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Differential Recovery of Photosystem II Complex from Low-Temperature Photoinhibition in Plants with Different Chilling Sensitivity

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To examine the chilling tolerance of photosynthetic machinery in relation to membrane lipids, we compared the chilling susceptibility of photosystem II of wild type tobacco plants with that of transgenic tobacco plants, in which the sensitivity to chilling had been enhanced by genetic modification of fatty acid unsaturation of chloroplast membrane lipids. The transgenic tobacco plants were found to contain reduced levels of unsaturated membrane fatty acids by being transformed with cDNA for glycerol-3-phosphate acyltransferase from squash. For the purpose of studying on the functional integrity of photosystem II during low-temperature photoinhibition, the photochemical efficiency was measured as the ratio of the variable to the maximun fluorescence of chlorophyll (Fv/Fm) of photosystem II. In parallel with an investigation on the transgenic plants, susceptibility of chilling-resistant species, such as spinach and pea, and of chilling-sensitive ones, such as squash and sweet potato, to low-temperature photoinhibition was also compared in terms of room temperature-induced chlorophyll fluorescence from photosystem II. When leaf disks from the two genotypes of tobacco plants were exposed to light at 5℃, the transgenic plants showed more rapid decline in photochemical activity of photosystme II than wild-type plants. When they were pretreated with lincomycin, an inhibitor of chloroplast-encoded protein synthesis, the extent of photoinhibition was even more accelerated. More importantly, they showed a comparable extent of photoinhibition in the presence of lincomycin, making a clear contrast to the discrepancy observed in the absence of lincomycin. Restoration of Fv/Fm during recovery from low-temperature photoinhibition occurred more slowly in the transgenic tobacco plants than the wild-type. These findings are discussed in relation to fatty acid unsaturation of membrane phosphatidylglycerol. It appears that the ability of plants to rapidly regenerate the active photosystem II complex from photoinhibited form might explain, in part, why chilling-resistant plants can tolerate low-temperature photoinhibition.

key words: chilling sensitivity, chlorophyll fluorescence, low-temperature photoinhibition, glycerol-3-phospahate acyltransferase, fatty acid unsaturaion

INTRODUCTION

Low temperature and high-intensity light are important examples of environmental stresses on plants, and they act synergistically to induce low-temperature photoinhibition [1,2]. The photoinhibition is caused by photo-induced damage to the D1 protein of the photosystem II protein (PS II) complex [3]. Under light conditions that allow normal growth of plants, the photo-induced damage to the D_1 protein of PS II is reversed by repair system that includes degradation and *de novo* synthesis of the D_1 protein [4,5]. By contrast, under strong illumination that causes photoinhibition of the PS II complex, the rate of the photo-induced damage to the D_1 protein is higher than that of the repair and thus, the amount of active D_1 protein decreases, with resultant loss of PS II activity [6,7].

In previous studies, we demonstrated that sensitivity of tobacco plants to low-temperature photoinhibition could be modified by altering the level of unsaturated fatty acids in phosphatidylglycerol (PG) in thylakoid membranes. Such manipulation was achieved by transformation of tobacco plants with cDNA of glycerol-3-phosphate acyltransferase isolated from squash [8]. We also found that unsaturaion of fatty acids in membrane lipids accelerated the recovery of the photosynthetic machinery from photoinhibition but did not affect the process of photo-induced inactivation that is associated with photoinhibition [9,10].

The aim of the present study was to examine in further detail the sensitivity of the photosystem II to chilling temperatures of plants having different chilling sensitivity. For this purpose, we employed cabbage and spinach as chilling-resistant plants versus squash and sweet potato as chilling-sensitive ones. Moreover, in order to study on how the sensitivity of plants to low-temperature photoinhibition is affected by the degree of membrane lipid unsaturation, the

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transgenic tobacco plants were also used, whose sensitivity to chilling had been enhanced *via* a decrease in levels of unsaturated fatty acids in membrane PG, due to transformation with cDNA for glycerol-3-phosphate acyltransferase from squash [8].

In the present study, we report that tolerance of plants to low-temperture photoinhibition is supported by rapid recovery from photoinhibition.

