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The effect of low growth temperature on Hill reaction and photosystem 1 activities and pigment contents in maize inbred lines and their F_1 hybrids

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Abstract

Young plants of maize inbred lines CE777, CE704, and CE810 and their F_1 hybrids displaying a positive heterotic effect in various photosynthetic characteristics were exposed to low temperature during their early growth developmental stage. The photochemical activity of isolated mesophyll chloroplasts and the contents of photosynthetic pigments in leaves of stressed and non-stressed plants were compared with the aim to find out the possible changes in the relationship between parents and hybrids, and to determine the genetic basis of heterosis in F_1 generation. Strong decrease in the content of chlorophylls was observed for all genotypes examined when plants were subjected to low growth temperature. Similar change was recorded for Hill reaction activity (HRA) of inbred lines but not of their F_1 hybrids, and no significant response at all was found for photosystem 1 (PS1) activity or the total carotenoids content. The intraspecific variation due to differences between genotypes was found for most of photosynthetic characteristics examined. This variation was caused by the additive and dominance genetic effects. Positive dominance was the main cause of positive heterosis in HRA and in the contents of photosynthetic pigments and was much more pronounced in the stressed plants compared to the non-stressed ones. The maternal additive effects participated in the inheritance of contents of photosynthetic pigments in plants exposed to low temperature, too.

Additional key words: carotenoids; chlorophylls; genetic analysis; heterosis; intraspecific variation; photosystem 2; *Zea mays*.

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Abbreviations: Car - carotenoids; Chl - chlorophyll; HRA - Hill reaction activity; LA - leaf area; PS - photosystem; RC - reaction centre.

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Introduction

The primary photosynthetic processes in higher plants are strongly influenced by low temperature. Cold stress can negatively affect the fluidity of chloroplast thylakoid membranes and change the composition of thylakoid lipids (Yordanov 1992). Plants stressed by low temperature have usually less chlorophyll (Chl) and a greater amount of xanthophyll cycle pigments compared to the non-stressed ones (Haldimann 1997). The synthesis of several chloroplast- or nuclear-encoded thylakoid proteins, as well as their stabilisation in membranes and assembly of pigment-protein complexes can also be affected by low temperature (Nie and Baker 1991, Bredenkamp *et al.* 1992, Yordanov 1992). This can result in the reduction of the whole electron-transport chain activity or the activity of individual pigment-protein complexes of thylakoid membranes. Among them, photosystem (PS) 2 is the most sensitive and its activity can strongly decrease with cold treatment of plants (Baker *et al.* 1983, Nie and Baker 1991). On the other hand, PS1 may be less affected by various stresses including low temperature (Nie and Baker 1991). Cold stress can also lower the rate of photophosphorylation and the activity of thylakoid ATP-synthase (Yordanov 1992, Markowski and Skrudlik 1995). The relative importance of these individual processes with regard to low temperature remains yet to be established, but together they result in the lowering of plant photosynthetic capacity.

The response of photosynthetic apparatus to low temperature is not the same in all plants, however. Strong differences exist not only between various plant species but within one species as well. Some genotypes are less sensitive to low temperature and can better adapt to stress than the other ones. Such differences are especially apparent in the species of tropical or sub-tropical origin, which may be more susceptible to cold stress than the plants of moderate climate. The differential response of photosynthetic rate to low temperature and its dependence on genotype was observed in rice (*Oryza sativa* L.) (Kishitani and Tsunoda 1974), almond (*Prunus amygdalus* Batsch) (Matos *et al.* 1997), or peanut (*Arachis hypogaea* L.) (Bell *et al.* 1994a,b). In peanut, the differences in CO_2 exchange rate were accompanied by differences in some Chl fluorescence parameters which can serve as a measure of PS2 activity (Bell *et al.* 1994a). On the other hand, no such phenomenon was found for tomato (*Lycopersicon esculentum* Mill.) (van de Dijk and Maris 1985, Nieuwhof and van de Dijk 1988). Comparison of two winter cultivars and one spring cultivar of wheat (*Triticum aestivum* L.) stressed or non-stressed by low temperature showed the existence of genetic variation in Chl fluorescence parameters as well as in the oxygen production by leaf discs (Öquist *et al.* 1993).

