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## Photosynthetic responses of greenhouse tomato plants to high solution electrical conductivity and low soil water content

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### SUMMARY

Greenhouse tomato plants (*Lycopersicon esculentum* Mill. cv. Capello) were grown in a peat-moss based substrate and supplied with nutrient solutions of high ( $4.5 \text{ mS cm}^{-1}$ ) or low ( $2.3 \text{ mS cm}^{-1}$ ) electrical conductivity (EC) and under high ( $95 \pm 5\%$ ) or low ( $55 \pm 8\%$  of capillary capacity) soil water content, to elucidate how EC and soil water status affect plant photosynthesis and related physiological processes. Two weeks after beginning the treatments, photosynthesis (Pn) was measured during changes of photosynthetic photon flux (PPF) from 0 to  $1200 \mu\text{mol m}^{-2} \text{s}^{-1}$  using a gas exchange method. The rectangular hyperbolic model ( $P_n = P_{\text{max}} \text{KI} (1 - \text{KI})^{-1} - r$ ) provided a good fit for the photosynthetic light-response curve. High EC treatment changed the curve by increasing the initial slope (quantum yield) and decreasing photosynthetic capacity at high PPF. However, soil water deficit not only decreased the photosynthetic capacity, but also decreased quantum use efficiency. Depression of Pn was attributed to decreased stomatal ( $g_s$ ) and mesophyll ( $g_m$ ) conductances, but  $g_s$  was depressed more than  $g_m$ . The ratio of  $g_m/(g_m + g_s)$ , an indicator of water use efficiency and a measure of relative control of Pn by carboxylation and  $\text{CO}_2$  supply, was higher for high-EC treated plants. Chlorophyll content was increased by high EC treatment, and was consistent with quantum yield. Leaf water potential was decreased by high EC and/or low soil water content and the decreases in leaf water potential ultimately accounted for the Pn depressions. The effects of high EC and soil water deficit were additive on photosynthesis and most related physiological processes.

SOIL water deficit affects plant growth and physiology through reduction in leaf water potential. Decrease in leaf water potential affects many physiological processes. Photosynthesis is sensitive to water stress. Although severe water stress does not occur frequently in greenhouse crop production, short-term mild water stress can occur. One factor limiting plant water uptake is high electrical conductivity (EC) of the nutrient solution. High EC affects plant physiology through decreasing plant water uptake (Ehret and Ho, 1986). Even under sufficient water supply, substrate salinity can induce water stress, especially when evapotranspiration demand is high (Adams, 1991; Awang *et al.*, 1993). Salt-buffering substrates

are often used to avoid salinity stress. Peat-moss based substrates are popularly used in greenhouse tomato production because of their high salt-buffering capacity, good aeration and high capillary water capacity (Derouin *et al.*, 1988). To improve fertigation of greenhouse tomato grown in peat-moss based substrates, it is important to determine the effects of EC and substrate water content on plant growth and physiology. The mechanism of water deficit on farm crops is well documented (Radin, 1993). However, there are few reports on the effect of EC on photosynthesis in greenhouse tomatoes. Therefore, the first part of the present research was designed to examine the effects of high EC and soil water deficit on photosynthesis and

related processes such as CO<sub>2</sub> diffusion conductances and chlorophyll content as well as the leaf water potential of greenhouse tomato plants grown in a peat-moss based substrate.

To analyze the photosynthetic response characteristics, the rectangular hyperbolic model has been used to fit the light-response curve (Jones, 1983; Givnish, 1988). From the light-response curve, it is possible to identify which physiological process of photosynthesis is affected. When the dark reaction and CO<sub>2</sub> supply are not limiting, for example under low PPF levels, the light reaction determines the photosynthetic rate (Harbison *et al.*, 1989). Hence, the activity of light reaction is obtained from the initial slope of the light-response curve, which is also called the quantum yield. Under high PPF levels, the limiting factors are CO<sub>2</sub> supply and carboxylation. CO<sub>2</sub> supply is determined by the rate of CO<sub>2</sub> diffusion from ambient air through stomata to photosynthetic mesophyll. Stomatal conductance is dominant in this process. Carboxylation is determined by Rubisco activity and reflected in mesophyll conductance, i.e. CO<sub>2</sub> diffusion rate from mesophyll cell wall to the carboxylation site. Therefore, in the present work, effects not only on photosynthesis but also on the related processes are measured and discussed.

