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The evaluation of photosynthetic parameters in maize inbred lines subjected to water deficiency: Can these parameters be used for the prediction of performance of hybrid progeny?

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Abstract

The response of selected photosynthetic and morphological parameters of plants to drought was examined in 5 inbred lines of maize (*Zea mays* L.) and their 10 F1 hybrids. The aim of the study was to establish whether the photosynthetic performance of parental genotypes under drought conditions correlates with the performance of their progeny and whether the net photosynthetic rate, the chlorophyll fluorescence parameters or the content of photosynthetic pigments could be used as reliable physiological markers for early breeding generations. The relative importance of the additive and the nonadditive (dominance, maternal) genetic effects in the inheritance of these parameters was also assessed by means of the quantitative genetics analysis. The results showed that the nonadditive genetic effects associated with a particular combination of genotypes or a particular direction of crossing are at least equally and often even more important as the additivity and that these genetic effects almost totally change with the exposure of plants to drought conditions. This was reflected in the inability to predict the response of F1 hybrids to drought on the basis of the photosynthetic performance of their parents, which indicates that the practical usability of such parameters in maize breeding programs is rather limited.

Additional keywords: additivity; chlorophyll fluorescence; dominance; drought; genetic analysis; maternal effects; photosynthesis; stress tolerance.

Introduction

Maize (*Zea mays* L.) is susceptible to various abiotic and biotic stress factors including drought. The economic losses in maize production due to this stressor are quite substantial and breeding for the improved drought tolerance is thus one of the most important tasks maize breeders are currently confronted with. Current strategies for the improvement of maize drought tolerance try to employ newer, genomics-related tools (for reviews, *see e.g.* Bruce *et al.* 2002, Tuberosa *et al.* 2002, 2007; Campos *et al.* 2004, Parry *et al.* 2005, Bäzinger and Araus 2007, Ribaut and Ragot 2007, Mullet 2009). Unfortunately, these approaches still suffer from some shortcomings. Experiments with gene manipulations and production of transgenic plants with better response to

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Abbreviations: *ANOVA* – analysis of variance; Cars – carotenoids; Chl – chlorophyll; *E* – transpiration rate; F_0 – minimum chlorophyll fluorescence; F1 – the first filial generation; F_m – maximum chlorophyll fluorescence; F_v/F_m – maximum quantum efficiency of photosystem II; g_s – stomatal conductance; LSD – least significant difference; P_N – net photosynthetic rate; PAR – photosynthetically active radiation; PS – photosystem; QTL – quantitative trait loci; Rubisco – ribulose-1,5-bisphosphate carboxylase/oxygenase; RuBP – ribulose-1,5-bisphosphate; RWC – relative water content; SLM – specific leaf mass; WUE – water-use efficiency.
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water deficiency were mostly undertaken in model plant species such as *Arabidopsis* or tobacco and only rarely have been transferred to crop plants. The identification of any quantitative trait loci (QTL) for a particular trait strongly depends on the genetic background and epistatic effects can be strong source of variation for traits that affect plant tolerance to stress factors; moreover, QTLs associated with stress tolerance often show low heritability. There is also the question of the reception of transgenic crops by the general public and of the economic benefits associated with the incorporation of a particular molecular marker or new technology into breeding program (Brennan and Martin 2007). This means that the original expectations of a significant improvement in maize drought tolerance, thought to occur with the advent of modern biotechnology techniques, have yet to be fulfilled.

An alternative approach to improve plant breeding for the better stress tolerance is the introduction of physiological selection markers. The physiological traits used in breeding for plant stress tolerance should show a good correlation both to tolerance/sensitivity to the target stress factor and to yield parameters relevant for the respective crop species, an adequate genetic variation in the evaluated population/genotype collection, and a high heritability and repeatability (Sayar *et al.* 2008). Among physiological parameters that are the most frequently proposed as the indirect selection markers, parameters associated with photosynthesis play undoubtedly the main role. It is not surprising, given that the photosynthetic apparatus responds very rapidly to the majority of stress factors that plants have to cope with (recently reviewed, *e.g.*, in Kreslavski *et al.* 2007, Sage and Kubien 2007, Allakhverdiev *et al.* 2008, Chaves *et al.* 2009, Lawlor and Tezara 2009 *etc.*). There are three major categories of photosynthetic parameters usually recommended for the evaluation of stress tolerance and the introduction into breeding programs: characteristics based on the gas exchange measurements, the determination of the content of photosynthetic pigments, and the chlorophyll fluorescence measurements. All have been abundantly tested in many crop species regarding their possible utility for introduction into breeding programs aimed at the improvement of plant stress tolerance. However, the results of these studies are somewhat ambiguous: although some authors have recommended at least one of the above-mentioned parameters as a suitable secondary selection criterion, others did not.

The net photosynthetic rate (P_N) measured with some portable gas analysis system has been examined (together with the stomatal conductance (g_s), and sometimes transpiration rate (E) as potential selection marker for the detection of drought tolerance in wheat (Di Marco *et al.* 1988, Subrahmanyam *et al.* 2006, Živčák *et al.* 2008), triticale (Hura *et al.* 2007), maize (Zarco-Perelló *et al.* 2005), potato (Schapendonk *et al.* 1989b), sugar beet (Ober *et al.* 2005), faba bean (Khan *et al.* 2007), or sunflower (Jamaux *et al.* 1997). With the exception of Ober *et al.* (2005), all the above-mentioned studies recommended the P_N (or g_s) as a secondary selection marker in breeding for an increased drought tolerance. The chlorophyll (Chl) content is another physiological trait often suggested as a suitable marker for the selection against sensitivity to drought (*e.g.* Araus *et al.* 1998, Royo *et al.* 2000, Fotovat *et al.* 2007, Olivares-Villegas *et al.* 2007 and Paknejad *et al.* 2007 in wheat, Li *et al.* 2006 in barley, Betrán *et al.* 2003 and O'Neill *et al.* 2006 in maize, Silva *et al.* 2007 in sugarcane). As with gas-exchange parameters, the usefulness of this parameter for screening and selection for drought tolerance is somehow ambiguous, as some authors advise for its use (Li *et al.* 2006, Fotovat *et al.* 2007, Silva *et al.* 2007) while others advise against it (Royo *et al.* 2000, O'Neill *et al.* 2006).

However, the third group of photosynthetic parameters suggested for the improvement of crop production strategies, *i.e.* Chl fluorescence parameters, clearly stands out due to the rapidity and non-invasivity of this technique and the major recent progress in the development of the appropriate instruments. Parameters associated both with the fast-transient and slow-transient phase of Chl fluorescence induction kinetics, and in some cases (Hura *et al*. 2007) the fluorescence emission spectra, have been widely examined as possible tools for screening and selection for stress tolerance, particularly in connection with breeding for drought tolerance. Such studies have been made *e.g.* in wheat (Di Marco *et al.* 1988, Dib *et al.* 1994, Flagella *et al.* 1995, 1998, Araus *et al.* 1998, Royo *et al.* 2000, Subrahmanyam *et al.* 2006, Paknejad *et al.* 2007, Sayar *et al*. 2008, Živčák *et al.* 2008), barley (Flagella *et al.* 1998, Li *et al.* 2006, Oukarroum *et al.* 2007), triticale (Hura *et al.* 2007), maize (Selmani and Wassom 1991, O'Neill *et al.* 2006), sugarcane (Silva *et al.* 2007), *Lolium-Festuca* hybrids (Koscielniak *et al.* 2006), potato (Schapendonk *et al.* 1989a, Schapendonk and Tonk 1991), or groundnut (Clavel *et al*. 2006).