MATERIALS AND METHODS

Plant materials Leaves from 45-day-old plants of spinach (*Spinacia oleracea* var. Glabra) and 30 day-old-plants of squash (*Cucurbita moschata* Duch var. Shirakikuza) that had been cultivated hydroponically at 22℃ under continuous light (0.2 mmol $m^{-2}s^{-1}$) and at 25℃ with 16h of light daily (0.2 mmol $m^{-2}s^{-1}$), respectively, were used for experiments. Leaves of sweet potato and cabbage were obtained from plants that had been cultivated in a green house. Wild-type and transgenic (Rbcs-SQ) tobacco plants (*Nicotiana tabacum* var. Samsun) were obtained as described previously and were grown as described [10]. Leaves from 3-month-old tobacco plants were used for experiments.

Exposure of Leaf Disks to Light at Photoinhibitory Intensity For low-temperature photoinhibition of the PS II complex, leaf disks (Φ3.8 cm) were illuminated at 1℃ by light at an intensity of 1.5 mmol $m^{-2}s^{-1}$ from two 100-W tungsten lamps (RF flood lamp; Toshiba Co., Tokyo, Japan). A 5 cm layer of circulating cold water was placed between the light source and the samples, and two fans provided continuous air circulation over the samples.

To study the role of the chloroplast genome-encoded proteins in the tolerance of leaves to photoinhibition, the extent of photoinhibition of photosynthesis was studied in the presence and in the absence of lincomycin or of chloramphenicol. Leaf disks were illuminated at a light intensity of 0.6 mmol $m^{-2}s^{-1}$ at different temperatures. Lincomycin was administered to a leaf blade through the cut end of the petiole during transpiration by immersing it in a solution of 0.6mM lincomycin for 4h at 25℃ at a light intensity of 0.1 mmol $m^{-2}s^{-1}$.

To repair the damage to the PS II complex due to photoinhibition, we incubated leaf disks after photoinhibitory treatment at 17℃ in darkness or in light at an intensity of 0.07 or 0.3 mmol $m^{-2}s^{-1}$. The intensity of light was regulated with various neutral density filters (Hoya Glass, Tokyo, Japan).

Measurement of Chlorophyll Fluorescence Measurements of the ratio of variable chlorophyll fluorescence (Fv) to maximum chlorophyll fluorescence (Fm) of leaf disks were carried out with a fluorometer (PAM-2000; Walz Co., Effeltrich, Germany) [11,12]. For these measurements, a leaf disk was placed in a chamber in which the temperature was controlled by circulating water from a refrigerated water bath. The chamber was equipped with a fiber-optic system. Ratios of Fv to Fm (Fv/Fm) were taken as a measure of the photochemical efficiency of the PS II complex [13,14].

RESULTS

Photoinhibition and Recovery in Transgenic Tobacco Plants

To gain insight into the mechanism of chilling sensitivity in the photosynthetic machinery of transgenic tobacco plants in relation to the unsaturation of membrane lipids, light-dependent changes in photochemical efficiency of photosystem II was examined in terms of chlorophyll fluorescence. Figure 1A and B show the profiles of the decline in Fv/Fm during