#### MATERIALS AND METHODS

##### *Plant material*

Seeds of tomato (*Lycopersicon esculentum* Mill., cv. Capello) were sown in small rockwool cubes (Pargro Co., Ltd., Caledonia, Ontario, Canada) in seedling packs. When the seedlings were established, they were transplanted to rockwool blocks (Pargro, 0.1 m × 0.1 m × 0.1 m). Five weeks after sowing, the seedlings were transplanted into plastic pots (0.00625 m<sup>3</sup> in volume) filled with peat-moss based substrate (70% peat and 30% perlite (v/v), Premier Peat Moss, Rivière-du-loup, Quebec, Canada). The plants were grown in a double-layer polyethylene greenhouse. The temperature was controlled at 21 ± 1°C/18 ± 1°C (day/night). The relative humidity fluctuated with time of day between 50% and 80%. When the fifth leaf from the base was fully expanded, the plants were divided into four plots and supplied with nutrient solutions of high (4.5 mS cm<sup>-1</sup>) and low (2.3 mS cm<sup>-1</sup>) EC combined with high

(95 ± 5%) and low (55 ± 8% of the capillary capacity) substrate water content. Each treatment had five replicate plots and each plot included six plants, at a density of three plants per square metre. Concentrated nutrient solutions were made and stored in tanks. The irrigation nutrient solutions were made by diluting the concentrated solutions into water in appropriate ratios, and EC was adjusted by decreasing or increasing the total nutrient concentration. The concentrations of nutrient elements are presented in Table I. The levels of all nutrients in the low EC treatment were normal and non-limiting for tomato plant growth and physiology. The soil water levels were adjusted by weighing and watering the pots twice a day. Each pot was placed in a plastic dish. The salts were allowed to accumulate in the substrate without overwatering. Nutrient analysis of irrigation solution or substrate was conducted by atomic absorption spectrophotometry at the laboratory of Quebec Ministry of Agriculture, Fishery and Food. The salt or nutrient levels on day 1 and day 14 are presented in Table II.

##### *Measurement of photosynthesis*

Two weeks after beginning the treatments, photosynthesis (P<sub>n</sub>) was measured using an infrared CO<sub>2</sub> analyzer (ADC-225-MK3, Analytical Development Co., Ltd., Hoddesdon, England) in an open gas exchange system (Yue *et al.*, 1992). The top leaflet of the youngest fully expanded leaf was placed in an assimilation chamber. Photosynthetic rates of the same leaflet were measured at different PPF at 0, 50, 100, 250, 400, 600, 800, and 1200 μmol m<sup>-2</sup> s<sup>-1</sup>. The measurement at one level of PPF took 20 min. Two assimilation chambers were used at the same time for two leaf samples, each from different treatments. To prevent large changes in soil water status by transpiration, other leaves were shaded from strong light. After the photosynthetic measurement, leaf area was measured with a leaf area meter (LI-COR Model 3000). Air temperature in the assimilation chambers was 23 ± 0.4°C depending on PPF. Leaf temperature was about 0.5°C lower than air temperature at low PPF (0 to 250 μmol m<sup>-2</sup> s<sup>-1</sup>) and 0.5 to 1.0°C higher than air temperature at high PPF. The vapour pressure deficit (vpd) of the air from the assimilation chamber inlet was

TABLE I  
Composition ( $\text{mol m}^{-3}$ ) of nutrient solutions used in this experiment

Element	Electrical conductivity (EC)	
	$2.3 \text{ mS cm}^{-1}$	$4.5 \text{ mS cm}^{-1}$
N	10.1	19.7
P	1.7	3.3
K	7.2	13.8
Ca	3.5	6.8
Mg	1.8	3.5
Fe	0.0726	0.1421
Mn	0.0180	0.0354
Zn	0.0076	0.0149
Cu	0.0016	0.0030
B	0.0282	0.0510
Mo	0.0006	0.0011

controlled at 1.3 KPa. The air inside the assimilation chambers was circulated by a fan mounted on a side wall of the assimilation chamber and the vpd varied with transpiration rate between 1.2 and 0.9 KPa.