However, almost all the above-mentioned studies have been made with the single purpose to examine the suitability of various photosynthetic parameters for simple screening of large sets of genotypes for their tolerance of various stressors. The assessment whether the genotypes selected in such a manner will transmit this tolerance into the next generation and, thus, whether the respective parameter can be used for the prediction of the performance of hybrid progeny as well, has been made very rarely (Fracheboud *et al.* 1999, Betrán *et al.* 2003, Koscielniak *et al.* 2005). Yet, without this information, even the most suitable parameter can not help much with the improvement of breeding programs for crop species such as maize, which is bred with the primary purpose of creating hybrid progeny that will eventually be cultivated commercially. We have thus decided to examine whether the photosynthetic performance of parental genotypes of maize subjected to water deficiency conditions correlates

with the performance of their progeny under such conditions, *i.e.* whether various photosyntetic parameters could be used as reliable physiological markers for early breeding generations. As such parameters should also display high heritability (preferably the narrow-sense heritability, *i.e.*, the main genetic effects determining the intraspecific variability in these parameters should be of an additive character), we have also assessed the relative importance of the additive and the non-additive (dominance, maternal) genetic effects by means of the quantitative genetics analysis.

Materials and methods

Five inbred lines of maize (*Zea mays* L.) designated as 2013, 2023, 2086, CE704, and CE810 were selected for the study. These genotypes are part of a breeding programme of the CEZEA Maize Breeding Station (Čejč, Czech Republic) and differ in various yield, morphological, and physiological parameters. As one aim of our study was to examine the possible effect of the maternal genotype on the response of its progeny, only those parental combinations that could be successfully crossed in both directions were used for the breeding of F1 generation, thus yielding ten F1 hybrids: 2013×CE704, CE704×2013, 2013×CE810, CE810×2013, 2023×2086, 2086×2023, 2023×CE704, CE704×2023, CE704×CE810, and CE810×CE704.

The cultivation of the plants and the experiments were performed during April – May 2007. Kernels of both parental and hybrid genotypes were sown into pots (12 cm in diameter, 13 cm deep, one kernel per pot) filled with a mixture of garden soil and sand (2:1 v/v) and placed in a naturally lit greenhouse of the Faculty of Science, Charles University in Prague, and of the Faculty of Agrobiology, Food and Natural Resources, Czech University of Life Sciences Prague (Czech Republic), under semi-controlled conditions at a temperature of 25±2/20±2°C and air humidity 50±5/70±5% day/night for 32 days (*i.e.*, till the V3 developmental stage of plants). At this time point, the plants were divided into two groups, "stress" and "control" (each approx. 30 plants per each genotype). In the "stress" group, the water supply to the plants was withheld for 6 days and the soil was allowed to gradually dry up till the volumetric soil water content was approx. 12.5% and the plants showed visible symptoms of water deficiency stress (*i.e.*, leaf rolling). The control plants were normally watered during this period. In both "stress" and "control" treatments, the completely randomized design of experiments was used. All physiological and morphological measurements were made in the middle part of the 4th leaf (counting from the plant base) blade of 38-d-old plants (or, in some cases, they were determined from the whole plants) with 6 to 10 replicates per each genotype/treatment combination. At the time of the measurements, the 4th leaves were the youngest fully developed leaves in the "stress" group of plants and usually the second youngest ones in the "control" group. The irradiance in the greenhouse at the time of the measurements and at the level of the measured leaves was approx. 800 ± 350 μmol m^{-2} s^{-1}.

The P_N, E and g_s were measured in the leaves *in situ* using the portable gas-exchange system *LCpro+* (*ADC BioScientific Ltd.*, Hoddesdon, Great Britain) from 8:00 to 11:30 of Central European time. The irradiance was 650 μmol m^{-2} s^{-1} PAR, the temperature in the measurement chamber was 25ºC, the CO_2 concentration was 550 ± 50 cm^3 m^{-3}, the air flow rate was 205 ± 30 μmol s^{-1} and the duration of the measurement of each sample was 10 min after the establishment of steady-state conditions inside the measurement chamber. The water-use efficiency (WUE) was calculated as P_N/E. The minimum Chl fluorescence (F_0) and the maximum Chl fluorescence (F_m) were measured also *in situ* with the portable Chl fluorometer *OS30P* (*ADC BioScientific Ltd.*, Hoddesdon, Great Britain) with 1-s excitation pulse (660 nm) and saturation intensity 3,000 μmol m^{-2} s^{-1} after 20-min dark adaptation of the leaves. The maximum quantum efficiency of photosystem (PS) II was calculated as F_v/F_m ($F_v = F_m - F_0$). The content of Chls *a* and *b* and the content of total carotenoids (Cars) were determined after their extraction from leaf discs with N,N-dimethylformamide (Wellburn 1994). The relative water content (RWC) in the leaves was established as 100 × (FM – DM)/(SM – DM), where FM represents the fresh mass of 10 leaf discs (diameter 6 mm) cut from the middle part of leaf blade and immediately weighed on an analytical balance with 0.1 mg readability, SM is the saturated mass of the same discs after their hydration in the dark for 5 h, and DM is the dry mass of these discs after they were oven-dried at 80ºC for 24 h. The specific leaf mass (SLM), *i.e.*, the dry mass of the 4th leaf expressed per the unit of leaf area, was determined from the same leaf discs as RWC. The DM of the 4th leaf, the total DM of the shoot and roots and the height of each plant (measured from the ground to the ligule of the youngest fully developed leaf) were also recorded at the end of the cultivation period.

All data were statistically evaluated by two-way analysis of variance (*ANOVA*) with genotypes (G), water treatments (T) and G×T interaction as possible sources of variability. The statistical significance of the differences between individual genotype/treatment combinations was tested using *LSD* (least significant difference) test with 0.05 probability level as the significant one. The relationship between individual photosynthetic/morphological parameters as well as the relationship between the response of the hybrids to water deficiency and the response of their maternal or paternal parents was examined using the calculation of Pearson´s correlation

coefficient (*r*). The response to water deficiency was in this case always expressed as 100 × (the mean value of the respective parameter measured in the stressed plants/the mean value of this parameter measured in the control plants). All statistical evaluations were made with the *CoStat* computer programme, version *6.204* (*CoHort Software*, Monterey, CA, the U.S.A.). The quantitative genetic analysis aimed at the establishment of the relative importance of the additive and the non-additive (dominance, maternal) genetic effects was based on the modified model of Eberhart and Gardner (1966) as described in Kočová *et al.* (2009). The computer programme *CBE3* from the *Software Package CBE*, *version 4.0*, was used for this analysis (Wolf 1996).

