ACCLIMATION OF LIGHT-HARVESTING AND LIGHT-UTILIZATION CAPACITIES IN RESPONSE TO GROWTH TEMPERATURE

KENNETH L. STEFFEN AND JIWAN P. PALTA, DEPARTMENT OF HORTICULTURE, UNIVERSITY OF WISCONSIN, MADISON, WI 53706, USA.

1. INTRODUCTION

There is considerable variation in the ability of tuber-bearing *Solanum* species to increase their frost tolerance during a two week exposure to low, non-freezing temperature (1). We have previously shown that there is a light-dependent inhibition of photosynthetic capacity during this acclimation period (2). The degree of inhibition was significantly greater in a species which can not acclimate than in a species which is able to acclimate. We have suggested that an important component of the ability to frost acclimate is the ability to maintain high levels of photosynthetic metabolism during the acclimation period. While there are a number of protective compounds and mechanisms present within the chloroplast which are known to provide protection from the damaging effects of excess light energy (3), it is reasonable to expect that a plant's first and most efficient response would be an adaptation which would balance its light-trapping and light-utilization capacities to the prevailing environmental conditions.

The objectives of the present study were: 1) to determine if a moderate level of low temperature stress results in an adapted state which has increased tolerance to short term photoinhibitory stresses; 2) to test the hypothesis that acclimation involves an adjustment of light-trapping and light-utilization capacities in order to prevent over-excitation of the photosynthetic apparatus; and 3) to determine if alterations in levels of total soluble and membrane-bound proteins are of sufficient magnitude to explain changes in photosynthetic parameters in response to growth temperature.

2. PROCEDURE

2.1. Materials and methods

2.1.1. Plant material: Two potato species, *S. commersonii*, which is cold-tolerant and can frost acclimate, and *S. tuberosum*, which is cold-sensitive and can not frost acclimate, were used in this study. Plants were established at 18 C with 70-80% RH and a 12 h photoperiod of 400 µmol/m^2-s light quanta at the canopy level. After 2 weeks, plants were transferred to either 12 or 24 C with all other conditions held constant. Following 4 weeks of growth temperature treatments, leaflets that were 80-90% of full expansion and at an incident light level of 400 ± 50 µmol/m^2-s were removed and leaf discs excised for immediate use in treatments or assays.

2.1.2. Photoinhibitory stress treatments: Leaf discs were floated on a buffer at 1.0 C (50 mM Hepes-KOH, 0.5 mM $CaSO_4$, pH 7.2) in individual wells of a specially designed chilling chamber. Paired leaf discs from a single leaflet were held in either the dark or in the presence of 1000 µmol/m^2-s white light, for 4 hours. Following the stress treatment, all leaflets were held in the dark at 1.0 C until photosynthetic assays could be conducted.

Biggens, J. (ed.), Progress in Photosynthesis Research. Vol. IV. ISBN 90 247 3453 3
© 1987 Martinus Nijhoff Publishers, Dordrecht. Printed in the Netherlands.

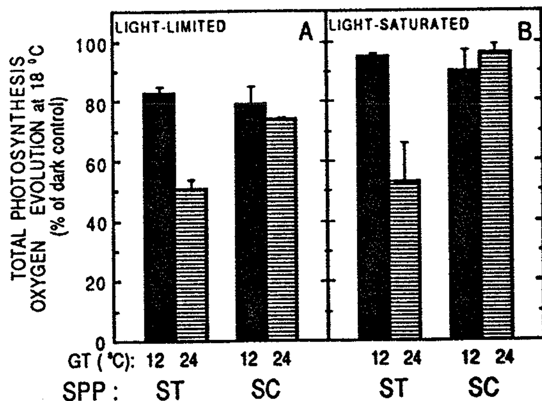
2.1.3. Photosynthetic and respiratory assays: Leaf discs were sliced, infiltrated with assay buffer (50mM Hepes-KOH, 0.5 mM $CaSO_4$, pH 7.2) and then placed in the chamber of an oxygen electrode at 18 with a final bicarbonate concentration of 20 mM. Respiratory oxyg uptake was first measured and then light-limited photosynthesis w initiated by supplying 114 µmol/m^2-s light quanta, followed by t measurement of light-saturated photosynthesis at 1750 µmol/m^2-s light quanta (see ref. 2 for details). Apparent photosynthetic quantum yields were determined by measuring photosynthesis at 41, 72 and 114 µmol/m^2-s light quanta. Following the assays, chlorophyll was extracted from the tissue and quantitated in 96% ethanol. Respiration rates were added to all net photosynthetic rates to obtain total photosynthesis per chlorophyll. Dark-chille controls had photosynthetic rates similiar to unstressed tissue.