#### Determination of stomatal and mesophyll conductances

Stomatal conductance for  $\text{CO}_2$  diffusion was calculated on the basis of water vapour diffusion conductance as described by Jones (1983). Stomatal conductance for water vapour diffusion was calculated from transpiration rate and the water vapour content difference between air in the assimilation chamber and that in the intracellular space under the leaf temperature. Transpiration was measured synchronously with photosynthesis by an infrared  $\text{H}_2\text{O}$  analyzer (ADC-225-MK3, Analytical Development Co., Ltd., Hoddesdon, England) in the same open gas exchange system. Leaf temperature was measured using a copper-constantan thermocouple (type T) touching the undersurface of the leaf blade. Air temperature in the

assimilation chamber was measured with a similar copper-constantan thermocouple placed underneath the leaf blade. Mesophyll conductance was calculated from photosynthetic rate and intracellular  $\text{CO}_2$  concentration as described by Jones (1983).

#### Modelling the photosynthetic light-response curve

The following rectangular hyperbolic model was used to fit the photosynthetic light-response curve (Givnish, 1988):

$$P_n = P_{\max} KI (1 + KI)^{-1} - r,$$

where  $P_n$  ( $\mu\text{mol m}^{-2} \text{ s}^{-1}$ ) is the net photosynthetic rate at a given PPF ( $I$ ,  $\mu\text{mol m}^{-2} \text{ s}^{-1}$ );  $P_{\max}$  ( $\mu\text{mol m}^{-2} \text{ s}^{-1}$ ), the saturation photosynthetic rate at maximum  $I$ ;  $K$  ( $\mu\text{mol}^{-1} \text{ m}^2 \text{ s}$ ), the Michaelis-Menten constant, equals the reciprocal of the PPF level at which  $P_n = P_{\max}/2$ ;  $r$  ( $\mu\text{mol m}^{-2} \text{ s}^{-1}$ ), dark respiration rate. The value of  $P_{\max} K$  ( $\text{mol CO}_2 \text{ mol}^{-1} \text{ PPF}$ ) shows the maximum quantum use efficiency when PPF ( $I$ ) tends to 0 (Figure 1).

#### Determination of quantum yield

General quantum requirement was expressed by plotting the PPF/ $P_n$  ratio against PPF. The intersection of the regression line with the vertical axis was defined as the minimum quantum requirement (quantum mole required for assimilation of one mole  $\text{CO}_2$ ) and its reciprocal was defined as quantum yield. Quantum yield can also be seen from the slope of the initial linear part of the light-response curve (Figure 1).

#### Water use efficiency and chlorophyll content

Since  $g_s$  is positively related to transpiration

TABLE II  
The concentrations of potassium (K), magnesium (Mg) calcium (Ca), nitrate nitrogen ( $\text{NO}_3^-$ ) and phosphorus (P) in the substrate and electrical conductivity (EC) in the soil solution on day 1 and day 14 from beginning of the treatments

Treatment	$\text{mol m}^{-3}$										$\text{mS cm}^{-1}$	
	K		Mg		Ca		$\text{NO}_3^-$		P		EC	
	1 d	14 d	1 d	14 d	1 d	14 d	1 d	14 d	1 d	14 d	1 d	14 d
HW/LEC	2.2	7.8	0.33	2.8	0.72	5.2	0.35	2.0	0.42	1.8	1.0	4.1
LW/LEC	1.8	6.4	0.25	1.7	0.50	3.0	0.32	1.5	0.36	1.5	1.3	4.7
HW/HEC	2.4	15.0	0.45	4.8	0.80	9.5	0.40	5.4	0.48	3.5	1.2	8.0
LW/HEC	2.1	12.8	0.37	3.4	0.62	6.5	0.33	3.2	0.36	2.3	1.5	9.3

HW/LEC, high soil water content combined with low EC; LW/LEC, low soil water content combined with low EC; HW/HEC, high soil water content combined with high EC; LW/HEC, low soil water content combined with high EC. The same abbreviations for the treatments are used in the following tables.