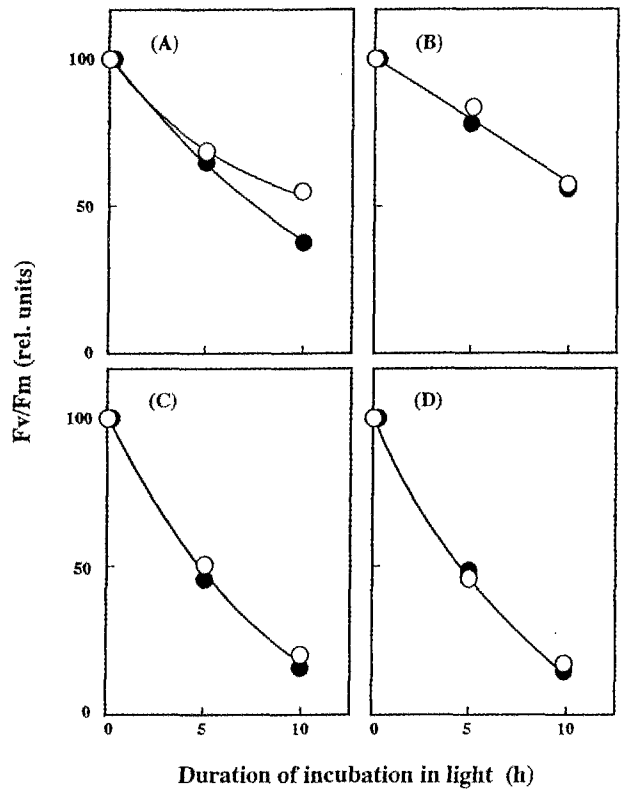

Figure 1. Photoinhibition of PS II photochemical efficiency, monitored in terms of ratios of Fv to Fm, in leaves of wild-type and Rbcs-SQ transgenic tobacco plants in the presence or absence of lincomycin. Lincomycin was administered by immersing petioles in 0.6mM lincomycin for 4h at 25℃ at a light intensity of 0.1 mmol quanta $m^{-2}s^{-1}$. To induce photoinhibition, leaf disks were incubated at a light intensity of 0.6 mmol quanta $m^{-2}s^{-1}$ at designated temperatures. The initial values of Fv/Fm measured at 25℃ were taken as 100% and corresponded to 0.79 and 0.80 for wild-type and transgenic tobacco, respectively. The values were obtained from the results of three independent experiments. The deviation of values was within ±5%. (A) 5℃, without lincomycin; (B) 15 ℃, without lincomycin; (C) 5 ℃, with lincomycin; (D), 15℃, with lincomycin; (○) Wild type; (●) Rbcs-SQ transgenic tobacco plants.

photoinhibition in leaves of wild-type tobacco plants and in the leaves of transgenic tobacco plants. When leaf disks were exposed to light at an intensity of 0.6 mmol $m^{-2}s^{-1}$, the extent of photoinhibition was higher at 5℃ than at 15℃ in both types of plant. Although no differences between the two types of plant were discernible in terms of the extent of photoinhibition during photoinhibition at 15℃, photoinhbition at 5℃ made the two type of plants show clear dicrepancy in the degree of decline in Fv/Fm.

Figure 1C and D show the time course of induction of photoinhibition at 5℃ and 15℃ in leaves of tobacco plants in the presence of lincomycin. Lincomycin accelerated the photoinhibition in the two types of tobacco plant. Moreover, the difference in the extent of photoinhibition between wild-type and transgenic plants that had been observed during exposure to 5℃, in the absence of lincomycin, disappeared in the presence of lincomycin (Figs. 1A and C).

Figure 2A shows the return to normal values of Fv/Fm during recovery of leaves of wild-type and transgenic tobacco plants from photoinhibition. Leaf disks were exposed to strong light at low temperature until Fv/Fm had fallen to about 20% of the original level. Then Fv/Fm ratios were monitored after shifting of the leaf disks to 25℃ under dim light. After 4h-incubation under these conditions, the ratio in wild-type plants reached about twice the ratio in transgenic plants.

The effects of light and chloroamphenicol, which are modulators of the repair that occurs in the phtosystem II complex during recovery from photoinhibition, were also studied in wild-type tobacco plants (Fig. 2B). For this purpose, photoinhibition in tobacco leaf disks was induced by strong light at 1℃ in the presence, or in the absence of chloramphenicol. Then the leaf disks were allowed to recover at 17 ℃ in darkness or under dim light. Increase in Fv/Fm were markedly promoted during incubation under dim light. In contrast to this, recovery in darkness or in the presence of chloramphenicol was very slow or negligible, suggesting that the recovery of the PS II complex from low-temperature photoinhibition requires photosynthetically-supported protein synthesis in chloroplasts.

Photoinhibition and Recovery in Chilling-Sensitive and Chilling-Resistant Plants

To compare the chilling sensitivity of plants having different adaptive capabilities to thermal environment, photo-induced inactivation of their PS II photochemical efficiency was examined according to the incubation temperatures in terms of chlorophyll fluorescence. Table 1 shows the extent of photoinhibition at 15℃ of the PS II complex in leaf disks of spinach and squash, as determined by Fv/Fm. The results indicates that strong light induced an marked inactivation of the PS II photochemical efficiency in squash plants, although spinach also exhibited a slight decrease, suggesting that the