Maize (*Zea mays* L.) is another species which has been often studied with respect to molecular mechanisms of plant adaptation to cold stress. Plants subjected to low temperatures usually show greater genetic variation in the rate of photosynthesis, Chl fluorescence parameters, or the Chl content than the plants grown at optimum temperature (Greer and Hardacre 1989, Haldimann 1997, Janda *et al.* 1998). Although many studies deal with the difference in photosynthetic characteristics between cold-sensitive and -tolerant cultivars of maize, there has been as yet no examination of the parents-hybrid relationship regarding their response to this type of stress. Maize has another advantage for its choice as a model organism in such analysis: a positive heterotic effect in F_1 hybrids has often been found for various photosynthetic characteristics (*e.g.*, Fousová and Avratovščuková 1967, Fousová 1971, Monma and Tsunoda 1979, Albergoni *et al.* 1983, Baer and Schrader 1985, Kočová 1992, Mehta *et al.* 1992, Qi *et al.* 1995, Krebs *et al.* 1996, Holá *et al.* 1999). The exact causes of this phenomenon are still unknown. It has been speculated that heterosis could be associated with the possible ability of hybrids to better adapt to diverse, changing, and often unfavourable environments. From this point of view, it is very important to study heterotic effect in photosynthetic characteristics both in optimum and stressed conditions, and to determine its genetic mechanisms. As far as we know, heterosis usually arises from strong positive overdominance or complete dominance, together with the non-allelic interactions of several types (Fousová and Avratovščuková 1967, Fousová 1971, Baer and Schrader 1985, Mehta *et al.* 1992, Krebs *et al.* 1996, Holá *et al.* 1999). However, these genetic effects are strongly influenced by external factors, and their expression can probably change with various environments. The purpose of this work, therefore, was to examine the genetic basis of heterosis in several photosynthetic characteristics by comparison of maize inbred lines and their F_1 hybrids grown in optimum and low-temperature conditions, and to ascertain if the inbred lines and hybrids differently respond to cold stress.

Materials and methods

The photochemical activity of isolated mesophyll chloroplasts and the photosynthetic pigments content were measured in three maize (*Zea mays* L.) inbred lines CE777, CE704, and CE810 and their F_1 hybrids (CE777×CE704, CE704×CE777, CE704×CE810, CE810×CE704) stressed by low growth temperature and compared to the control, non-stressed ones. Seeds, obtained from the maize breeding station *CEZEA* in Čejč (Czech Republic), were sown to low planting dishes with soil and placed in the temperated glasshouse (21-25/18-19 °C day/night, 80-85 % air humidity, no additional irradiance). Each genotype was represented by 90-100 plants. After 10 d of this treatment, one half of the seedlings was placed in the non-temperated glasshouse (11-18/-5 to +5 °C day/night, 80-85 % air humidity, no additional irradiance). Leaves of 4 to 6 week-old seedlings were used for the measurements of photosynthetic characteristics. The first fully developed leaf (counting from the top) was taken from each plant; 10 to 13 plants were usually needed to get sufficient amount of leaf tissue.

The isolation of chloroplasts from maize leaves and measurements of HRA and PS1 activity was performed as shown in Holá *et al.* (1999). The final chloroplast suspensions contained mostly mesophyll chloroplasts with broken envelope membranes, which still showed high photochemical activity. Photochemical activity of isolated mesophyll chloroplasts was measured polarographically as Hill reaction activity (HRA) and PS1 activity, *i.e.*, the amount of oxygen formed (or, in case of PS1 activity, consumed) by the suspensions of isolated chloroplasts irradiated by " "white light" (170 W m^{-2} PAR) after the addition of artificial electron acceptors or donors. We used 7 mM $K_3[Fe(CN)_6]$ as electron acceptor in case of HRA, and 0.15 mM reduced DCPIP as electron donor, 0.1 mM methylviologen as electron acceptor, and 0.1 mM DCMU as PS2 activity inhibitor in case of PS1 activity. Each sample, representing one genotype in one treatment, was measured two to four times and the values were expressed per leaf area (LA) or Chl content units and time unit.

