EFFECT OF CHILLING AND ACCLIMATION ON THE ACTIVITY OF GLUTAMINE SYNTHETASE ISOFORMS IN MAIZE SEEDLINGS

ANA D. SIMONOVIĆ1,2 and M. D. ANDERSON1

1Department of Biological Sciences, North Dakota State University, Fargo, ND 58105, USA
2Department of Plant Physiology, Siniša Stanković Institute for Biological Research, 11060 Belgrade, Serbia

Abstract – Effects of chilling and acclimation on the activity of cytosolic (GS1) and plastidic (GS2) isoforms of glutamine synthetase (E.C. 6.3.1.2) were studied in chilling-sensitive and acclimation-responsive maize inbred G50. Glutamine synthetase activity in mesocotyls and roots of chilled (7 d/4°C) and rewarmed (1 d/27°C) etiolated plants was ≈1/3 that of controls. In coleoptiles+leaves of light-grown plants, GS1 was reduced to 75%, and GS2 to 50%. Acclimation (3 d/14°C) increased GS activity and alleviated the effects of chilling. Exposure to H2O2 or menadione also reduced GS activity. Since chilling causes oxidative stress in maize, acclimation probably preserves GS activity by protecting GS from oxidative inactivation.

Key words: Acclimation, chilling, glutamine synthetase, maize, oxidative stress, phosphate precipitation

INTRODUCTION

Chilling injury occurs in the absence of ice nucleation in plant cells between 0°-12°C. It is a two-stage process, where a primary event, which might include changes in membrane fluidity, protein denaturation, reduction of metabolic rates, and alteration of the cytoskeleton, is followed by a series of secondary events (Raison and Lyons, 1986; Raison and Orr, 1990). Chilling may cause disfunction in respiration, photosynthesis, chlorophyll synthesis, amino acid incorporation, ethylene production, phenolic metabolism, sugar metabolism, and redox regulation (Raison and Orr, 1990; Anderson et al., 1994). Chilling delays the circadian pattern of nitrate reductase activity in tomato (Jones et al., 1998) but, to our best knowledge, no reports on the effect of chilling on other N-assimilating enzymes have been published.

The molecular basis of chilling injury and tolerance in maize has been studied by comparing cultivars with different degrees of chilling sensitivity (Kocsy et al., 1996; Hodges et al., 1997; Kaniuga et al., 1999; Lepner et al., 1999) and by comprehensive study of low temperature-induced biochemical changes within one genotype, Pioneer G50. In G50, chilling at 4°C for 7 days in darkness induces significant H2O2 increase in shoots, which promotes oxidation of proteins and lipids, inducing severe oxidative stress (Prasad et al., 1994a; Prasad, 1996). An acclimation phenomenon has been characterized in this system, during which antioxidative enzymes, such as catalase and peroxidases are induced (Anderson et al., 1994; 1995; Prasad et al., 1994b).

The aim of this work is to assess the effects of low temperatures on activity of the core enzyme of nitrogen metabolism, glutamine synthetase (GS, E.C. 6.3.1.2), using a well-described experimental system, the G50 inbred. Plant GS enzymes are octamers with subunits of about 40 kD. Higher plants have one plastidic (GS2) and one or more cytosolic (GS1) isoforms (Hirel and Gadal, 1980; Lam et al., 1996). In maize, GS isoforms are encoded by six nuclear genes, one for GS2 (Snustad et al., 1988) and five for cytosolic isoforms, named GS1-1 through GS1-5 (Sakakibara et al., 1992a; Li et al., 1993). The GS2 isoform is expressed primarily in green leaves, where it is involved in reassim-
ilation of photorespiratory ammonia, but is also implicated in nitrate assimilation in both leaves and roots (Sakakibara et al., 1992a, b; Li et al., 1993; Redinbaugh and Campbell, 1993). The major cytosolic isoforms are GS1-3 and GS1-4, which are constitutively expressed throughout the plant, and a pair of root isoforms, GS1-1 and GS1-5 (Sakakibara et al., 1992a, b; 1996; Li et al., 1993). The GS1-2 isoform is found in vascular root tissues (Li et al., 1993) and is involved in nitrogen remobilization during the grain fill (Mudgett, 2003).

The effects of chilling, rewarming and acclimation on the activities of GS isoforms were studied in roots, mesocotyls and coleoptiles + leaves (c+l). Comparative study of etiolated and light-grown plants was performed because light often exaggerates the effects of chilling (van Hasselt, 1990). The results are discussed in relation to chilling-induced oxidative stress and acclimation-induced antioxidative defense.