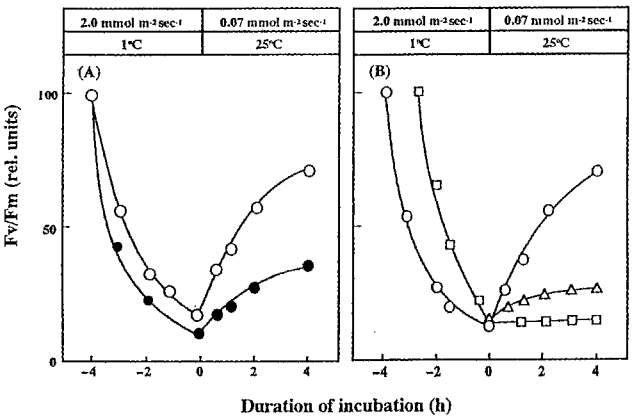

Figure 2. (A) Restoration of PS II photochemical efficiency, monitored in terms of ratios of Fv to Fm, after photoinhibition in leaves of wild-type and Rbcs-SQ transgenic tobacco plants. Leaf disks were exposed at 1℃ to light at an intensity of 1.5 mmol quanta $m^{-2}s^{-1}$ for 4h. To induce recovery from low-temperature photoinhibition, then the leaf disks were incubated at 25℃ in dim light at an intensity of 0.07 mmol quanta $m^{-2}s^{-1}$. (○) Wild type; (●) Rbcs-SQ transgenic tobacco plants. (B) Effects of light and chloramphenicol on restoration of PSII activity after photoinhibition in leaves of wild type tobacco plants. To induce photoinhibition at low temperature, leaves were exposed to strong light at an intensity of 1.5 mmol quanta $m^{-2}s^{-1}$ at 1℃ in the absence or presence of chloramphenicol (0.1mM). Then they were incubated for recovery from low-temperature photoinhibition at 25℃ in darkness or in light (0.07 mmol $m^{-2}s^{-1}$). (○) Light; (□) light plus chloramphenicol; (△) darkness. The values were obtained from the results of three independent experiments. The deviation of values was within ±5%.

PS II complex of squash plants was more susceptible to chilling temperature than spinach.

The extent of photoinhibition in leaves is determined by the counteracting two processes, namely the light-induced inactivation of the PS II complex and its repair from the photoinhibited form [6]. These two processes were seperated into the individual ones by inducing photoinhibition in the presence of lincomycin, as lincomycin serves as an inhibitor of protein synthesis in chloroplasts (Table 1). Although the extent of photoinhibition was accelerated by lincomycin in both types of plant, previously observed differences in susceptibility to photoinhibition during exposure of leaf disks to strong light was no more apparent in the presence of lincomycin between the two types of plant. This observation suggests that the rate of the photo-induced inactivation was about the same in the two species but that the rate of recovery from photoinhibition in spinach was faster than that in squash.

Figure 3 shows the time course of restoration of PS II photochemical efficiency, measured in terms of the ratios of Fv to Fm, in the chilling-resistant species, spinach and cabbage, and in the chilling-sensitive ones, sqaush and sweet potato. Leaf disks were first exposed to strong light at 1℃ for 3 or 4h to reduce the ratios to about 25% of the original values. Then, increases in each ratio were followed during incuba-