For determination of contents of photosynthetic pigments, six leaf discs, each corresponding to 0.5 cm^2, were cut into small pieces which were put into 10 cm^3 of N,N-dimethylformamide and stored in a dark and cool place. After 2 d, Chl *a*, Chl *b*, and total carotenoid (Car) contents in the extracts was determined spectrophotometrically (*Spekol 211, Carl Zeiss*, Jena, Germany) (Porra *et al.* 1989, Wellburn 1994). Each genotype/treatment was represented by three samples and the values were expressed per LA unit. The ratios of Chl *a/b* and Chl/Car were also calculated.

Two series of experiments with the same experimental pattern were done, in April and October. The average values characterising each genotype/treatment on each experimental day were used for the genetic analysis of photosynthetic characteristics. The differences between genotypes were analysed using Student's *t*-test. A generation mean analysis according to Hayman (1958) was applied to determine the genetic mechanisms of the inheritance of photosynthetic characteristics in the two hybrid combinations examined. Each hybrid combination was first analysed by the simple additive-dominance model, and the adequacy of this model was tested by χ^2 test. The extended genetic model was then applied to examine the possibility of existence of additive maternal effects. The statistical computations were made with the aid of CBE computer programs (Wolf 1996).

Results

The content of Chls in leaves of the plants stressed by low growth temperature was significantly lower than that of the non-stressed plants in all genotypes examined. This applied to the Chl *a*, Chl *b*, and their sum. The Chl/Car ratios were also lower in the stressed plants compared to the non-stressed ones, whereas the ratio of Chl *a/b* did not change on the treatment. The total Car content was usually not affected by low growth temperature. As for HRA, the inbred lines and their F_1 hybrids responded somewhat differently to low growth temperature. The decrease of HRA values from non-stressed to stressed plants was especially pronounced in the inbred lines, while the differences between both treatments were usually not statistically significant in the hybrids. The activity of PS1 did not show any response to low temperature treatment, or even (when expressed per Chl content unit) significantly increased in stressed plants (Fig. 1).

The analysis of photosynthetic characteristics of maize leaves in both hybrid combinations revealed high intraspecific variation due to genotypic differences for HRA and the content of photosynthetic pigments. Such variation, however, was usually not found for PS1 activity. The differences between inbred lines mostly decreased with low temperature treatment. Among inbred lines, the highest contents of photosynthetic pigments, the highest values of Chl *a/b* or Chl/Car ratios, and the highest HRA were observed for CE810. Inbred line CE777 had low Chl content but displayed rather high photochemical activity of isolated mesophyll chloroplasts, while for CE704 the situation was reverse.