MATERIALS AND METHODS

Chilling and acclimation treatments

Maize inbred G50 was grown in pots with Sunshine Germinating Mix #3 in darkness or continuous white light under eight different temperature regimes (Fig. 1). Since maize grows very little during acclimation at 14/16°C, and not at all during chilling at 4°C, then A, C, and even 14 day-old AC plants can be considered to be at the same developmental stage as G4 controls (Anderson et al., 1994). Likewise, AR, CR, and ACR plants can be considered to be at the same stage as G5 plants. The intensity of combined fluorescent and incandescent white light in growth chambers was 100 mmol m⁻² s⁻¹ in the 27°C chamber, 80 mmol in the acclimation chamber, and 220 mmol in the chilling chamber.

Hydrogen peroxide and menadione treatments

Seeds were germinated in Petri dishes in darkness at room temperature. Three-day-old plants, ≈5 cm long, were wrapped in kimwipes and inserted in tubes with H₂O₂ or menadione solutions to the seed level, so that roots were completely immersed, while shoots were in contact with the wet paper. Plants were exposed to oxidants for 48 hours at 27°C in darkness or in the light. At least five plants were used for each treatment, and experiments were repeated three times.

Protein extraction, GS assay and data analysis

Soluble proteins were extracted from frozen c+l, mesocotyls, or roots, separated by native PAGE and assayed for GS activity as described earlier (Simonovíc et al., 2004). Plastids were isolated from fresh roots of dark-grown 6 days old plants using a Percoll gradient (Esposito et al., 2003). The activity of a plastidic marker, alkaline pyrophosphatase (APPase, E.C. 3.6.1.1), in crude and purified preparations was used to calculate the yield of plastids. It was assayed in a system containing 2.5 mM sodium pyrophosphate, 5 mM MgCl₂, and 50 mM Tris-HCl, pH 8.6 (Hemalatha and Prasad, 2002), followed by determination of released orthophosphate (Chifflet et al., 1988). The obtained plastid pellet (≈1.5 ml) was washed once in 30 ml of plastid buffer (50 mM Tricine-KOH, pH 7.9, 330 mM sorbitol, 4 mM EDTA, and 1 mM MgCl₂) to eliminate Percoll and cytosolic contaminations, recovered (7 min at 4,000g and 4°C), and used for protein extraction and GS assay by the same procedure as for frozen tissue samples. The optical

Fig. 1. Temperature treatments. G4–control plants, germinated for 4 days at 27°C; A–G4 plants acclimated for additional 3 days at 14°C (in darkness) or 16°C (in light); C–G4 plants chilled at 4°C for 7 days; AC–G4 plants acclimated for 3 days and chilled for 7 days; GS–plants grown at 27°C for 5 days; AR–acclimated (A) plants that were rewarmed for 1 day at 27°C; CR–chilled (C) plants rewarmed for 1 day; ACR–acclimated and chilled (AC) plants that were rewarmed.

Fig. 2. Activity of cytosolic (GS1) and plastidic (GS2) isoforms of glutamine synthetase in crude preparations of soluble cellular proteins from roots of dark-grown plants (C, 100 mg) and soluble proteins from plastids from of same tissue (P, 15 mg).
density of bands on GS zymograms was quantified using ImageJ software (version 1.32J, National Institutes of Health, USA). To minimize gel-to-gel differences, the background was subtracted, and data were normalized by setting values of GS1 activities in the control (G4) samples as 100% activity. All data are average values of three independent repeats.

RESULTS AND DISCUSSION

GS activity in tissue and plastid protein extracts

The GS assay revealed one activity band for mesocotyls and two for c+l and roots (Figs. 3-8). That both bands represent GS activity was confirmed earlier by sensitivity to L-methionine-sulfoximine, insensitivity to L-buthionine-S,R-sulfoximine (Simonović et al., 2004), and complete inhibition by glufosinate (data not shown). To assign the two activity bands to GS1 and GS2 isoforms, we isolated plastids from roots of dark-grown plants and compared the GS activity profile of plastidic proteins to that of total proteins from the same tissue (Fig. 2). The plastidic sample (15 mg) had the same intensity of the upper band as the total protein sample (100 mg), a nearly seven-fold enrichment, and virtually lacked the lower band, demonstrating that the upper band is GS2. This is in agreement with migration of maize GS isoforms on SDS-PAGE (Sakakibara et al., 1992a, b; Becker et al., 2000). Since the MW difference between GS2 and GS1 monomers is ≈4 kDa (Becker et al., 2000), the isoforms could be separated in their native, octameric, forms only after prolonged electrophoresis at high voltage.