2.1.4. Protein determinations: Approximately 0.5 g of leaf disc tissue was thoroughly homogenized in ice cold buffer (50 mM Tris-HCl, 2 mM DTT, pH 7.8) using a Brinkman polytron. The slurr was spun at 20,000 x g for 30 min, the supernatant poured off and saved. The pellet was resuspended with sonication in 2.5 ml of grinding buffer and spun again at 20,000 x g for 30 min. The supernatants were pooled and spun at 109,000 x g for one hour. A pellets were combined and resuspended in 5.0 ml grinding buffer using sonication. Aliquots of the supernatant and pellet fractio were incubated in 1 N NaOH for 2 hours. A modified Lowry (4) was used to determine total soluble and membrane-bound protein conten

3. RESULTS AND DISCUSSION

3.1. The combination of high light and low temperature, caused a greater reduction in light-limited versus light-saturated photosynthetic capacity, of plants grown at both temperatures (Fig. 1). Both the light-limited and the light-saturated photosynthetic capacity of tissue from 24 C-grown *S. tuberosum* was reduced more than 50% by hig light and low temperature treatment, compared to dark-chilled tissue The 12 C-grown *S. tuberosum,* however, showed an adaptive response which resulted in little photoinhibitory damage by the same treatmen (Fig. 1). Visual observations suggested that *S. tuberosum* was under slight stress at 12 C, as evidenced by marginal yellowing and occa- sional necrotic flecking. *S. commersonii* appeared more resistant to the photoinhibitory treatment, regardless of the growth temperature. Furthermore, this species showed no visual signs of stress at 12 C.

3.2. The photosynthetic capacities of both species showed a consistent response to decreased growth temperature. Light-saturated photo- synthetic capacity, assayed at 18 C, increased markedly with de- creasing growth temperature. This increase was 32% in *S. tuberosum* a 50% in *S. commersonii* for 12 C compared to 24 C-grown plants (Fig. 2 This suggests an enhancement of light-utilization capacity in spite reduced enzyme reaction rates at the lower growth temperature. At t same time there was, in both species, a 20-25% decline in the appare quantum efficiency at the lower growth temperature (Fig.3). These results indicate a reduction in light-trapping and/or electron trans- port capacities in response to lowered temperature, again suggesting an adaptive response to prevent or minimize over-excitation of the thylakoid membranes under stress conditions.



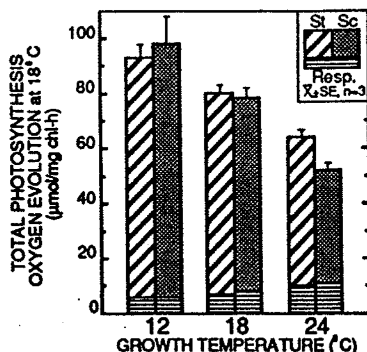
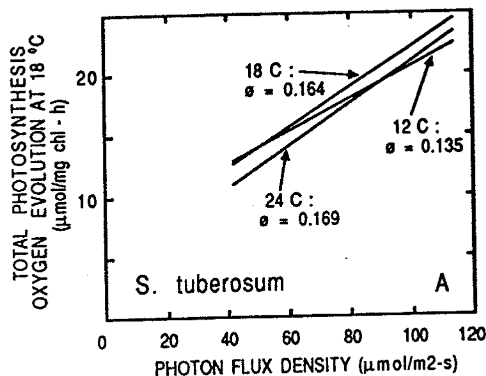


FIGURE 1. Total light-limited (A) and light-saturated (B) photosynthetic oxygen evolution at 18 C following 4 h of high light and low temperature stress. Plant material: *S. tuberosum* (ST) and *S. commersonii* (SC) grown at 12 or 24 C. Rates are expressed as a percentage of the paired dark controls. Values are means ± SE, n = 3.