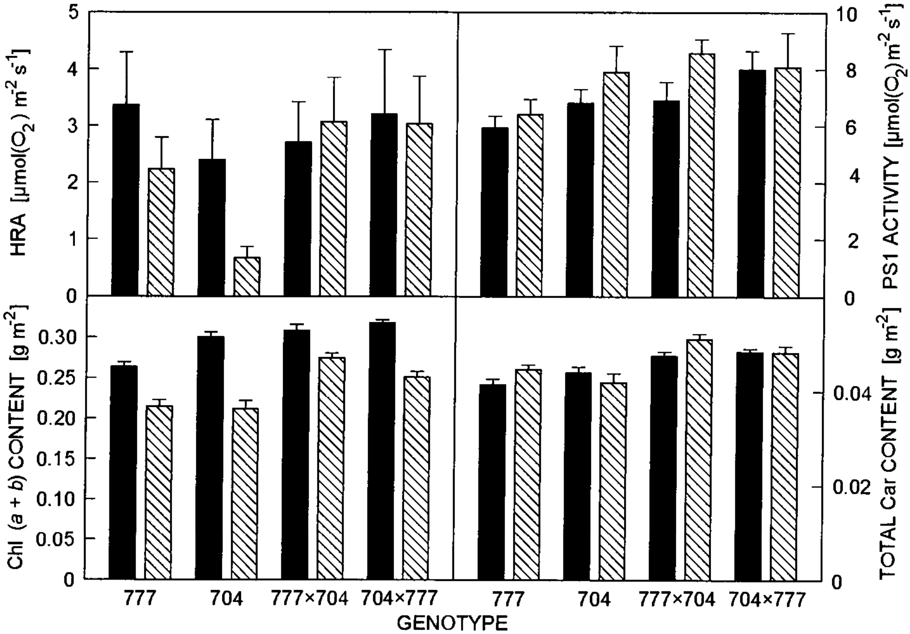

Fig. 1. The effects of low growth temperature on Hill reaction activity (HRA), photosystem (PS) 1 activity, chlorophyll (Chl) and total carotenoid (Car) contents in the leaves of maize inbreds CE777 (777) and CE704 (704), and their reciprocal F_1 hybrids. Means ± SEM are shown. *Hatched bars* represent plants stressed by low growth temperature, *solid bars* represent non-stressed plants.

The reciprocal F_1 hybrids did not significantly differ in either HRA, PS1 activity, or ratios of photosynthetic pigments. However, statistically significant differences between F_1 hybrids in the Chl or Car contents were found for the stressed plants of hybrid combination CE777xCE704 in both experimental series. Similar differences were observed between the non-stressed plants of genotypes CE704xCE810 and CE810xCE704 in the second series of experiments.

The differences between inbreds and hybrids were more pronounced in the stressed plants compared to the non-stressed ones. The positive heterotic effect in F_1 generation was observed for photosynthetic pigments' content in plants under both treatments. On the other hand, the positive heterosis for HRA or PS1 activity was found in stressed plants only and was higher in hybrid combination CE777xCE704 compared to CE704xCE810. The dramatic increase of heterosis in plants subjected to low growth temperature was found especially in HRA (from about 95 to 200 or 170 % of mid-parent value in the hybrid combinations CE777xCE704 or CE704xCE810, respectively). As for Chl content, the mid-parent heterosis in F_1 generation was also usually higher in stressed plants than in the non-stressed ones. This was noted especially for the content of Chl (*a*+*b*) and Chl *a*; the differences in Chl *b* content were less prominent. The heterotic effect in the content of total Car did not change with temperature treatment (Fig. 2).

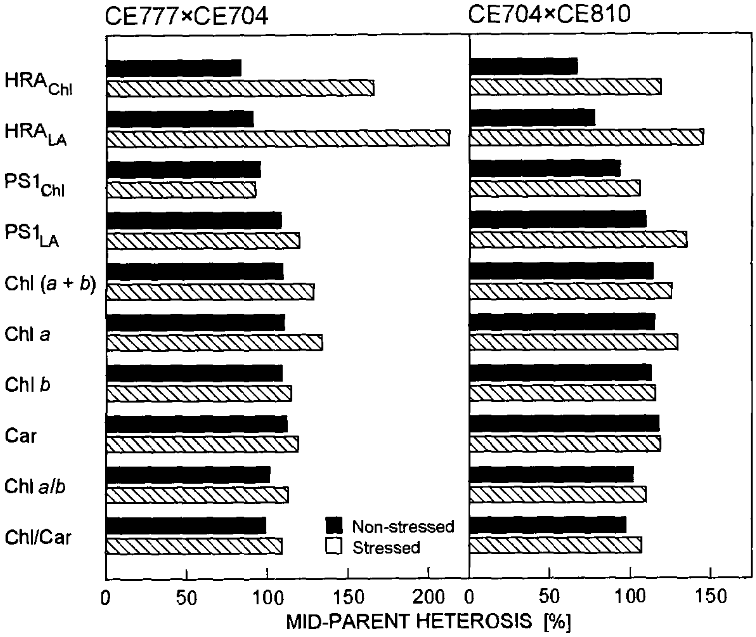

Fig. 2. The mid-parent heterosis in Hill reaction activity (HRA), photosystem (PS) 1 activity [both expressed per leaf area unit (LA) or chlorophyll (Chl) content unit], the contents of Chl (*a*+*b*), Chl *a*, Chl *b*, and total carotenoids (Car), and the ratios of photosynthetic pigments, displayed by F_1 hybrids CE777×CE704 (*left*) or CE704×CE810 (*right*) of maize stressed or non-stressed by low-growth temperature.