Results

The exposure of maize plants to six days of water deficiency resulted in a statistically significant decrease of the leaf RWC (Table 1), maximum quantum efficiency of PSII (F_v/F_m), and Chl *a* and *b* contents (Table 2). The content of Cars, the shoot DM and the plant height were also lower in the stressed plants compared to the control ones, although this difference was not statistically significant in some genotypes (Tables 2, 3). On the other hand, the values of F_0 parameter significantly increased after the exposure of the plants to a water deficiency period in almost all examined genotypes (Table 2), and a similar effect was observed for WUE, although in this case the increase was only exceptionally statistically significant (Table 1). Both the DM and SLM of the 4th leaf, as well as the DM of the roots mostly did not show any significant changes resulting from disrupted water supply (Table 3). As regards the gas-exchange parameters (P_N, E, and g_s), no general trend in their response to drought simulation could be observed: their values decreased in some of the examined genotypes but increased or did not significantly change in others (Table 1). This dependence of leaf gas-exchange response to water deficiency on the genotype was confirmed by the presence of statistically significant G×T interaction in the results of two-way *ANOVA* (Table 4). Such interaction was not found for any other photosynthetic or morphological parameter with the exception of RWC (Table 4). This means that although the individual genotypes differed in the values of these parameters between themselves (as shown by both *ANOVA* and *LSD* tests, Tables 1 to 4), the response of all examined genotypes to the respective water treatment was always similar.

The relationship between various photosynthetic and morphological parameters was examined using the calculation of Pearson´s correlation coefficients. The positive, statistically highly significant ($P \leq 0.01$) correlation ($r = 0.79 \pm 0.12$) was found between the P_N and E parameters, as well as between P_N and g_s ($r = 0.91 \pm 0.08$) or E and g_s ($r = 0.84 \pm 0.10$). Further positive correlations were observed between F_v/F_m, the content of Chls and Cars, RWC, the plant height and the DM of shoot, whereas the F_0 parameter negatively and significantly correlated with the content of photosynthetic pigments, RWC and the shoot DM (Table 5). Gas-exchange characteristics did not correlate with either RWC, Chl fluorescence parameters, photosynthetic pigments content or morphological parameters. Other correlations between photosynthetic and morphological parameters of the plants were also statistically nonsignificant (data not shown). WUE did not correlate with any photosynthetic or morphological parameter examined.

Among parental genotypes, the inbred line 2086 showed a high rate of both photosynthesis and transpiration, as well as high stomatal conductance, whereas the CE704 inbred line ranked the lowest in this respect (Table 1). The inbred line 2013 was characterized by the highest content of photosynthetic pigments under the control conditions but was replaced in this position by the CE810 inbred line when the plants were stressed by water deficiency (Table 2). The inbred line 2023 usually showed the lowest values of the content of both Chls and Cars and the F_m or F_v/F_m parameters among the parental genotypes subjected to six days without watering (Table 2). The ranking of the F1 hybrids strongly depended both on the parameter examined and the water-treatment conditions (Tables 1–3) and no clear order could be thus established.

When the response of individual F1 hybrids to water deficiency was compared to the response of their respective maternal or paternal parents, no statistically significant correlation was detected regardless of the photosynthetic or morphological parameter examined (Table 6). This clearly shows that it is not possible to predict the response of hybrid genotype to drought based on the behaviour of either its maternal or paternal parent.

In order to ascertain which type of genetic effects is more important for the inheritance of the photosynthetic parameters in maize, the quantitative genetic analysis was performed and its results are shown in Tables 7 and 8. Three principal conclusions could be made from these results. First, the mode of inheritance of each parameter examined in this study markedly differed between plants grown in the control and the stress conditions, *i.e.*, the genetic effects significant under optimum water treatment were frequently nonsignificant under water-deficiency treatment and *vice versa*, or, in the case the same effect was significant in the plants under both water treatments, its character often changed between positive and negative. Second, although the additive genetic effects played an important role in the inheritance of the examined parameters, the nonadditive effects associated with a particular combination of genotypes (dominance effects) or a particular direction of crossing (maternal

Table 1. Mean values ± SD (n = 10) of selected parameters of plant water status and leaf gas exchange measured in five inbred lines of maize and their F1 hybrids grown either under normal conditions (control) or subjected to water deficiency (stress). *Letters* a–m denote the statistical significance (as determined by *LSD* test) of the differences between individual genotypes/ treatments (only those marked with different letters differ significantly at $P \leq 0.05$). The LSD values significant at P=0.05 are shown at the end of the table. RWC – relative water content; g_s – stomatal conductance; E – transpiration rate; P_N – net photosynthetic rate; WUE – water-use efficiency.

Genotype	RWC [%]		g_s [mol m^{-2} s^{-1}]		E [mmol H_2O m^{-2} s^{-1}]		P_N [μmol CO_2 m^{-2} s^{-1}]		WUE [μmol $mmol^{-1}$]	
	Control	Stress	Control	Stress	Control	Stress	Control	Stress	Control	Stress
2013	96.46±4.30^{a}	58.86±11.00^{b}	0.06±0.02^{ghij}	0.12±0.03^{cd}	1.04±0.18^{defgh}	1.01±0.09^{fgh}	8.65±4.41^{bcdefg}	10.01±1.02^{abcde}	7.78±3.75^{def}	9.96±0.97^{abcd}
2023	99.29±3.87^{a}	39.35±5.96^{cde}	0.11±0.04^{cde}	0.03±0.01^{ijk}	1.26±0.25^{bcde}	0.55±0.04^{jk}	10.32±4.08^{abcd}	4.55±0.61^{jklm}	7.88±2.39^{cdef}	8.30±1.12^{bcdef}
2086	92.28±13.45^{a}	47.74±14.65^{c}	0.15±0.06^{ab}	0.09±0.04^{defg}	1.50±0.33^{b}	1.04±0.35^{efgh}	11.94±3.28^{a}	10.41±3.02^{abc}	7.86±0.87^{cdef}	10.79±3.12^{abc}
CE704	96.42±3.76^{a}	35.97±9.59^{def}	0.05±0.01^{hijk}	0.02±0.00^{k}	0.83±0.18^{hi}	0.28±0.02^{l}	5.58±2.24^{hijkl}	2.39±1.51^{m}	6.39±1.96^{ef}	8.38±4.96^{bcdef}
CE810	89.90±4.14^{a}	37.17±12.97^{def}	0.13±0.05^{bc}	0.08±0.02^{efgh}	1.26±0.43^{bcde}	0.90±0.20^{ghi}	10.40±5.65^{abc}	9.41±1.52^{abcdef}	7.38±3.01^{def}	10.90±2.52^{ab}
2013×CE704	93.51±8.95^{a}	34.00±4.35^{def}	0.06±0.02^{ghi}	0.06±0.02^{ghi}	0.96±0.24^{fghi}	0.91±0.10^{ghi}	8.03±1.26^{cdefghi}	8.28±1.90^{cdefgh}	8.67±1.15^{bcdef}	9.22±2.32^{bcde}
CE704×2013	91.43±9.79^{a}	28.28±9.99^{f}	0.08±0.04^{fgh}	0.05±0.02^{hijk}	1.03±0.36^{efgh}	0.75±0.18^{ij}	6.88±3.72^{fghij}	5.30±2.85^{ijkl}	6.02±2.94^{f}	7.04±3.94^{def}
2013×CE810	94.86±5.60^{a}	29.73±9.98^{ef}	0.11±0.03^{ghij}	0.09±0.04^{defg}	0.90±0.33^{ghi}	1.15±0.27^{cdef}	5.74±2.57^{hijkl}	9.43±3.65^{abcdef}	6.73±4.09^{ef}	8.82±6.11^{bcdef}
CE810×2013	93.44±7.99^{a}	32.28±8.52^{def}	0.10±0.03^{cdef}	0.02±0.00^{k}	1.28±0.24^{bcd}	0.48±0.10^{kl}	8.98±2.62^{bcdefg}	3.23±2.52^{lm}	6.87±1.00^{ef}	6.40±3.35^{ef}
2023×2086	92.39±8.55^{a}	38.99±11.53^{cde}	0.10±0.04^{cdef}	0.07±0.03^{ghi}	1.29±0.35^{bc}	0.93±0.05^{fghi}	9.45±4.01^{abcdef}	7.65±0.50^{defghi}	6.86±1.98^{ef}	8.22±0.50^{bcdef}
2086×2023	97.02±1.71^{a}	38.50±15.20^{cde}	0.09±0.01^{defg}	0.03±0.00^{jk}	1.12±0.06^{cdefg}	0.47±0.06^{kl}	7.49±1.12^{efghi}	3.79±2.28^{klm}	6.68±0.83^{ef}	8.00±4.73^{bcdef}
2023×CE704	94.36±4.01^{a}	34.94±6.33^{def}	0.11±0.05^{cdef}	0.16±0.06^{a}	1.30±0.33^{bc}	1.47±0.35^{b}	9.97±4.71^{abcde}	11.40±3.32^{a}	7.21±2.79^{def}	7.53±1.15^{def}
CE704×2023	97.35±2.54^{a}	40.31±4.37^{cd}	0.06±0.01^{ghij}	0.11±0.02^{cd}	1.02±0.11^{fgh}	1.74±0.24^{a}	6.28±2.61^{ghijk}	11.12±0.92^{ab}	6.18±2.90^{f}	6.50±1.11^{ef}
CE704×CE810	90.93±5.37^{a}	35.58±12.22^{def}	0.12±0.06^{bcd}	0.13±0.06^{bc}	1.25±0.51^{cde}	1.35±0.41^{bc}	11.10±5.72^{ab}	12.08±4.81^{a}	7.87±2.82^{cdef}	8.48±1.96^{bcdef}
CE810×CE704	92.90±6.40^{a}	34.43±7.60^{def}	0.11±0.06^{cd}	0.07±0.01^{ghi}	1.29±0.42^{bcd}	0.94±0.35^{fghi}	10.34±3.55^{abcd}	9.60±3.33^{abcdef}	8.08±0.90^{bcdef}	12.19±11.33^{a}
LSD (P=0.05)		9.85		0.04		0.26		2.97		3.17