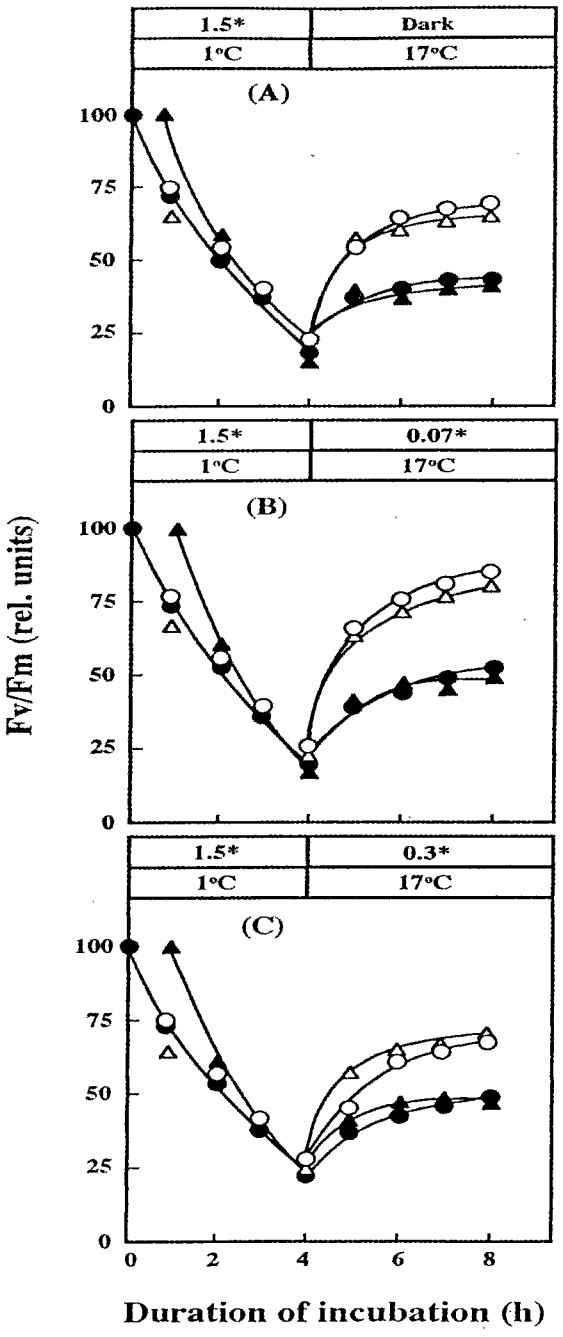

Figure 3. (A) Restoration of PS II photochemical efficiency, monitored in terms of ratios of Fv to Fm, after photoinhibition in leaves of spinach, cabbage, squash and sweet potato. To induce photoinhibition, leaf disks were exposed to light at 1°C at an intensity of 1.5 mmol quanta $m^{-2}s^{-1}$ for 3h with repect to squash and for 4h with respect to spinach. To allow recovery from low-temperature photoinhibition, then the leaf disks were incubated at 17°C in darkness (A), in light of at an intensity of 0.07 mmol quanta $m^{-2}s^{-1}$ (B), or in light at an intensity of 0.3 mmol quanta $m^{-2}s^{-1}$ (C). The initial values of Fv/Fm measured at 25°C were taken as 100% and corresponded to 0.80, 0.82, 0.81 and 0.79, in spinach, cabbage, squash and sweet potato, respectively. The values were obtained from the results of three independent experiments. The deviation of values was within ±5%. (○) Spinach; (△) cabbage; (●) squash; (▲) sweet potato. * light intensity in mmol quanta $m^{-2}s^{-1}$.

tion of the photoinhibited leaves at 17°C in darkness or in the light. During incubation under dim light following photoinhibition, there was a clear difference in the extent of recovery of the PS II complex between the chilling-sensitive and chilling-resistant species. After recovery for 4h under irradiation by light of 0.07 mmol $m^{-2}s^{-1}$, ratios of Fv to Fm of cabbage and spinach reached 70% of the original levels, whereas those of squash and sweet potato had reached only 40%.

DISCUSSION

In the present study, we examined a possibility that photoinhibition of PS II complex, as monitored in terms of chlorophyll fluorescence would be affected by alteration in the degree of unsaturation of membrane lipis. The purpose of the test was achieved by using wild-type tobacco plants and transgenic tobacco plants that had been transformed with cDNA for glycerol-3-phosphate acyltransferase from squash [8]. The transgenic tobacco plants were found to contain reduced levels of unsaturated fatty acids in membrane lipids, thus having higher sensitivity of photosynthesis to chilling [10].

The presented results showed that photochemical efficiency of PS II of the transgenic plants was more sensitive to low-temperature photoinhibition than the wild-type plants. Furthermore, when the extent of low-temperature photoinhibition was compared between chilling-sensitive squash and chilling-resistant spinach (Table 1), squash was found to be more sensitive to photoinhibition than spinach, indicating that there are clear discrepancy in the photochemical efficiency of the PS II complex between those plants having different chilling sensitivity.

During photoinhibition *in vivo*, two processes are suggested to counteract against each other [6]. One is the inactivation process induced by high light, which includes the photodamage

Table 1. Photoinhibition of the PS II complex at 15°C, monitored in terms of ratios of Fv to Fm, in leaves of spinach and squash in the presence or absence of lincomycin

Time of photoinhibition at 15°C (h)	Fv/Fm (% of control)			
	minus Lm		plus Lm	
	spinach	squash	spinach	squash
0	100	100	100	100
5	92	73	68	57
10	90	54	43	32

Experimental details were the same as described in the legend of Figure 1. The initial values of Fv/Fm measured at 25°C were taken as 100% and corresponded to 0.81 and 0.80 for squash and spinach, respectively. The values were obtained from the results of three independent experiments. The deviation of values was within ±5%. Lm, lincomycin.