The positive heterosis in photosynthetic characteristics of maize leaves was caused by the positive dominance in both hybrid combinations. Genetic analysis also demonstrated the importance of additive genetic effects for the intraspecific variation in photochemical activity of isolated mesophyll chloroplasts and in the contents of photosynthetic pigments. The additive effects were the most important component of genetic variation in HRA and PS1 activity, while the prominent role of positive dominance was found especially in the content or ratio of photosynthetic pigments. The nature of genetic effects was the same for both hybrid combinations examined, with the exception of PS1 activity, where the presence of additivity and (in some cases) positive dominance was confirmed for CE704×CE810 only (Tables 1 and 2).

Comparison of the genetic effects participating in the inheritance of HRA between the stressed and non-stressed plants showed the dramatic increase in importance of positive dominance in plants stressed by low growth temperature. Contrary to this, the genetic mechanisms responsible for the intraspecific variation in PS1 activity did

Table 1. Genetic analysis of photosynthetic characteristics in hybrid combination CE777×CE704 of maize stressed and non-stressed by low growth temperature. Two experimental series. For abbreviations see legend to Fig. 2. The estimates of genetic parameters (±SEM) after Hayman (1958) are shown together with their statistical significance ($^{**}p = 0.01$, $^{*}p = 0.05$).

	Non-stressed			Stressed		
	Mean	Additivity	Dominance	Mean	Additivity	Dominance
Series 1						
HRA (Chl)	11.91±0.51**	2.92±0.56**	-1.84±1.01	11.73±0.34**	4.78±0.55**	6.15±0.68**
HRA (LA)	3.56±0.21**	0.62±0.20**	-0.21±0.92	2.86±0.07**	0.97±0.11**	2.07±0.13**
PS1 (Chl)	23.51±0.79**	0.04±1.08	0.87±1.59	30.98±0.88**	-3.17±1.31**	-2.09±1.77
PS1 (LA)	6.98±0.27**	-0.46±0.25	1.41±0.53**	7.78±0.29**	-0.77±0.45	1.40±0.58**
Chl (*a*+*b*)	30.05±0.25**	-1.88±0.41**	3.72±0.49**	24.16±0.34**	0.04±0.57	5.56±0.68**
Chl *a*	23.05±0.19**	-1.39±0.32**	2.98±0.37**	18.42±0.32**	0.15±0.55	4.93±0.04**
Chl *b*	6.98±0.06**	-0.49±0.10**	0.69±0.12**	5.67±0.15**	-0.10±0.30	0.50±0.30
Car	4.49±0.04**	-0.14±0.07*	0.56±0.08**	4.60±0.06**	0.12±0.10	0.67±0.13**
Chl *a/b*	3.28±0.001**	0.03±0.01*	0.06±0.03*	3.25±0.08**	0.10±0.05	0.38±0.15**
Chl/Car	6.68±0.03**	-0.23±0.04**	-0.07±0.07	5.22±0.04**	-0.14±0.06*	0.38±0.08**
Series 2						
HRA (Chl)	18.82±0.60**	1.86±0.80*	-3.05±1.19**	14.26±0.86**	4.14±1.31**	4.01±1.71*
HRA (LA)	5.93±0.21**	0.06±0.26	-0.28±0.41	3.59±0.21**	0.94±0.27**	1.07±0.42**
PS1 (Chl)	29.11±1.40**	3.69±2.65	-2.35±2.79	50.14±2.41**	0.99±4.13	-7.07±4.82
PS1 (LA)	9.03±0.35**	0.06±0.58	1.45±0.71*	10.82±0.51**	0.71±0.74	1.90±1.01
Chl (*a*+*b*)	31.73±0.36**	-2.74±0.42**	5.35±0.72**	23.17±0.41**	0.47±0.71	5.52±0.81**
Chl *a*	24.47±0.28**	-2.23±0.32**	4.23±0.56**	17.86±0.29**	0.35±0.48	4.32±0.58**
Chl *b*	7.25±0.08**	-0.51±0.11**	1.11±0.17**	5.31±0.07**	0.12±0.13	1.22±0.15
Car	4.74±0.04**	-0.24±0.04**	0.69±0.08**	4.68±0.08**	0.18±0.14	0.75±0.17**
Chl *a/b*	3.38±0.01**	-0.08±0.01**	0.07±0.02**	3.38±0.01**	-0.01±0.02	0.08±0.03**
Chl/Car	6.63±0.04**	-0.27±0.07**	0.18±0.07**	4.98±0.03**	-0.09±0.05	0.44±0.06