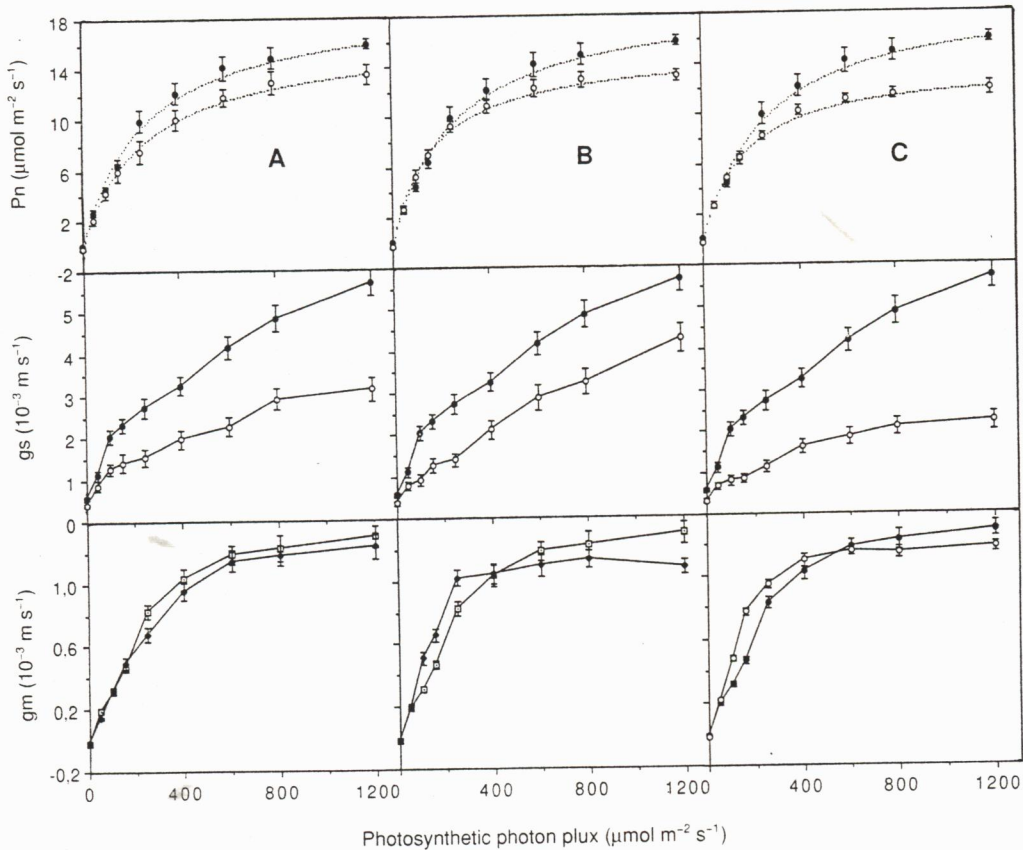


FIG. 1

Responses of photosynthetic rates ( $P_n$ ) and stomatal ( $g_s$ ) and mesophyll ( $g_m$ ) conductances in tomato plants to changing photosynthetic photon flux. A, low EC and low soil water content; B, high EC and high soil water content; C, high EC and low soil water content. The closed circles show the results of control (low EC and high soil water content). Data at each PPF level are the mean values of three measurements with a standard error shown by the vertical bars.

and  $g_m + g_s$  is positively related to photosynthesis, the  $g_m/(g_m + g_s)$  ratio shows water use efficiency if there is no large fluctuation in intracellular  $CO_2$  concentration (Jones, 1980). In the present work, the  $g_m/(g_m + g_s)$  ratio was plotted against PPF to elucidate the effects of high EC and substrate water deficit at different PPF levels. Chlorophyll content was measured by the method of Arnon as described by Hokujo and Ishitsuka (1988) and Xu (1994).

#### Measurement of leaf water potential

Xylem water potential in the leaf was measured at predawn and midday with a pressure chamber (Model 3000, Soilmoisture Equipment Corp., Santa Barbara, CA 93105, USA) to show the leaf water potential (Turner, 1988). The youngest expanded leaf was cut with

five leaflets attached, immediately put into a polyvinyl bag and then placed in the pressure chamber. Air humidity in the pressure chamber was maintained at saturation, to prevent transpirational water loss. The speed of pressure application was fast at the beginning of the measurement and then slowed to  $0.01 \text{ MPa s}^{-1}$ , when the pressure was close to the level of leaf water potential. When water exuded from the xylem and saturated the cut end of the petiole, pressure application was stopped. The balance pressure was then assumed to be the leaf water potential. To see the effect of PPF level on leaf water status during photosynthetic measurement, leaf water potential was also measured after placing the leaf in the assimilation chamber for 30 min to adapt different PPF and chamber conditions.