Table 2. Mean values ± SD (n = 6) of chlorophyll fluorescence parameters and the content of photosynthetic pigments in leaves of five inbred lines of maize and their F1 hybrids grown either under normal conditions (control) or subjected to water deficiency (stress). *Letters* a–m denote the statistical significance (as determined by *LSD* test) of the differences between individual genotypes/ treatments (only those marked with different letters differ significantly at $P \leq 0.05$). a.u. – arbitrary units for the measurement of F_0 with portable chlorophyll fluorometer *OS30P* (*ADC BioScientific Ltd.*, Hoddesdon, Great Britain). The *LSD* values significant at P=0.05 are shown at the end of the table. F_0 – minimum chlorophyll fluorescence; F_v/F_m – maximum quantum efficiency of photosystem II; Chl – chlorophyll; Cars – carotenoids.

Genotype	F_0 [a.u.]		F_v/F_m		Chl *a* content [mg m^{-2}]		Chl *b* content [mg m^{-2}]		Cars content [mg m^{-2}]	
	Control	Stress	Control	Stress	Control	Stress	Control	Stress	Control	Stress
2013	33.25±1.08^{gh}	37.42±5.19^{defgh}	0.81±0.00^{a}	0.64±0.15^{ab}	292.00±16.21^{abc}	190.78±56.32^{ghij}	73.25±6.29^{abcd}	50.54±13.57^{hij}	52.20±1.50^{abcd}	42.89±6.26^{ijkl}
2023	34.50±4.43^{efgh}	51.17±12.14^{a}	0.79±0.03^{a}	0.30±0.27^{d}	236.63±19.79^{defg}	108.19±24.58^{k}	64.32±7.10^{cdefg}	30.05±11.71^{l}	42.29±2.98^{jkl}	34.27±4.99^{m}
2086	29.92±1.20^{h}	37.08±7.30^{defgh}	0.81±0.01^{a}	0.50±0.33^{bcd}	264.66±28.71^{bcde}	199.32±65.33^{ghij}	65.81±10.01^{bcdef}	50.71±13.06^{hij}	51.41±3.93^{bcde}	51.16±6.16^{bcdef}
CE704	32.25±2.89^{h}	49.83±11.04^{ab}	0.80±0.01^{a}	0.44±0.26^{bcd}	252.87±30.74^{cdef}	171.10±59.49^{ij}	69.96±7.64^{abcde}	50.03±14.71^{hij}	44.65±4.49^{ghijkl}	43.55±3.89^{ijkl}
CE810	32.08±3.40^{h}	37.33±8.37^{defgh}	0.80±0.03^{a}	0.47±0.23^{bcd}	226.23±80.99^{defgh}	214.55±57.23^{efghi}	60.21±20.24^{defgh}	56.59±14.00^{efghi}	44.20±10.47^{hijkl}	45.01±6.75^{fghijkl}
2013×CE704	31.75±1.37^{h}	37.25±8.94^{defgh}	0.80±0.02^{a}	0.46±0.25^{bcd}	301.29±11.57^{abc}	205.36±50.51^{fghij}	78.38±6.31^{ab}	58.27±11.69^{efghi}	53.07±2.50^{abc}	46.79±7.41^{cdefghij}
CE704×2013	31.58±1.72^{h}	40.08±3.14^{defg}	0.81±0.01^{a}	0.47±0.21^{bcd}	308.97±32.24^{ab}	190.34±79.43^{ghij}	80.20±11.45^{a}	52.02±17.31^{ghij}	53.82±4.23^{ab}	50.61±6.05^{bcdefg}
2013×CE810	32.83±1.97^{gh}	41.75±4.33^{cdef}	0.80±0.02^{a}	0.40±0.22^{cd}	312.50±24.71^{ab}	187.30±58.37^{ghij}	81.30±9.38^{a}	52.18±11.85^{ghij}	57.88±1.99^{a}	48.09±10.38^{bcdefghij}
CE810×2013	34.50±2.24^{fgh}	40.17±8.53^{defg}	0.80±0.01^{a}	0.35±0.22^{cd}	309.72±22.99^{ab}	175.46±50.85^{hij}	79.47±7.68^{a}	52.27±8.09^{ghij}	58.19±3.81^{a}	49.14±4.93^{bcdefghi}
2023×2086	33.00±1.64^{gh}	35.00±8.28^{efgh}	0.81±0.00^{a}	0.51±0.26^{bcd}	326.30±33.68^{a}	199.91±50.34^{ghij}	81.76±12.05^{a}	49.90±11.00^{hij}	58.03±5.52^{a}	47.78±4.76^{bcdefghij}
2086×2023	32.42±1.02^{gh}	42.50±5.50^{bcde}	0.81±0.01^{a}	0.50±0.32^{bcd}	289.65±11.82^{abc}	195.55±47.68^{ghij}	74.45±6.53^{abc}	49.55±11.29^{hij}	51.95±3.12^{abcde}	47.89±8.20^{bcdefghij}
2023×CE704	33.92±2.27^{fgh}	42.42±13.40^{bcde}	0.80±0.01^{a}	0.31±0.27^{cd}	267.15±18.33^{bcd}	155.96±43.01^{jk}	71.84±8.49^{abcd}	40.33±12.20^{jk}	48.05±3.13^{bcdefghij}	39.74±3.81^{klm}
CE704×2023	32.67±2.60^{gh}	48.75±18.54^{abc}	0.80±0.01^{a}	0.51±0.26^{bc}	262.49±15.43^{bcde}	171.66±60.73^{ij}	70.01±8.38^{abcde}	45.73±16.83^{ij}	47.76±2.41^{bcdefghij}	39.01±6.53^{lm}
CE704×CE810	33.33±1.44^{gh}	43.50±5.43^{abcd}	0.80±0.01^{a}	0.43±0.26^{cd}	275.01±41.66^{abcd}	190.28±51.05^{ghij}	74.76±13.16^{abc}	53.79±13.15^{fghi}	50.26±7.06^{bcdefgh}	45.69±5.93^{efghijk}
CE810×CE704	32.92±2.27^{gh}	44.75±10.24^{abcd}	0.80±0.01^{a}	0.46±0.19^{bcd}	293.65±35.56^{abc}	181.84±44.55^{hij}	77.56±11.51^{abc}	51.54±11.93^{ghij}	53.90±5.19^{ab}	46.08±3.25^{deefghij}
LSD (P=0.05)	7.90		0.20		51.60		13.43		6.29	