not differ between both treatments. As for the photosynthetic pigments' contents, the additive genetic effects were statistically significant in non-stressed plants, but non-significant in stressed plants of the hybrid combination CE777xCE704 (Table 1). In this case, the simple additive-dominance model did not fully suit the requirements of genetic analysis, and the maternal additive effects were therefore also included into the model. These effects (Table 3) were statistically significant and positive for the Chl and Car contents in plants grown under low temperature. The maternal additive effects participated in the accumulation of photosynthetic pigments in the second series of experiments with hybrid combination CE704xCE810, too. In this case, however, their value was negative and statistically significant in non-stressed plants only (Table 3).

Discussion

The analysis of photochemical activity of isolated mesophyll chloroplasts and

Table 2. Genetic analysis of photosynthetic characteristics in hybrid combination CE704×CE810 of maize stressed and non-stressed by low growth temperature. Two experimental series. For abbreviations see legend to Fig. 2. The estimates of genetic parameters (±SEM) after Hayman (1958) are shown together with their statistical significance ($^{**}p = 0.01$, $^{*}p = 0.05$).

	Non-stressed Mean	Additivity	Dominance	Stressed Mean	Additivity	Dominance
Series 1						
HRA (Chl)	11.83±0.95**	-3.81±0.66**	-3.76±1.91*	13.85±0.83**	-9.36±0.67**	1.23±1.66
HRA (LA)	4.08±0.36**	-1.36±0.23**	-0.66±0.71	3.49±0.21**	-2.13±0.13**	1.02±0.43**
PS1 (Chl)	27.11±1.50**	-4.43±1.89*	-0.68±3.00	38.99±1.84**	-1.48±2.67	4.64±3.68
PS1 (LA)	8.57±0.56**	-1.29±0.48**	1.07±1.13	9.71±0.52**	-0.25±0.74	3.18±1.04**
Chl (*a*+*b*)	33.03±0.41**	-0.51±0.46	4.86±0.82**	24.48±0.31**	-0.37±0.50	5.52±0.62**
Chl *a*	25.40±0.32**	-0.56±0.37	3.80±0.65**	18.81±0.28**	-0.68±0.46	4.64±0.56**
Chl *b*	7.60±0.09**	0.06±0.10	1.06±0.18**	5.64±0.12**	0.30±0.22	0.84±0.63**
Car	4.87±0.06**	-0.11±0.07	0.84±0.18**	4.81±0.07**	-0.26±0.09**	0.82±0.14**
Chl *a/b*	3.36±0.02**	-0.11±0.02**	0.06±0.03	3.36±0.06**	-0.27±0.11**	0.26±0.12*
Chl/Car	6.78±0.03**	0.07±0.04	-0.19±0.06**	5.10±0.04**	0.21±0.05**	0.27±0.09**
Series 2						
HRA (Chl)	17.70±0.46**	-0.49±0.81	-2.53±0.92**	13.97±0.72**	-3.16±1.43*	5.40±1.44**
HRA (LA)	6.55±0.17**	-0.64±0.28*	-0.20±0.33	3.66±0.18**	-0.86±0.34**	1.99±0.35**
PS1 (Chl)	28.35±0.86**	-2.80±1.56	-2.09±1.72	52.63±1.68**	-2.38±3.02	-4.85±3.35
PS1 (LA)	9.91±0.37**	-1.69±0.58**	-0.04±0.74	11.38±0.31**	-1.44±0.54**	1.57±0.62**
Chl (*a*+*b*)	36.67±0.19**	-2.47±0.27**	4.82±0.39**	23.61±0.46**	-1.34±0.66*	4.66±0.91
Chl *a*	28.44±0.16**	-2.03±0.23**	3.65±0.33**	18.33±0.36**	-1.15±0.51*	3.67±0.71**
Chl *b*	8.19±0.04**	-0.45±0.06**	1.10±0.09**	5.28±0.10**	-0.20±0.15	1.01±0.20**
Car	5.38±0.04**	-0.38±0.05**	0.67±0.08**	4.78±0.11**	-0.35±0.15**	0.63±0.21**
Chl *a/b*	3.47±0.01**	-0.06±0.02**	-0.02±0.03	3.47±0.01**	-0.10±0.02**	0.05±0.02*
Chl/Car	6.79±0.02**	0.01±0.03	-0.02±0.02	4.62±0.04**	0.08±0.06	0.31±0.07**

