrapa Maize and Sorghum has dedicated action strategic in its improvement program, considering preliminary, intermediate and advanced phenotyping protocols. These approaches comprise various germplasm sources with their diverse genetic background, focus on the development of tolerant plants to multiple stresses. The results available about eighty contrasting maize lines selected mainly for ASI and GY; and, also a segregating families (RILs) and two inbred lines groups characterized under different water regimes.

References

Gama, E.E.G.; Durães, F.O.M.; Santos, M.X. dos. Tropical maize synthetics improvement for moisture-stress tolerance for small-scale farmers. **In:** 7th Eastern and Southern Africa Regional Maize Conference and Symposium on Low-Nitrogen and Drought Tolerance in Maize. Nairobi, Kenya, 11-15 February 2002. pp. 288-291.

Durães, F.O.M.; Machado, R.A.F.; Magalhães, P.C.; Santos, M.X.dos; Silva, R. and Molina, M. 1999. Adaptação de milho às condições de seca: 1. Caracterização de genótipos contrastantes quanto ao parâmetro fenotípico IFMF. **In:** Congresso Brasileiro de Fisiologia Vegetal, 5th, Resumos ..., Brasília, 1999. SBFV, Brasília, DF.

Durães, F.O.M.; Magalhães, P.C.; Ferrer, J.L.R.; and Machado, R.A.F. 2000a. Adaptação de milho às condições de seca: 2. Florescimento e maturidade fisiológica de sementes de linhagens contrastantes para o parâmetro fenotípico IFMF. **In:** Congresso Nacional de Milho e Sorgo, 23rd, Resumos ..., Uberlândia, 2000. ABMS, Uberlândia, MG.

Durães, F.O.M.; Santos, M.X.; Paiva, E.; Couto, L.; Oliveira, A.C. 2000b. Estratégia de melhoramento de milho visando tolerância a seca. **In:** Congresso Nacional de Milho e Sorgo, 23rd, Resumos ..., Uberlândia, 2000. ABMS, Uberlândia, MG.

Durães, F.O.M.; Magalhães, P.C.; Oliveira, A.C.; Santos, M.X.; Gomes, E.E.G.; Guimarães, C.T. Combining ability of tropical maize inbred lines under drought stress conditions. **Crop Breeding and Applied Biotechnology**, v.2, n.2, p. 291-298, 2002.

Durães, F.O.M.; Gama, E.E.G.; Gomide, R.L.; Albuquerque, P.E.P.; Andrade, C.T.L.; Guimarães, C.T.; Magalhães, J.V. Phenotyping maize for drought response in Brazilian tropical areas: Approaches to breeding programs and genomics studies. p.7.09. **In:** InterDrought II. Rome, Italy, September 24-28, 2005.

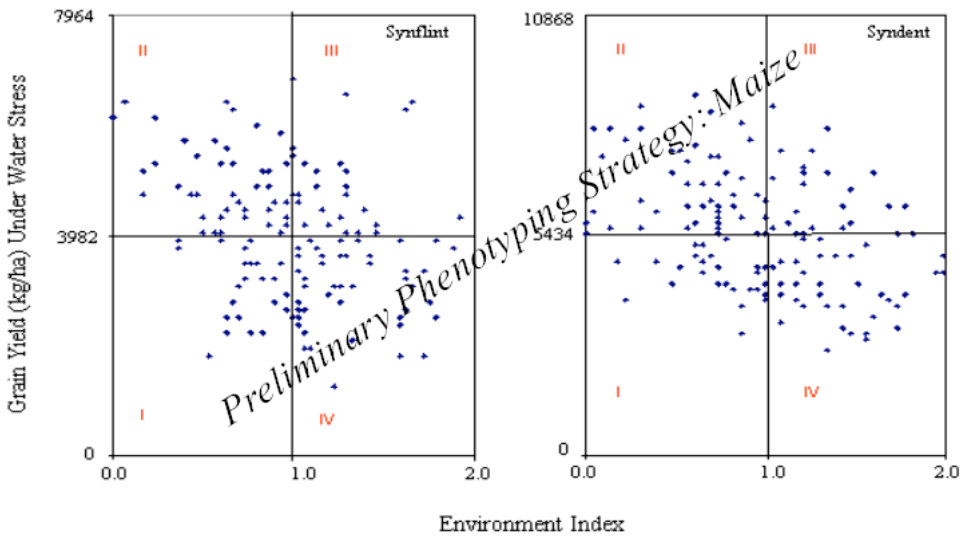

Figure 1. Ear yield as function of *environment index* [*or, Water Stress Index, WSI* = $\mathbf{(Y_{ww}-Y_{ws})}$ / $\mathbf{(Ya_{ww}-Ya_{ws})}$] for 144 S_2 progenies trials each across two replications from 2 tropical maize synthetics under two water regimes at Janauba, Brazil. (Gama et al., 2002).