FIGURE 2. Total light-saturated photosynthetic oxygen evolution from *S. tuberosum* and *S. commersonii* plants grown at 12, 18 or 24 C and assayed at a common temperature of 18 C. Values are means ± SE, n = 3.



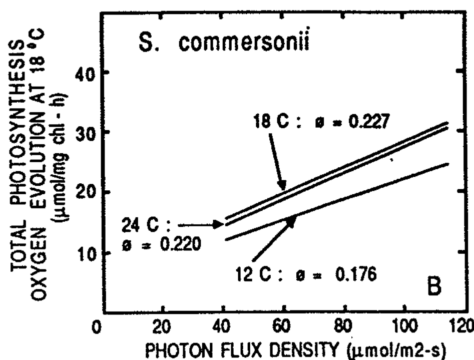


FIGURE 3. Apparent total photosynthetic quantum yields assayed at 18 C in the presence of 41, 72 and 114 µmol/m^2-s light quanta. Regression lines were fitted to the means of 4 replications of each temperature treatment at each light assay level and r^2 = 0.999 or better. a) *S. tuberosum* and b) *S. commersonii*.

3.3. Soluble protein per chlorophyll increased under 12 C-growth conditions compared to 24 C, by 29% in *S. tuberosum* and 40% in *S. commersonii* (Tab. 1). Since soluble leaf protein is a reasonable estimate of levels of enzyme involved in carbon fixation, the data indicates that changes in protein/chl are of the magnitude necessary to explain the increase in light-saturated photosynthetic capacity

TABLE 1. Changes in chlorophyll and protein content with growth temperature.

Species	Growth Temp. (C)	Chl content per FW (mg/g)	Soluble protein per FW (mg/g)	Soluble protein per Chl (ratio)	Membrane protei per FW (mg/g)	Membrane protei per C (rati
S. tuberosum	24	2.1±0.06[a]	16±1.0	7.6	31±2.0	14.
	12	1.7±0.17	18±2.3	10.6	24±1.4	14.
S. commersonii	24	2.4±0.09	28±0.9	11.7	25±1.8	10.
	12	1.7±0.14	32±1.5	18.8	22±0.7	12.

[a]Means ± SE, n = 3.

associated with lowered growth temperature (Fig. 2). While both species show a similiar decline (20-30%) in apparent photosynthetic quantum yield at the lower growth temperature (Fig. 3), the changes chlorophyll per membrane protein (1/membrane protein per Chl; rati are quite different (Tab. 1). There is essentially no change in thi ratio in *S. tuberosum* while there is a 20% decline in chlorophyll p membrane-bound protein in *S. commersonii*. This data taken together with the proportionally greater loss of chlorophyll than membrane protein, suggests that in *S. commersonii* there may be a reduction in antenna size. In *S. tuberosum*, however, the substantially greater loss of protein, balanced by the loss of chlorophyll, suggests a reduction in the number of light-harvesting units.

3.4. In conclusion, our results show: 1) that growth under a mild, low temperature, stress condition, results in an acclimated state in whi tissue tolerance to photoinhibitory treatments is dramatically increased 2) this low temperature, acclimated state has an increased enzymatic capacity for light energy-utilization and a decreased light-trapping efficiency, and 3) increases in total soluble protein per chlorophyll are in the range to explain most of the increases in photosynthetic capacity at the lower growth temperature. The data suggests that the adjustment of light-harvesting and light-utilizati capacities in response to growth temperature plays a role in preventing photoinhibition. However, the relative importance of oth protective mechanisms needs to be considered in order to determine t biological relevance of this adaptation.

REFERENCES

1 Palta, J.P. and P.H. Li. 1979. Crop Sci. 19: 665-671.

2 Steffen, K.L. and J.P. Palta. 1986. Physiol. Plant. 66: 353-359.

3 Powles, S.B. 1984. Annu. Rev. Plant Physiol. 35: 15-44.

4 Markwell, M.A.K., S.M. Haas, L.L. Bieber and N.E. Tolbert. 1978. Anal. Biochem. 87: 206-210.