to the D_1 protein of the PS II complex, and the other is the recovery process, which includes degradation of the photodamaged D_1 protein, synthesis of the D_1 protein *de novo* and reintegration of the D_1 protein into the PS II complex [4]. From these perspectives, we seperated the inactivation process from the recovery process by applying lincomycin to leaves through transpiration stream, and subsequently exposed them to high light. Our results showed that by lincomycin treatment, the decline in Fv/Fm by exposure of leaf disks to high light, especially at low temperature, was accelerated even more than before lincomycin treatment. More importantly it was noted that the previoulsly observed discrepancy in the extent of Fv/Fm drop between spinach and squash, during exposure of them to high light in the absence of lincomycin, as well as between wild-type and transgenic tobacco plants, became almost abolished when they were photoinhibited in the presence of lincomycin. Since lincomycin serves as an inhibitor of repair reactions of photodamaged PS II complex due to low-temperature photoinhibition, it is assumed that photoinhibition in the presence of lincomycin consists only of the photo-induced inactivation in the PS II complex. Therefore, our observations suggest that the light-induced inactivation of the PS II complex occurs at almost equivalent rate in spinach and squash plants, as well as in wild-type and transgenic tobacco plants.

We tried to examine whether the recovery of the PS II complex from low-temperature photoinhibition was responsible for the observed differences in chilling sensitivity between the wild-type and transgenic tobacco plants, and also between chilling-resistant species and chilling-sensitive ones. For this purpose, we exposed leaf disks of each plant to strong light at 1°C to reduce Fv/Fm values. Then we directly measured restoration of Fv/Fm from leaf disks that had been incubated in darkness or under dim light in order to induce recovery of the PS II complex from the photoinhibited state.

Our results showed that there were clear differences in the rate of recovery from photoinhibition between the two types of plants; chilling resistant pea and spinach as well as wild-type tobacco plants showed much faster restoration of Fv/Fm than the chilling-sensitive groups such as sweet potato, squash and Rbcs-SQ transgenic plants. Furthermore, when we analyzed the process of recovery under various light conditions, the recovery under low-intensity light (0.07 mmol $m^{-2}s^{-1}$) was shown to be faster than in darkness, suggesting that photosynthetically produced energy is important in the recovery process. Chloramphenicol completely blocked the recovery, a result suggesing that the recovery process was supported by active protein synthesis.

Tolerance of plants to chilling is closely related to the level of unsaturated species of phosphatidylglycerol (PG) [15,16,17]. When the relative levels of molecular species of PG in cabbage, spinach, squash and sweet potato were calculated from our earlier results (Table 2), both chilling-sensitive species and transgenic tobacco plants contained low amounts of cis-unsaturated PG in membrane lipids, suggesting that low levels of fatty acid unsaturation of membrane PG is related to slower recovery of PS II complex from low-temperature photoinhibition. In contrast to this, both chilling-resistant species and wild-type tobacco plants contained higher amounts of cis-unsaturated PG, suggesting that higher degree of fatty acid unsaturation of membrane PG accelerates the recovery from low-temperature photoinhibition. It is widely accepted the repair of photodamaged PS II centers constitute a complex cycle, including degradation of photodamaged D_1 protein, *de novo* synthesis, and insertion of newly synthesized D_1 protein into PS II complex, migration of PS II complexes between appressed and nonappressed thylakoid regions [18].

Adir *et al* (1990) [19] have suggested that the rate of the repair cycle of PS II is controlled by the migration of photodamaged PS II reaction centers from the appressed to nonappressed membrane regions for repair. It is possible that low-temperature photoinhibition affects fluidity of thylakoid membranes, which in turn could possibly impede migration of the damaged molecules through altered lipid environments of thylakoid membranes. On the basis of these possibilities, we speculate that unsaturation of membrane lipids is responsible for maintaining membrane fluidity of thylakoid membranes at low temperature, which might be crucial in the turnover of photodamaged D_1 protein.

Table 2. Estimated contents of cis-unsaturated PG molecules relative to the total molecular species of PG isolated from leaves of some plants. S and R stand for chilling-sensitive and chilling-resistant plants, respectively.

Plant	S or R	*Cis*-unsaturated PG species (mol %)
Cabbage	R	96
Spinach	R	94
Squash	S	57
Sweet potato	S	29
Tobacco:		
Wild type	-	64
Rbcs-SQ[a]	-	24

The values were calculated from previously published data [8,15].
[a]Rbcs-SQ plants stand for transgenic tobacco plants transformed by cDNAs for glycerol-3-phosphate acyltransferse from squash.