Table 3. Mean values ± SD (n = 6) of selected morphological parameters of five inbred lines of maize and their F1 hybrids grown either under normal conditions (control) or subjected to water deficiency (stress). *Letters* a–m denote the statistical significance (as determined by LSD test) of the differences between individual genotypes/ treatments (only those marked with different letters differ significantly at $P \leq 0.05$). SLM – specific leaf mass. DM – dry mass. The *LSD* values significant at P=0.05 are shown at the end of the table.

Genotype	SLM of the 4th leaf [mg m^{-2}]		DM of the 4th leaf [g]		Shoot DM [g]		Root DM [g]		Plant height [mm]	
	Control	Stress	Control	Stress	Control	Stress	Control	Stress	Control	Stress
2013	16.22±2.34^{abcd}	19.09±9.09^{a}	0.08±0.02^{klm}	0.06±0.03^{lm}	0.74±0.34^{defg}	0.40±0.27^{g}	0.18±0.09^{h}	0.17±0.06^{h}	159±46^{defghijk}	126±28^{klm}
2023	15.63±2.34^{abcd}	14.86±1.61^{bcd}	0.15±0.02^{bcde}	0.16±0.02^{bcd}	1.19±0.26^{cd}	0.96±0.37^{cdef}	0.32±0.07^{fgh}	0.45±0.12^{cdef}	178±19^{bcdef}	143±30^{ghijkl}
2086	16.30±5.62^{abcd}	18.36±4.73^{abc}	0.06±0.03^{m}	0.07±0.02^{klm}	0.69±0.52^{defg}	0.48±0.27^{fg}	0.22±0.15^{gh}	0.23±0.10^{gh}	107±38^{mn}	91±29^{n}
CE704	16.57±1.35^{abcd}	17.15±1.14^{abcd}	0.09±0.04^{ijklm}	0.09±0.04^{hijjklm}	1.32±0.33^{bc}	0.85±0.47^{cdefg}	0.45±0.14^{cdef}	0.45±0.18^{cdef}	169±27^{cdefg}	134±28^{ijklm}
CE810	14.74±2.10^{cd}	17.88±5.53^{abc}	0.10±0.02^{ghijkl}	0.09±0.02^{jjkl}	0.97±0.28^{cdef}	0.59±0.23^{efg}	0.25±0.07^{gh}	0.24±0.09^{gh}	168±21^{cdefghi}	143±19^{ghijkl}
2013×CE704	15.86±2.80^{abcd}	15.69±3.10^{abcd}	0.13±0.04^{cdefgh}	0.11±0.04^{efghijk}	1.92±0.70^{a}	0.96±0.33^{cdef}	0.58±0.20^{abcd}	0.45±0.18^{cdef}	189±39^{abcd}	146±11^{efghijkl}
CE704×2013	17.89±2.36^{abc}	17.99±2.56^{abc}	0.13±0.02^{cdefghi}	0.12±0.04^{cdefghij}	2.08±0.71^{a}	1.15±0.45^{cd}	0.57±0.19^{abcd}	0.55±0.16^{abcd}	185±27^{bcd}	146±29^{fghijkl}
2013×CE810	16.75±1.68^{abcd}	16.09±2.61^{abcd}	0.14±0.03^{bcdef}	0.13±0.03^{cdefg}	2.03±0.55^{a}	1.11±0.28^{cde}	0.54±0.16^{abcd}	0.43±0.10^{def}	221±26^{a}	176±20^{bcdefg}
CE810×2013	18.40±1.27^{ab}	13.82±4.16^{d}	0.16±0.02^{bc}	0.14±0.03^{cdefg}	2.03±0.56^{a}	1.18±0.49^{cd}	0.60±0.14^{abc}	0.44±0.16^{def}	200±31^{abc}	180±27^{bcde}
2023×2086	18.16±1.76^{abc}	15.39±2.69^{bcd}	0.21±0.04^{a}	0.18±0.05^{ab}	1.83±0.60^{ab}	1.02±0.33^{cde}	0.56±0.15^{abcd}	0.48±0.13^{bcdef}	161±27^{defghij}	133±27^{jklm}
2086×2023	15.16±2.63^{bcd}	16.16±2.61^{abcd}	0.12±0.04^{defghij}	0.15±0.03^{bcde}	1.15±0.70^{cd}	0.76±0.45^{defg}	0.33±0.18^{fgh}	0.32±0.15^{fgh}	158±34^{defghijk}	122±33^{lmn}
2023×CE704	16.10±2.96^{abcd}	15.63±2.06^{abcd}	0.18±0.07^{ab}	0.15±0.02^{bcde}	1.75±0.63^{ab}	0.99±0.31^{cdef}	0.52±0.14^{bcde}	0.45±0.14^{cdef}	182±35^{bcd}	143±37^{hijkl}
CE704×2023	15.39±2.44^{bcd}	17.40±1.27^{abcd}	0.14±0.02^{bcdef}	0.11±0.03^{efghijk}	1.82±0.31^{ab}	0.85±0.24^{cdefg}	0.63±0.10^{ab}	0.37±0.04^{efg}	181±30^{bcd}	138±17^{hijklm}
CE704×CE810	16.04±0.91^{abcd}	15.83±2.96^{abcd}	0.15±0.05^{bcde}	0.10±0.05^{fghijk}	1.96±0.76^{a}	1.00±0.41^{cdef}	0.69±0.25^{a}	0.44±0.14^{cdef}	187±32^{abcd}	161±33^{defghij}
CE810×CE704	17.04±1.82^{abcd}	14.80±0.72^{bcd}	0.12±0.05^{defghij}	0.11±0.02^{efghijk}	1.97±0.67^{a}	0.96±0.29^{cdef}	0.58±0.17^{abcd}	0.45±0.14^{cdef}	210±35^{ab}	170±28^{cdefgh}
LSD (P=0.05)	3.63		0.06		0.53		0.16		33.74	

effects) were at least equally and often even more important (particularly for the gas-exchange parameters in the plants stressed by water deficiency). And third, each evaluated parameter displayed a unique combination of genetic effects participating in its inheritance, thus preventing any selection of a particular genotype/genotype combination as a general bearer of strong additive or nonadditive effects.