contents of photosynthetic pigments in two maize hybrid combinations has revealed a significant decrease of Chl content and HRA in leaves of plants stressed by low growth temperature. Contrary to this, the activity of PS1 usually did not change with cold treatment. This agrees with the findings of Nie and Baker (1991), who studied the effect of low temperature on the development of thylakoids in maize leaves. They observed a great reduction of PS2 activity but not of PS1 activity, which is associated with the reduction in PS2 reaction centre (RC) complexes. The accumulation and stability of PS2 RC proteins, especially D1 protein, may be severely affected by cold stress. These changes in pattern of thylakoid proteins probably favour the cyclic electron transport around PS1. As HRA is, to some extent, a measure of the PS2 activity (or, rather, of whole photosynthetic electron-transport chain activity), it is not surprising that its values in cold stressed plants were reduced compared to the non-stressed ones in our study.

The reduction in the content of Chls is another phenomenon associated with the low-temperature stress in maize (Baker *et al.* 1983, Haldiman 1997, *etc.*). The content of Chl *a* is the most strongly affected, which is probably connected to the

Table 3. Genetic analysis of photosynthetic characteristics in hybrid combinations CE777×CE704 and CE704×CE810 of maize. Plants were (CE777×CE704) or were not (CE704×CE810) stressed by low growth temperature. The maternal additive effects were included in the model. The estimates of genetic parameters (±SEM) after Hayman (1958) are shown together with their statistical significance ($^{**}p = 0.01$, $^{*}p = 0.05$).

	Mean	Additivity	Dominance	Maternal additive
CE777×CE704, series 1				
Chl (*a*+*b*)	23.98±0.35**	-1.07±0.69	5.20±0.69**	1.11±0.39**
Chl *a*	18.28±0.32**	-0.71±0.65	4.64±0.65**	0.85±0.35**
Chl *b*	5.70±0.15**	-0.35±0.30	0.56±0.30	0.26±0.06**
CE777×CE704, series 2				
Chl (*a*+*b*)	22.86±0.41**	-1.69±0.82*	4.91±0.82**	2.16±0.42**
Chl *a*	17.65±0.29**	-1.37±0.59*	3.90±0.59**	1.72±0.34**
Chl *b*	5.21±0.08**	-0.32±0.15*	1.02±0.15**	0.44±0.08**
Car	4.60±0.09**	-0.21±0.17	0.59±0.17**	0.39±0.10**
CE704×CE810, series 2				
Chl (*a*+*b*)	36.05±0.25**	-0.82±0.49	3.58±0.49**	-1.65±0.41**
Chl *a*	27.99±0.20**	-0.74±0.41	2.75±0.41**	-1.29±0.34**
Chl *b*	8.06±0.05**	-0.09±0.11	0.83±0.11**	-0.36±0.09**
Car	5.31±0.05**	-0.18±0.09	0.54±0.09**	-0.20±0.97**

decreased amount and stability of thylakoid pigment-protein complexes. The decrease in the content of light-harvesting antenna proteins (LHC) which bind both Chl *a* and *b* has also been well documented (Nie and Baker 1991, Bredenkamp *et al.* 1992). The reduction of Chl *b* amount, observed in our case, is therefore expected. Nevertheless, the Chl *a/b* ratio did not significantly change with the temperature treatment, even if the content of PS2 RC should be more reduced relative to LHC (Nie and Baker 1991, Yordanov 1992). However, Chl *a* is not exclusively bound to PS2 RC proteins, but (in much greater amounts) to PS1 and LHC proteins as well, which could explain the above fact. On the other hand, the ratio Chl/Car was lower in stressed plants than in the non-stressed ones.