Genotype		Background		Characteristic
Sigla	**Name**	**Inbreeding/Cycle**	**Genetics**	
		(Parental lines/F1´s and reciprocals)		
L1	L1170	S8	inbred	High ASI*
L2	L1147	S8	inbred	High ASI
L3	L13.1.2	S8	inbred	Low ASI
L4	L6.1.1	S8	inbred	Low ASI
L5	L10.1.1	S8	inbred	Low ASI
L6	L8.3.1	S8	inbred	Low ASI
		(Hybrid check)		
C4	HS93H	Early maturity	SC	Experimental SC
C5	BRS3060	Early	TWC	Commercial TWC
C6	BRS2114	Early	DC	Commercial DC

* ASI (Anthesis-silking interval, in days)

Table 1. Characteristics of the inbred lines and check hybrids of maize. Embrapa Maize and Sorghum, Brazil. (Durães et al., 2000b)

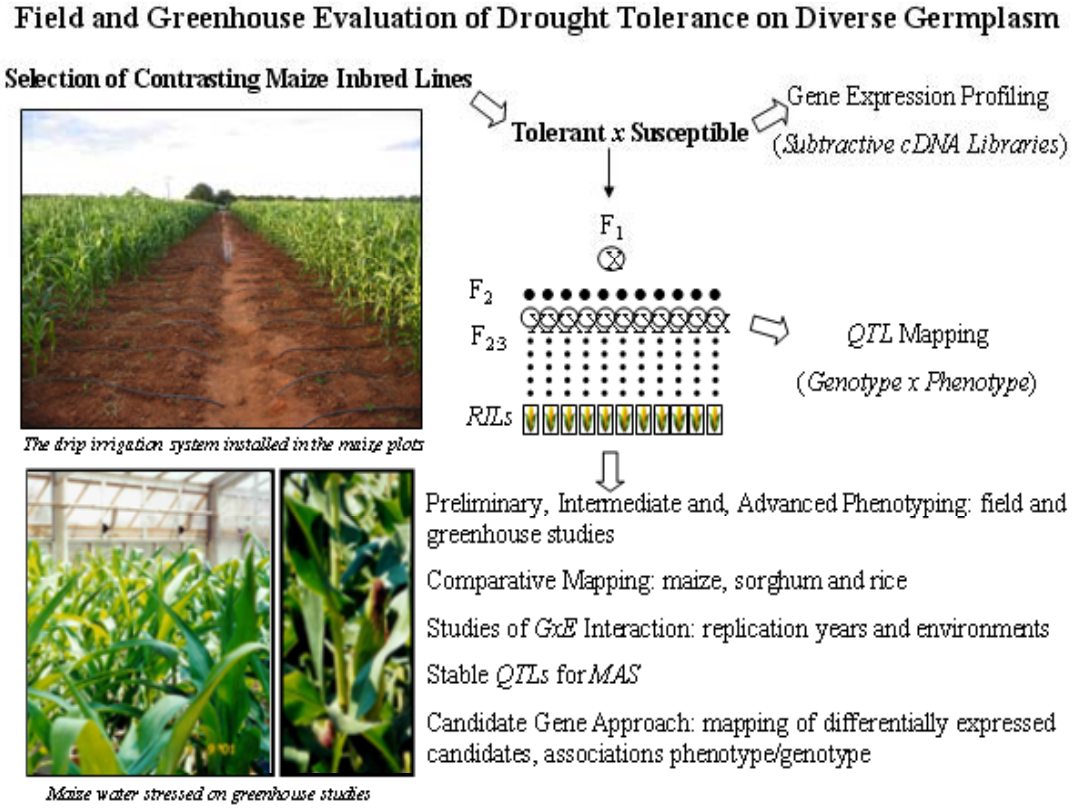

Figure 2. Schematic procedures to segregation populations production of contrastants inbred lines to ASI, with objectives of grain yield and drought tolerance. (Durães et al., 2005)

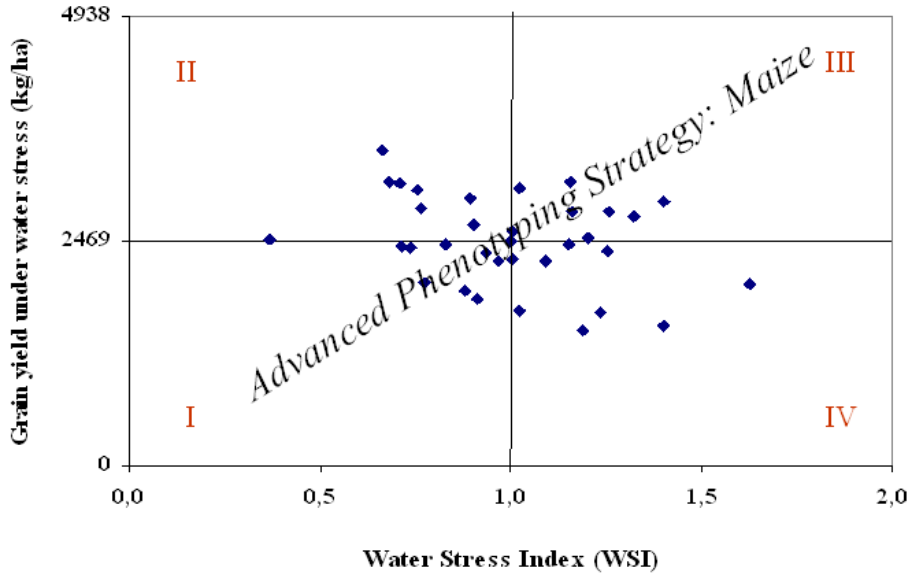

Figure 3. Mean grain yield (kg/ha, at 13% moisture) of 3 hybrid checks and 30 maize F_1's, evaluated in two experiments with and without water stress. (Durães et al., 2002)