In G4 plants, relative GS2 activity in c+l was only 20% and 30% of that of GS1 for dark- and light-grown plants, respectively (Figs. 3 and 6). From comparative studies of Panicum species having C₃, C₄, and intermediate photosynthetic pathways, it was concluded that the high GS1:GS2 ratio in the leaves of C₄ plants is correlated with C₄ metabolism (Hirel et al., 1983). The GS2 activity in green c+l (Fig. 6) was higher than in etiolated ones (Fig. 3), due to light induction of GS2 in leaves (Sakakibara et al., 1992a, b; Lam et al., 1996). No GS2 was detected in mesocotyl samples (Figs. 4 and 7). Sakakibara et al. (1992a) did not find GS2 protein in mesocotyls either. Relative GS2 activity found in roots of dark-grown plants (Fig. 5) was higher in comparison to roots of light-grown plants (Fig. 8). The regulation of GS2 expression is not only organ-specific, but also cell-specific, while GS2 isoforms isolated from different cells have different biochemical characteristics (Sakakibara et al., 1992b; Redinbaugh and Campbell, 1993; González-Moro et al., 2000). Light induction of GS2 is characteristic of leaves, where it is restricted to bundle sheath cells (Sakakibara et al., 1992b). Our results indicate an inhibitory effect of light on root GS2.

We were unable to separate the individual GS1 iso-
forms. The coding regions of the five cytosolic GS cDNAs exhibit 80-96% sequence identity (Li et al., 1993), and their monomer MW ranges from 38.981 for GS1-4 to 40.094 Da for GS1-2, making the separation by native PAGE and activity staining virtually impossible. The GS1 activity band found in c+l (Figs. 3 and 6) was sharp, corresponding to one GS1 band detected by immunoblots in leaves (Sakakibara et al., 1992a, b; Becker et al., 2000). The smear-like activity of GS in mesocotyls, especially in light-grown plants (Fig. 7), indicates the presence of abundant isoforms, likely GS1-3 and GS1-4, whose transcripts have been detected in this organ (Sakakibara et al., 1992a). The GS1 activity bands for root samples were broad (Figs. 5 and 8), corresponding to the overlapping activity of all GS1 isoforms whose transcripts were found in roots (Li et al., 1993), while Western analysis revealed two protein bands labeled as GS1 and Gsr, which are products of the GS1-3/GS1-4 and GS1-1/GS1-5 genes respectively (Sakakibara et al., 1992b, 1996).

Effect of chilling and rewarming on GS activity

The effect of chilling for 7 days at 4°C on GS activities was estimated at the end of the chilling period (C) and after rewarming for one day at 27°C (CR). Exposure to chilling reduced GS activity in a light-modulated and organ-specific manner. In dark-grown plants, the non-photosynthetic organs, mesocotyls and roots, were most affected. Chilling did not reduce GS1 activity in etiolated coleoptiles (Fig. 3) and caused only a 2% reduction of GS2, but CR treatment significantly reduced GS2 activity (19%) in comparison to G5 plants (39%). In mesocotyls, the effect of chilling was pronounced, C plants having 66% and CR plants less than 40% of control activity (Fig. 4). In roots of dark-grown plants, C and CR treatments caused reduction of GS1 to 48% and 42%, respectively, while GS2 activity after these treatments was barely detectable (Fig. 5).

It has been shown that chilling at 4°C for 7 days in darkness induced severe oxidative stress in G50 seedlings, that only 2% of plants were capable of surviving, and that chilling injury was most obvious in mesocotyls (Anderson et al., 1994; Prasad et al., 1994a). Chilling causes significant \( \text{H}_2\text{O}_2 \) increase in shoots, but not in roots (Prasad et al., 1994a; Anderson et al., 1995). In heterotrophic maize seedlings, mitochondria are very active in supporting growth and are also a major source of superoxide and \( \text{H}_2\text{O}_2 \) in the cell (Prasad et al., 1994a). Chilling impairs respiratory activity, the cytochrome pathway and ATPase activity, resulting in greater production of ROS in mitochondria (Prasad et al., 1994a, b). Chilled G50 seedlings have 2-3 times higher levels of oxidized protein (Prasad, 1996), and the literature suggests that GS proteins are particularly sensitive to oxidative inactivation and degradation.