We did not find any significant response to cold stress in the content of Car in maize leaves. However, it is impossible to draw any definitive conclusion from these results. According to Haldimann (1997), the exposition of plants to low temperature for several days during the early vegetative growth stages resulted in the decreased content of β-carotene, increase in contents of xanthophyll cycle pigments (together with the accelerated conversion of violaxanthin to zeaxanthin), and no effect on the contents of lutein or neoxanthin. A future detailed analysis of individual Car could bring more information about the effects of cold stress on these photosynthetic pigments in different maize genotypes.

The positive mid-parent heterosis in F_1 generation observed for both HRA and contents of photosynthetic pigments in maize confirms both our previous findings (Körnerová *et al.* 1998, Holá *et al.* 1998, 1999, Holá 1999) and the results of other authors (Kočová 1992, Mehta *et al.* 1992, Titok *et al.* 1994, Qi *et al.* 1995, Krebs *et al.* 1996). Contrary to it, the intraspecific variation in PS1 activity due to genotypic differences has been rather small. This was true also for the Chl *a/b* ratio which was in good agreement with other studies (Gimenez *et al.* 1992, Wilcox and Koller 1992, Pettigrew and Meredith 1994, Janssen *et al.* 1995, Krebs *et al.* 1996, Synková *et al.* 1997, Quinones *et al.* 1998). The heterotic effect was caused by strong positive dominance. The additive component of genetic variation was also statistically significant. These findings were supported for example by the results of Fousová (1971), Baer and Schrader (1985), Mehta *et al.* (1992), or Krebs *et al.* (1996), who studied the genetic basis of heterosis in the photosynthetic pigments' content or the functional characteristics of thylakoid membranes in maize.

The most important result of this study is the evidence that F_1 hybrids of maize can respond differently to low-temperature stress than their parental lines as concerns HRA and, to some extent, Chl content. The outstanding increase of heterosis in HRA in the stressed plants, as well as the changes of absolute values of this characteristic in hybrids and inbreds after cold treatment support the hypothesis of increased adaptive ability displayed by the F_1 generation. As the response of PS1 activity to low temperature is not so prominent, it is not surprising that we did not observe such effect on this characteristic.

Comparison of the results of genetic analysis of photosynthetic characteristics in plants stressed and non-stressed by low growth temperature has shown that the expression of genetic effects becomes more pronounced with cold treatment of plants. The degree of dominance has strongly increased for HRA and Chl content. The disappearance of statistically significant additive genetic effects in stressed plants of hybrid combination CE777×CE704 was caused by shift in the respective genetic parameter estimates from negative and rather large to positive, but small, values. Another example of the changes in expression of genetic effects is the presence of additive maternal effects which were found for the contents of photosynthetic pigments in stressed plants of hybrid combination CE777×CE704. Such importance of extranuclear genetic information in the inheritance of photosynthetic characteristics has not yet been definitely established. Maternal effects in the inheritance of the contents of Chls were already described (Fousová 1971, Synková *et al.* 1997). Others claim that such effects are of no special importance in the determination of this characteristic (Baer and Schrader 1985, Kirchhoff *et al.* 1989, Krebs *et al.* 1996). It is just possible that the maternal effects become more pronounced under the conditions which are not optimal for photosynthesis, as it was in our case.

We found that the response of photosynthetic characteristics to low temperature treatment of plants is not always the same in F_1 hybrids and their parental inbred lines of maize. The dramatic increase of mid-parent heterosis in HRA, accompanied by similar changes in the Chl content, was observed in cold-stressed plants compared to the non-stressed ones. These changes are caused by the higher degree of positive dominance together with the increase in expression of some other genetic effects.

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