Discussion

The effects of drought on plants have been studied for a long time and the changes induced by insufficient water supply have been thoroughly examined from the whole plant/plant population level to the biochemical and molecular levels (Bray 2007, Chaves *et al.* 2009, Farooq *et al.* 2009). The reduction of photosynthetic efficiency is a well-known symptom of drought-induced stress displayed by many plant species. In our study with inbred and hybrid genotypes of maize we observed the reduction of the P_N after the plants had been exposed to the 6-d period of water deficiency, and this reduction was usually accompanied by a decrease in the values of both g_s and E. Those genotypes that either did not show any changes of the stomatal function or even displayed an increased g_s after drought simulation compared to the nonstressed plants, were usually also characterized by increased values of both E and P_N, and there was a strong positive correlation between these parameters. At the initial stages of water deficit, the reductive effect of stomatal closure on E is thought to be greater than the effect on photosynthetic CO_2 assimilation, but with further development of water deficit, both processes are reduced rather dramatically (Chaves *et al.* 2002, Chaves and Oliveira 2004, Flexas *et al.* 2004). The actual contribution of the diminished stomatal conductance to drought-induced limitation of photosynthesis has been much discussed during past decades, particularly concerning its relative importance in comparison with that of the metabolic limitation and/or impaired ATP synthesis (Flexas and Medrano 2002, Lawlor 2002). The majority of scientists working in this area of research now accept the "stomatal control" model which proposes that stomatal closure and decrease of g_s are the primary causes of the reduction of P_N under mild drought conditions, with metabolic alterations (the inhibition of ATP synthesis and the inadequate RuBP regeneration associated with this inhibition) following later and developing gradually as drought progresses further (Chaves *et al.* 2002, 2009, Lawlor 2002, Reddy *et al.* 2004, Christensen and Feldman 2007, Lawlor and Tezara 2009).

Table 4. Analysis of variance of selected physiological and morphological parameters of maize plants of five inbred lines and their ten F1 hybrids grown either under normal conditions or subjected to six days of water deficiency. Genotypes (G), water treatments (T) and their interaction (G×T) were included in the analysis as the possible sources of variation and their levels of statistical significance (P) are shown. RWC – relative water content; WUE – water-use efficiency; P_N – net photosynthetic rate; E – transpiration rate; g_s – stomatal conductance; F_0 – minimum chlorophyll fluorescence; F_v/F_m – maximum quantum efficiency of photosystem II; Chl – chlorophyll; Cars – carotenoids; DM – dry mass.

Parameter	G	T	G×T
RWC	< 0.001	< 0.001	0.02
WUE	0.01	< 0.001	0.68
P_N	< 0.001	0.02	< 0.001
E	< 0.001	< 0.001	< 0.001
g_s	< 0.001	< 0.001	< 0.001
F_0	0.03	< 0.001	0.18
$F_v/F_m = (F_m–F_0)/F_m$	0.72	< 0.001	0.85
Chl *a* content	< 0.001	< 0.001	0.15
Chl *b* content	< 0.001	< 0.001	0.23
Cars content	< 0.001	< 0.001	0.19
Specific mass of the 4th leaf	0.78	0.78	0.20
DM of the 4th leaf	< 0.001	0.04	0.64
Shoot DM	< 0.001	< 0.001	0.24
Root DM	< 0.001	< 0.001	0.09
Plant height	< 0.001	< 0.001	1.00

Table 5. Correlations between selected chlorophyll fluorescence parameters (F_0, F_v/F_m), the content of photosyntetic pigments, the relative water content of leaves, the shoot dry mass and plant height, measured in five inbred lines of maize and their ten F1 hybrids grown either under normal conditions or subjected to six days of water deficiency. Values of Pearson´s correlation coefficients (r) ± SE(r), n = 30, are shown together with their statistical significance (* – significant at P=0.05, ** – significant at P=0.01, ns – nonsignificant). Chl – chlorophyll; Cars – carotenoids; RWC – relative water content; DM – dry mass.

	Chl *a* content r ± SE (r)	Chl *b* content r ± SE (r)	Cars content r ± SE (r)	RWC r ± SE (r)	Shoot DM r ± SE (r)	Plant height r ± SE (r)
F_0	–0.84 ± 0.10**	–0.82 ± 0.11**	–0.66 ± 0.14**	–0.80 ± 0.12**	–0.46 ± 0.17**	ns
F_v/F_m	0.90 ± 0.08**	0.89 ± 0.09**	0.58 ± 0.15**	0.97 ± 0.05**	0.57 ± 0.16**	0.46 ± 0.17*
Chl *a* content		0.99 ± 0.03**	0.82 ± 0.11**	0.86 ± 0.10**	0.70 ± 0.14**	0.56 ± 0.16**
Chl *b* content			0.80 ± 0.11**	0.85 ± 0.10**	0.74 ± 0.13**	0.64 ± 0.15**
Cars content				0.50 ± 0.16**	0.58 ± 0.15**	0.41 ± 0.17*
RWC					0.60 ± 0.15**	0.51 ± 0.16**
Shoot DM						0.84 ± 0.10**

Table 6. Correlations between the response of maize F1 hybrids and the response of their maternal or paternal parent to water deficiency. Values of Pearson´s correlation coefficients (r) ± SE(r), n = 30, are shown together with their statistical significance (ns – nonsignificant). The response of each genotype to water deficiency (*i.e.* the data the correlation coefficients were calculated from) was expressed as 100 × (the mean value of the respective parameter measured in stressed plants/ the mean value of the parameter measured in control plants). For abbreviations *see* Table 4.

Parameter	Hybrid-maternal parent $r \pm SE(r)$	Hybrid-paternal parent $r \pm SE(r)$
RWC	–0.32 ± 0.34 ^{ns}	–0.37 ± 0.32 ^{ns}
WUE	0.19 ± 0.35 ^{ns}	0.18 ± 0.35 ^{ns}
P_N	–0.07 ± 0.35 ^{ns}	–0.30 ± 0.34 ^{ns}
E	–0.21 ± 0.35 ^{ns}	–0.32 ± 0.34 ^{ns}
g_s	0.17 ± 0.35 ^{ns}	–0.47 ± 0.31 ^{ns}
F_0	0.19 ± 0.35 ^{ns}	0.34 ± 0.33 ^{ns}
$F_v/F_m = (F_m–F_0)/F_m$	0.11 ± 0.35 ^{ns}	–0.42 ± 0.32 ^{ns}
Chl *a* content	–0.05 ± 0.35 ^{ns}	–0.10 ± 0.35 ^{ns}
Chl *b* content	0.49 ± 0.31 ^{ns}	0.10 ± 0.35 ^{ns}
Cars content	0.42 ± 0.32 ^{ns}	–0.27 ± 0.34 ^{ns}
Specific mass of the 4th leaf	–0.29 ± 0.34 ^{ns}	–0.50 ± 0.31 ^{ns}
DM of the 4th leaf	0.40 ± 0.32 ^{ns}	0.06 ± 0.35 ^{ns}
Shoot DM	0.35 ± 0.33 ^{ns}	0.05 ± 0.35 ^{ns}
Root DM	0.34 ± 0.33 ^{ns}	–0.08 ± 0.35 ^{ns}
Plant height	0.34 ± 0.33 ^{ns}	0.25 ± 0.34 ^{ns}