Oxidative degradation of GS from different sources such as \textit{E. coli} (Levine et al., 1981; Levine 1983; Farber and Levine, 1986), yeast (Kim et al., 1985), rat liver (Fucci et al., 1983), pea chloroplasts (Stieger and Feller, 1997), wheat chloroplasts (Palatnik et al., 1999), and soybean roots (Ortega et al., 1999) apparently involves the same mechanism of metal-catalyzed site-specific oxidation, followed by dis-
crete fragmentation of the enzyme, as elaborated by Stadtman (1993) and Palatnik et al., (1999). It is likely that this mechanism is responsible for the observed chilling-induced deactivation of GS.

Chilling damage in G50 seedlings is observed in dark- rather than in light-grown seedlings (Prasad et al., 1994a), and chilling under a 16 h light/8 h dark cycle is not lethal (unpublished data). In this work, plants were grown in continuous light to assess whether excess light would influence the effect of chilling on GS degradation. In light-grown plants, C and CR treatments reduced GS1 activity in c+I to 88% and 74%, respectively, while CR samples showed nearly two-fold reduction of GS2 activity (Fig. 6). Mesocotyls of green plants appeared to be resistant to chilling (Fig. 7), but roots were affected by CR, which reduced GS1 activity to 72% (Fig. 8). The GS2 activity in roots of light-grown plants was very low for all treatments (Fig. 8).

Any condition that limits CO$_2$ fixation (chilling, drought, foliar senescence) or causes excessive PSI reduction (intense light) depletes NADP$^+$, causing O$_2$ to become the preferred electron acceptor (Stieger and Feller 1997; Palatnik et al., 1999). Maize leaves developing under chilling show an increased relative flux of photosynthetic reducing equivalents to O$_2$ via the Mehler reaction (Fryer et al., 1998; Leipner et al., 1999). Chloroplast-generated ROS cause extensive modifications of a variety of stromal and thylakoid proteins (van Hasselt, 1990; Palatnik et al., 1999). Even though C/CR treatments decreased leaf GS activities in green (Fig. 6) more than in etiolated leaves (Fig. 3), this effect was small in comparison to the extent of GS2 degradation in plants or chloroplasts exposed to oxidative stress in light described in C3 plants (Stieger and Feller 1997; Palatnik et al., 1999). The lack of a pronounced effect of light on GS inactivation in chilled maize leaves can be explained by its C4 physiology. It is known that GS2 is predominantly located in chloroplasts of BS cells (Sakakibara et al., 1992b; Gonzalez-Moro et al., 2000), where PSI is active, but oxygen (and ROS) production is low. On the other hand, GS1 is the predominant form in oxygen-generating mesophyll cells (Sakakibara et al., 1992b; Becker et al., 2000), but this isoform is sequestered in the cytoplasm and is not directly exposed to chloroplast ROS.

Light had a protective effect on GS in both mesocotyls (Fig. 7) and roots (Fig. 8). If mitochondria are the major source of ROS in heterotrophic seedlings, then the switch to autotrophic growth would indirectly alleviate chilling-imposed oxidative stress in nonphotosynthetic tissues. In addition, blue light inhibits cell extension in the mesocotyl elongation zone of maize seedlings and concomitantly induces lignification of primary walls and an increase in cell-wall stiffness (Schopfer et al., 2001). While elongated dark-grown mesocotyls, with little mechanical support, developed severe symptoms of chilling injury (discoloration and loss of turgor) and significant loss of GS activity, short and stiff mesocotyls of green seedlings appeared healthy after chilling and showed no reduction in GS. This supports the previously reported importance of acclimation-induced lignification of mesocotyls in acquisition of chilling tolerance (Anderson et al., 1995).
In all cases, CR treatment was more severe than chilling alone. Returning to non-chilling temperature after prolonged chilling exacerbates the symptoms of injury (Raison and Lyons, 1986; Raison and Orr, 1990). Most membrane damage occurs after rewarming, as shown by degradation of polar lipids (Kaniuga et al., 1999) and increased membrane leakage (Zhou et al., 2005). The observed decline in GS activity correlates with overall tissue decay, especially in dark-grown mesocotyls and roots.

**Effect of acclimation on GS activity**

Acclimation of G50 seedlings at 14°C in the dark induces mild oxidative stress, marked by transient \( \text{H}_2\text{O}_2 \) increase at the onset of acclimation, followed by induction of mitochondrial catalase 3, nine peroxidases including guaiacol peroxidase, cytC peroxidase, and two cell wall peroxidases implicated in lignification, as well as a change of the glutathione reductase isozyme profile (Prasad et al., 1994a; b; 1995; Anderson et al., 1994, 1995; Prasad, 1997). Here it was shown that 70% of acclimated plants survive chilling (Prasad et al., 1994a). The optimal temperature for acclimation of plants grown under a light/dark cycle was found to be 16°C (unpublished data).