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REFERENCES

1. Öquist, G. and N.P.A. Huner (1993) Cold-hardening-induced resistance to photoinhibition of photosynthesis in winter rye is dependent upon an increased capacity for photosynthe

sis. *Planta* **189**, 150-156.

2. Ottander C., T. Hundal, B. Andersson, N.P.A. Huner and G. Öquist (1993) Photosystem II reaction centers stay intact during low temperature photoinhibition. *Photosynth. Res.* **35**, 191-200.
3. Andersson, B., A.H. Salter, I. Virgin, I. Vass and S. Styring (1992) Photodamge to Photosystem II - primary and secondary events. *J. Photochem. Photobiol.* **15**, 15-31.
4. Aro, E.-M., I. Virgin and B. Andersson (1993) Photoinhibition of photosystem II. Inactivation, protein damage and turnover. *Biochim. Biophys. Acta* **1143**, 113-134.
5. Kanervo, E., E.-M. Aro and N. Murata (1995) Low unsaturation level of thylakoid membrane lipids limits turnover of the D1 protein of Photosystem II at hight irradiance. *FEBS Lett.* **364**, 239-242.
6. Greer, D.H., J.A. Berry and O. Björkman (1986) Photoinhibition of photosynthesis in intact bean leaves: role of light and temperature, and requirement for chloroplastprotein synthesis during recovery. *Planta* **168**, 253-260.
7. Miyao, M., M. Ikeuch N. Yamamoto and T.-A. Ono (1995) Specific degradation of the D1 protein of photosystem II by treatment with hydrogen peroxide in darkness: implications for the mechanism of the degradation of D1 protein under illumination. *Biochemistry* **34**, 10019-10026.
8. Murata, N., O. Ishizaki-Nishizawa, S.-I. Higashi, H. Hayashi, Y. Tasaka and I. Nishida (1992) Genetically engineered alteration in the chilling sensitivity of plants. Nature 356, 710-713.
9. Gombos, Z., H. Wada and N. Murata (1994) The recovery of photosynthesis from low-temperature photoinhibition is accelerated by the unsaturation of membrane lipids: a mech anism of chilling tolerance. *Proc. Natl. Acad. Sci. USA* **91**, 8787-8791.
10. Moon, B.Y., S.-I. Higashi, Z. Gombos and N. Murata (1995) Unsaturation of the membrane lipids of chloroplasts stabilizes the photosynthetic machinery against low-temperature photoinhibition in transgenic tobacco plants. *Proc. Natl. Acad. Sci. USA* **92**, 6219-6223.
11. Schreiber, U., W. Schliwa and U. Bilger (1986) Continuous recording of photochemical and non-photochemical chlorophyll fluorescence quenching with a new type of modulaiton fluorometer. *Photosynth. Res.* **10**, 51-62.
12. Moon, B.Y., C.B. Lee, Y.-G. Gong and I.-S. Kang (1998) Low dissipation of excitation energy in the photosynthetic machinery of chilling-sensitive plants during low-temperature photoinhibition. *J. Photosci.* **5**, 53-61.
13. Butler, W.L. (1978) Energy distribution in the photochemical apparatus of photosynthesis. *Annu. Rev. Plant Physiol.* 29, 345-378.
14. Krause, G.H. and E. Weis (1991) Chlorophyll fluorescence and photosynthesis: the basics. *Annu. Rev. Plant Physiol. Plant Mol. Biol.* **42**, 313-349.
15. Murata, N. (1983) Molecular species composition of phosphatidylglycerol from chilling-sensitive and chilling-resistant plants. *Plant Cell Physiol.* **24**, 81-86.
16. Roughan, P.G. (1985) Phosphatidylglycerol and chilling sensitivity in plants. *Plant Physiol.* **77**, 740-746.
17. Nishida, I. and N. Murata (1996) Chilling sensitivity in plants and cyanobacteria: The crucial contribution of membrane lipids. *Annu. Rev. Plant Physiol. Plant Mol. Biol.* **47**, 541-568.
18. Aro, E.-M., S. McCaffery and J.M. Anderson (1993) Photoinhibition and D1 protein degradation in peas acclimated to different growth irradiances. *Plant Physiol.* **103**, 835-843.
19. Adir, N., S. Shochat and I. Ohad (1990) Light-dependent D1 protein synthesis and translocation is regulated by reaction center II. *J. Biol. Chem.* **265**, 12563-12568.