The maximum quantum efficiency of PSII photochemistry and the content of Chls *a* and *b* also rather strongly decreased in our drought-stressed experimental plants, which indicates that the conditions of our drought simulation imitated severe drought stress. Primary photosynthetic processes are considered to be rather resilient to water deficit and the drought-induced decrease of photosynthetic electron transport efficiency occurs only secondarily, being caused by an imbalance between the generation of NADPH and its utilization in the photosynthetic carbon reduction cycle (Cornic and Fresneau 2002, Baker and Rosenquist 2004). Severe drought can nevertheless lead to an increased generation of reactive oxygen species leading to photooxidation and the degradation of photosynthetic membrane proteins (particularly D1, D2 and CP43 proteins of PSII) and associated pigments and lipids (Cornic and Fresneau 2002, Yordanov *et al.* 2003, Reddy *et al.* 2004). Although a certain relationship was observed between the effect of drought on the efficiency of PSII photochemistry and the P_N in some of our genotypes (*e.g.* in the inbred line 2023 or the F1 hybrid CE704×2023), no such correlation between P_N and F_v/F_m (or between P_N and the content of photosynthetic pigments) existed for the majority of genotypes, which, together with the above-mentioned observations, suggests that the net photosynthesis in the leaves of our drought-stressed plants was not limited by the efficiency of PSII or the amount of Chls or Cars but rather by the functioning of stomata.

We have also observed a rather interesting discrepancy between the results of our gasometric measurements and the consistently shown drought-induced decrease in RWC, the PSII efficiency or the content of photosynthetic pigments. Plant water status is not influenced only by water loss from leaves (associated with the efficiency of E and the stomatal closure) but also by water acquisition through roots and its transport into different parts of plants. Thus, some genotypes with greater root system and smaller stature could uptake more water from soil and transport it more efficiently, ensuring no need for stomatal closure at the time point the other, less-adapted genotypes already respond to drought by decreasing their transpiration rate. Maintaining the stomata open would then result in none or only a small diminishion of P_N, but could also lead to an increased E resulting in greater water loss, as reflected in the decrease of RWC. As to why there was no significant correlation between PSII response to drought and P_N, this could be explained in several ways. Following the experiments described in this paper, we have made a more detailed analysis of plant drought response in the most drought-tolerant (whose P_N was not much affected by drought) and -sensitive (which also had diminished P_N) parental line from the set analyzed here (Holá *et al.*, *manuscript in preparation*). Its results showed several things: (*1*) the activity of PSI increased in the tolerant line after drought period so the cyclic electron transport could compensate for the nonfunctional PSII; (*2*) the same applied for the amount and activity of Rubisco and the amount of Rubisco activase; (*3*) the amount of proteins of PSII oxygen-evolving complex and light-harvesting antennae decreased in the sensitive line and was less affected in the tolerant genotype; and (*4*) the proteosynthesis as a whole (and particularly the synthesis of various protective proteins) was intensified in the tolerant genotype. All these factors, together with the delayed stomatal closure, could contribute to the maintenance of high P_N (or, indeed, its increase) even under drought conditions

Table 7. Genetic analysis of selected parameters of plant water status and leaf gas exchange measured in maize plants grown either under normal conditions (control) or subjected to water deficiency (stress). The estimates of genetic effects are shown together with their statistical significance (* – significant at P=0.05, ** – significant at P=0.01, ns – nonsignificant). m – general mean across all genotypes, A – additive genetic effect, D – dominance genetic effect, M – maternal genetic effect (subscripts indicate the parental inbred line or – in the case of the dominance effects – the hybrid combination the respective genetic parameter is relevant to). NI – the respective parameters were not included into the applied model because a simpler model, *i.e.*, one including only the parameters representing the additive (or, in the case of the relative water content and the water use efficiency, the additive and dominance) effects showed better goodness-of-fit than the model including all three types of genetic effects. RWC – relative water content; g_s –stomatal conductance; E – transpiration rate; P_N – net photosynthetic rate; WUE – water-use efficiency.

Genetic parameters	RWC		g_s		E		P_N		WUE	
	Control	Stress	Control	Stress	Control	Stress	Control	Stress	Control	Stress
m	94.96^{**}	43.82^{**}	0.10^{**}	0.07^{**}	1.18^{**}	0.75^{**}	9.38^{**}	7.35^{**}	7.46^{**}	9.67^{**}
A_{2013}	1.39^{ns}	15.04^{**}	0^{ns}	0.04^{**}	0.08^{ns}	-0.18^{**}	0.82^{ns}	-1.08^{ns}	0.32^{ns}	-1.23^{ns}
A_{2023}	3.86^{**}	-4.47^{ns}	-0.02^{ns}	-0.08^{**}	-0.11^{ns}	-0.20^{**}	-1.22^{ns}	-3.69^{**}	0.42^{ns}	-1.26^{ns}
A_{2086}	-0.25^{ns}	3.92^{ns}	0.03^{ns}	0.01^{ns}	0.30^{*}	0.75^{**}	2.36^{ns}	6.03^{**}	0.40^{ns}	1.44^{ns}
A_{CE704}	0.03^{ns}	-7.85^{*}	-0.04^{**}	-0.05^{**}	-0.26^{**}	-0.74^{**}	-2.27^{ns}	-5.57^{**}	-1.07^{ns}	-0.16^{ns}
A_{CE810}	-5.02^{**}	-6.65^{ns}	0.03^{ns}	0.07^{**}	-0.02^{ns}	0.36^{**}	0.30^{ns}	4.31^{**}	-0.08^{ns}	1.21^{ns}
$D_{2013\times CE704}$	NI	-14.33^{**}	0.02^{**}	-0.01^{ns}	0.08^{ns}	0.18^{**}	0.80^{ns}	0.55^{ns}	0.42^{ns}	-1.17^{ns}
$D_{2013\times CE810}$	NI	-16.81^{**}	-0.02^{ns}	-0.04^{**}	-0.05^{ns}	-0.15^{**}	-2.15^{ns}	-3.42^{**}	-0.72^{ns}	-3.08^{**}
$D_{2023\times 2086}$	NI	-4.73^{ns}	-0.03^{**}	-0.01^{*}	-0.17^{*}	-0.09^{ns}	-2.66^{**}	-1.76^{**}	-1.16^{**}	-1.43^{ns}
$D_{2023\times CE704}$	NI	0.93^{ns}	0.01^{ns}	0.11^{**}	0.11^{ns}	1.19^{**}	0.18^{ns}	7.79^{**}	-0.42^{ns}	-1.33^{ns}
$D_{CE704\times CE810}$	NI	-1.82^{ns}	0.03^{ns}	0.05^{**}	0.22^{ns}	0.56^{**}	2.17^{ns}	5.01^{**}	1.17^{ns}	-0.51^{ns}
M_{2013}	NI	NI	-0.03^{**}	0.01^{ns}	-0.22^{**}	0.43^{**}	-1.55^{ns}	3.74^{**}	NI	1.53^{ns}
M_{2023}	NI	NI	0.03^{**}	0.05^{**}	0.19^{*}	-0.01^{ns}	2.16^{ns}	0.89^{ns}	NI	-0.10^{ns}
M_{2086}	NI	NI	0.02^{ns}	0.01^{ns}	0.02^{ns}	-0.47^{**}	0.20^{ns}	-2.97^{**}	NI	-0.32^{ns}
M_{CE704}	NI	NI	0^{ns}	0^{ns}	-0.09^{ns}	0.27^{**}	-1.53^{ns}	0.61^{ns}	NI	-1.13^{ns}
M_{CE810}	NI	NI	0^{ns}	-0.06^{**}	0.10^{ns}	-0.22^{**}	0.72^{ns}	-2.26^{*}	NI	0.03^{ns}