Acclimated plants showed increased GS1 activity in all organs except green leaves, from 106% for mesocotyls in light (Fig. 7) to 136% for mesocotyls in darkness (Fig. 4). Activity of GS2 was also increased in acclimated c+l, but not in roots. Acclimation followed by rewarming (AR) significantly increased GS activity in comparison to the G5 control only in mesocotyls, while in other organs both GS1 and GS2 were at the level of the G5 control. The effect of A/AR treatments on GS accumulation might be due in part to developmental increase, even though seedlings grow very slowly at 14/16°C. Another explanation is that antioxidant enzymes induced by acclimation protect GS from physiological turnover, which is also based on oxidative degradation (Ortega et al., 1999).

Acclimation as a pretreatment to chilling (AC) alleviated damaging effects of chilling on GS activity in all cases, except in roots of etiolated plants. The acclimation was particularly successful in mesocotyls of dark-grown plants (Fig. 4), where the AC sample had 1.7 times higher GS1 activity than the C sample. If acclimation preceded CR treatment (ACR), results were even better for shoots, but not for roots. Activity of GS2 in the ACR sample of green leaves was more than two times higher in comparison with CR (Fig. 6). Again, acclimation was most successful in the most sensitive tissue – mesocotyls in darkness – where the ACR sample had three times higher GS1 activity than the CR sample (Fig. 4).

The success of acclimation in protection of GS from chilling-induced inactivation in different organs correlates with the extent of induction of antioxidative enzymes in these organs. The best response to acclimation was found in mesocotyls of dark-grown seedlings, judged by induction of cat3 transcripts, CAT3 and total catalase activity, and activity of guaiacol peroxidase and other peroxidase and glutathione reductase isoforms (Prasad et al., 1994a; Anderson et al., 1994, 1995). Elevated levels of glutathione found in acclimated seedlings may contribute to the ability of c+l to scavenge \( \text{H}_2\text{O}_2 \), but none of the antioxidative enzymes was significantly affected by acclimation in c+l or roots (Prasad et al., 1994a; b; 1995; Anderson et al., 1994, 1995; Prasad, 1997).
creasing concentrations of \( H_2 \) in our system, 3-day-old seedlings were treated with in-
temperature-moderating capacity of soil. 

Experience the same temperature as shoots, because of the 

**Effect of oxidants on GS activity**

To test whether oxidants would reduce GS activity 
in our system, 3-day-old seedlings were treated with in-
creasing concentrations of \( H_2O_2 \) or menadione (MD), a 
redox-cycling quinone that generates superoxide (Fig. 9). 
In \( c+l \), neither oxidant had an effect on GS2 activity, 
while GS1 activity even slightly increased. Even the lowest 
concentrations of \( H_2O_2 \) (1 mM) or MD (0.1 mM) had an 
effect on GS1 in mesocotyls, but increase in oxidant concentration 
did not have a significant effect. Only GS1 from roots responded in a dose-response manner, giving 
a final reduction of activity of 75% for \( H_2O_2 \)-treated 
plants and about 50% for the MD treatment in both dark-
and light-grown plants. In all cases, MD appeared to be 
about 10 times more efficient than \( H_2O_2 \). A similar experi-
ment showed that mild oxidative stress induced by low 
concentrations of \( H_2O_2 \) or MD induces cat3 expression, 
leading to chilling tolerance, just like acclimation (Prasad et al., 1994a). Thus, if the intake of applied oxidants by shoots was low due to small contact area with the 
solution, the absence of a response could be attributed to induced antioxidative mechanisms.

We have shown that the activities of GS1 and GS2 
isofoms in different maize tissues detected by in-gel 
phosphate precipitation assay are in accordance with the 
published data on distribution and abundance of maize GS monomers and transcripts detected by Western and 
Northern blotting, confirming the reliability and sensitiv-
ity of our method. Our results suggest that one of the con-
sequences of chilling stress is impairment of ammonium 
assimilation through reduction of GS activity. Since chill-
ing induces oxidative stress in maize, the most likely 
mechanism of GS inactivation is oxidative degradation of 
the enzyme. Acclimation appears to preserve GS activity 
in chilling-sensitive tissues, perhaps by protecting GS from oxidative inactivation.

**Abbreviations:** A, acclimation; AC, acclimation and chilling; ACR, acclimation, chilling and rewarming; AR, 
acclimation and rewarming; C, chilling; \( c+l \), coleoptiles+leaves; CR, chilling and rewarming; G4, control 
plants grown for 4 days; G5, control plants grown for 5 
days; GS, glutamine synthetase; GS1, cytosolic isofom 
of GS; GS2, plastidic isofom of GS; MD, menadione; 
PAGE, polyacrylamide gel electrophoresis; ROS, react-
ive oxygen species.

**Acknowledgments** – Maize germ plasm was provided by Pioneer Hi-
Bred International, Inc. This research was supported by the Cooperative 
State Research, Education, and Extension Service, U.S. Department of 
Agriculture, under agreement No. 2002-35100-12054; by a grant from 
ND-EPSCoR; and by the Ministry of Science and Environment Protec-
tion of the Republic of Serbia, (Contract No. 143031).

**REFERENCES**

isozyme profiles of catalase, peroxidase, and glutathione reduct-
tase during acclimation to chilling in mesocotyls of maize seed-

Differential gene expression in chilling-acclimated maize seed-
lings and evidence for the involvement of abscisic acid in chill-

and glutamate dehydrogenase isoforms in maize leaves: locali-
ization, relative proportion and their role in ammonium assimila-

the determination of inorganic phosphate in the presence of labi-
le organic phosphate and high concentrations of protein: Applic-

Esposito, S., Massaro, G., Vona, V., Di Martino Rigano, V., and S. Carf-
agno (2003). Glutamate synthesis in barley roots: the role of the 
plastidic glucose-6-phosphate dehydrogenase. *Planta* **216**, 639-
647.

Farber, J. M. and R. L. Levine (1986). Sequence of a peptide suscepti-
tble to mixed-function oxidation. Probable cation binding site in 

Baker (1998). Relationship between CO2 assimilation, photosy-
thetic electron transport, and active O2 metabolism in leaves of 
maize in the field during periods of low temperature. *Plant 

vation of key metabolic enzymes by mixed-function oxidation 
reactions: possible implication in protein turnover and aging. 

González-Moro, B., Mené-Petite, A., Lacuesta, M., González-Murua, 
C., and A. Munoz-Rueda (2000). Glutamine synthetase from 
mesophyll and bundle sheath maize cells: isoenzyme comple-
ments and different sensitivities to phosphonothricin. *Plant Cell 
Rep.* **19**, 1127-1134.

Hemalatha, K. P. J., and D. S. Prasad (2002). Purification, physico-
chemical properties, and subcellular location of alkaline inor-
ganic pyrophosphatase from sesame (*Sesamum indicum L.*) cot-


Steger, P. A. and U. Feller (1997). Requirements for the light-stimulat-


**EFFECT OF CHILLING IN MAIZE SEEDLINGS**

В Русском языке:

ЕФЕКАТ ХЛАЂЕЊА И АКЛИМАТИЗЦИЈЕ НА АКТИВНОСТ ИЗОФОРМИ ГЛУТАМИН СИНТЕТАЗЕ КОД КЛИЈАНАЦА КУКУРУЗА

АНА Д. СИМНОВИЋ1,2 и М. Д. АНДЕРСОН1

Одељење биолошких наука, Државни Универзитет Северне Дакоте, Фарго, НД 58105, САД

Одељење за физиологију биљака, Институт за биолошка истраживања „Синиша Станковић”, 11000 Београд, Србија

У овом раду је праћен утицај хлађења и аклимације на активност цитосолне (ГС1) и пластидне (ГС2) изоформе глутамин синтетазе (E. C. 6.3.1.2) код култивираног врста кукуруза, који је осетљив на хладноћу, али се може аклиматизовати. ГС активност у мезокотилима и кореновима етиолираних клијанаца који су хлађени (7д/4℃) и потом угрејани (1д/27℃) је била ≈1/3 контроле. У колеоптилима и листовима клијанаца гајених на светлости, ГС1 је била редукована на 75%, а ГС2 на 50%. Аклимација (3д/14℃) је довела до повећања ГС активности и ублажила ефекте хлађења. Третман клијанаца пероксидом (H2O2) или менадионом је такође редуковао ГС активност. Како хлађење узрокује оксидативни стрес код кукуруза, претпостављамо да је ефекат аклимације на очувању ГС активности заснован на заштити овог ензима од оксидативне инактивације.