Table 8. Genetic analysis of chlorophyll (Chl) fluorescence parameters and the content of photosynthetic pigments in leaves of maize plants grown either under normal conditions (control) or subjected to water deficiency (ctress). The estimates of genetic effects from the most appropriate model are shown together with their statistical significance (* – significant at $P = 0.05$, ** – significant at $P = 0.01$, ns – nonsignificant). m – general mean across all genotypes, A – additive genetic effect, D – dominance genetic effect, (subscripts indicate the parental inbred line or – in the case of the dominance effects – the hybrid combination the respective genetic parameter is relevant to). NI – the respective parameters were not included into the applied model because a simpler model, *i.e.*, one including only the parameters representing the additive genetic effects showed better goodness-of-fit than the models including also the dominance and/or maternal genetic effects. F_0 – minimum chlorophyll fluorescence; F_v/F_m – maximum quantum efficiency of photosystem II; Chl – chlorophyll; Cars – carotenoids.

Genetic parameters	F_0		F_v/F_m		Chl *a* content		Chl *b* content		Cars content	
	Control	Stress	Control	Stress	Control	Stress	Control	Stress	Control	Stress
m	32.71^{**}	41.25^{**}	0.80^{**}	0.46^{**}	254.48^{**}	185.35^{**}	66.71^{**}	49.90^{**}	46.95^{**}	45.82^{**}
A_{2013}	0.27^{ns}	-3.86^{*}	0^{ns}	0.11^{*}	37.52^{**}	4.37^{ns}	6.54^{*}	2.55^{ns}	5.25^{**}	1.55^{ns}
A_{2023}	2.39^{**}	5.60^{ns}	0^{ns}	-0.11^{ns}	-17.85^{ns}	-63.06^{**}	-2.39^{ns}	-15.82^{**}	-4.66^{**}	-10.08^{**}
A_{2086}	-2.74^{**}	-5.53^{*}	0.01^{**}	0.09^{ns}	10.18^{ns}	48.65^{**}	-0.90^{ns}	6.77^{ns}	4.46^{**}	8.47^{**}
A_{CE704}	-1.22^{*}	2.88^{ns}	0^{ns}	-0.02^{ns}	-1.61^{ns}	0.83^{ns}	3.25^{ns}	2.55^{ns}	-2.30^{ns}	-1.40^{ns}
A_{CE810}	1.31^{*}	0.91^{ns}	-0.01^{**}	-0.07^{ns}	-28.25^{ns}	9.21^{ns}	-6.50^{ns}	3.94^{ns}	-2.75^{ns}	1.46^{ns}
$D_{2013\times CE704}$	NI	NI	NI	NI	29.73^{**}	NI	7.20^{*}	NI	4.84^{**}	NI
$D_{2013\times CE810}$	NI	NI	NI	NI	51.89^{**}	NI	13.48^{**}	NI	9.75^{**}	NI
$D_{2023\times 2086}$	NI	NI	NI	NI	43.03^{**}	NI	11.04^{**}	NI	6.57^{**}	NI
$D_{2023\times CE704}$	NI	NI	NI	NI	19.67^{*}	NI	3.77^{ns}	NI	4.40^{**}	NI
$D_{CE704\times CE810}$	NI	NI	NI	NI	46.24^{*}	NI	11.27^{*}	NI	8.20^{**}	NI

in genotypes displaying a greater tolerance to this stressor.

The utilization of any physiological parameter in plant breeding and selection programs aimed at the improvement of plant tolerance of stressors requires the fulfillment of several criteria such as the possibility of relatively simple and fast measurements of the respective parameter in many samples, its good correlation with the tolerance/sensitivity to the target stress factor, and an adequate intraspecific genetic variation (Brennan and Martin 2007, Sayar *et al.* 2008). The photosynthetic parameters examined in our study certainly satisfy the first condition (particularly the Chl fluorescence measurements) and more-or-less meet also the second condition (based on the presence of positive correlations between Chl fluorescence parameters or the content of photosynthetic pigments and the drought-induced changes in plant morphology and water status). Other authors have also described good association between maize drought tolerance and Chl fluorescence excitation spectra (Grzesiak *et al.* 2007a) or Chl content (Grzesiak *et al.* 2007b). From this point of view, the measurement of P_N seems to be the least suitable among the three categories of photosynthetic parameters examined, as it is rather time-consuming and the relationship between P_N and drought-induced changes in plant morphology and development is not unequivocal (Grzesiak *et al.* 2006).

As regards the intraspecific variability in photosynthetic characteristics, its existence in maize has been previously noted by various authors and confirmed also in our study. Genotypic differences in the content of Chls in maize leaves were found *e.g.* by Oelke and Andrew (1966), Rao *et al.* (1978), Monma and Tsunoda (1979), Baer and Schrader (1985), Crafts-Brandner and Poneleit (1992), Mehta *et al.* (1992) or Krebs *et al.* (1996), and the intraspecific variability in Chl fluorescence parameters was also described for this plant species in some previous papers (*e.g.* Csapó *et al.* 1991, Dolstra *et al.* 1994, Krebs *et al.* 1996). Thus, it would seem that both these categories of photosynthetic parameters are eminently suitable for their inclusion into breeding programs aimed at the improvement of maize drought tolerance. However, the situation is not so simple. Another condition that should be met by such parameters is their high heritability with particular regard to the additive component of genetic variation (Sayar *et al.* 2008). And herein lies the main problem: although the quantitative genetic analysis of our data showed that the additivity certainly can play an important role in the inheritance of the photosynthetic pigments content, the nonadditive (particularly dominance) genetic effects were even more important. Similar phenomenon was found by Oelke and Andrew (1966), Baer and Schrader (1985) or Mehta *et al.* (1992), who also analyzed various components of genetic variation in the Chl content in maize leaves. As regards P_N and other leaf gas-exchange parameters, we have found not only the presence of the dominance genetic effects, but the maternal effects as well, so the situation here is even more complex. The parameters associated with Chl fluorescence did not seem to suffer from such limitations in our study but their intraspecific variability was lower than that of the content of photosynthetic pigments. Moreover, our results clearly demonstrated that the genetic effects participating in the inheritance of the examined photosynthetic parameters almost totally changed when the plants were exposed to drought conditions. Körnerová and Holá (1999) and Kočová *et al.* (2009) described a similar phenomenon for the activities of PSI or PSII and the contents of Chls and Cars in maize plants stressed by low temperatures. Such changes of the relative importance of the additive and nonadditive genetic effects were probably the main cause of the absence of any significant correlation between the response of F1 hybrids to drought conditions and the response of their maternal or paternal parent.

We can thus conclude that although the determination of various Chl fluorescence parameters or the content of photosynthetic pigments can be used for a simple assessment of drought tolerance in collections of maize genotypes, the practical usability of such parameters in maize breeding programs is extremely limited, because their measurements in parental genotypes subjected to water deficiency cannot provide any information on the progeny performance under such conditions.

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