## RESULTS AND DISCUSSION

*Photosynthetic capacity, quantum yield and respiration*

*Effect of high EC:* Under both high and low soil water conditions, high EC treatment changed the light-response curve by increasing initial slope and decreasing photosynthetic rate at high PPF (Figure 1B, 1C). At low PPF, Pn was slightly higher for high-EC than for low-EC treated plants. However, as PPF increased, Pn became lower in high-EC treated plants than in low-EC treated plants (Figure 1B, 1C). This phenomenon was explained by the analysis of the light-response curve with a rectangular hyperbolic model (Figure 1, Table III). High EC treatment decreased photosynthetic capacity (photosynthetic rate at saturation PPF), as shown by both  $P_{max}$  and  $P_{1200}$  under high and low substrate water conditions (Table III). Another important parameter in this model is the Michaelis-Menten constant, K. The larger K is, the earlier Pn reaches half value of  $P_{max}$  and the more curved is the light photosynthesis regression. High EC treatment increased K and the maximum quantum use efficiency (MQUE) shown by  $P_{max}/K$  obtained from the model (Table III). In the present work, we also used another method to determine quantum use efficiency (Figure 2) and we called it quantum yield ( $Q_y$ ).  $Q_y$  obtained by this method was consistent with MQUE ( $P_{max}/K$ ) calculated from the model and showed positive effect of high EC treatment under both high and low substrate water conditions. MQUE and  $Q_y$  here show the quantum use efficiency when carboxylation activity is not the limiting factor at low PPF. However, at high PPF, quan-

tum requirement (the reciprocal of quantum yield) was larger in high EC treated plants (Figure 3). It was concluded that high-EC treated tomato plants could not use quanta efficiently at high PPF compared with tomato plants grown at low EC. However, this does not mean that the photochemical activity was lower in high EC treated plants. At high PPF, quantum supply is not the limiting factor. The limiting factors were carboxylation activity, mainly determined by the activity of Rubisco, and  $CO_2$  supply, mainly determined by stomatal conductance (Farquhar *et al.*, 1980). Therefore, at high PPF, high quantum requirement for high EC treated plants was attributed to the depression of Rubisco activity and/or stomatal conductance, rather than photochemical inhibition. Rubisco activity was determined not only by Rubisco content but also the ratio of activated/inactivated. Hence, it is not evident that Rubisco content was decreased. The Rubisco activity might have already been inhibited by high EC in the greenhouse; and/or might be, at least in part, inhibited at the measurement time because leaf water was lost (Figure 6) at high PPF levels. The decreases in leaf water potential at high PPF might be caused by the unbalance between water uptake and transpiration. High PPF itself may also inhibit non-stomatal activity in stressed plants (Givnish, 1988). The limiting factors for photosynthetic rate at low PPF are quantum supply and photochemically related factors, one of which is chlorophyll. If the activities of photochemically related factors are high, quantum can be used efficiently. So Pn is high and the initial slope of the light-response curve is large.

TABLE III  
Coefficients and parameters in light-photosynthesis curve for tomato plants maintained under different EC and soil water conditions

Treatment	$P_{max}/K$	K	r	$P_{max}$	$P_{1200}$	$Q_r$	$Q_y$
Low EC/High soil water	7.49 ± 0.52	3.81 ± 0.26	0.25 ± 0.03	19.7 ± 1.3	15.9 ± 0.4	15.1 ± 1.2	6.6 ± 0.5
Low EC/Low soil water	6.41 ± 0.33	3.89 ± 0.19	0.23 ± 0.02	16.5 ± 0.6	13.5 ± 0.8	18.0 ± 1.1	5.5 ± 0.3
High EC/High soil water	9.25 ± 0.42	5.99 ± 0.29	0.34 ± 0.04	15.4 ± 0.2	13.1 ± 0.5	12.4 ± 1.0	8.1 ± 0.6
High EC/Low soil water	8.71 ± 0.34	6.53 ± 0.32	0.31 ± 0.04	13.3 ± 0.7	11.9 ± 0.6	12.5 ± 1.0	8.0 ± 0.6

$P_{max}/K$  ( $10^{-2}$  mol  $CO_2$  mol<sup>-1</sup> PPF), the maximum quantum use efficiency when PPF tends to 0; K ( $10^{-3}$  m<sup>2</sup> s  $\mu$ mol<sup>-1</sup>), a constant and equals the reciprocal of the PPF when Pn =  $P_{max}/2$ ; r ( $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>), dark respiration rate;  $P_{max}$  ( $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>), the saturation photosynthetic rate at the maximum PPF level;  $P_{1200}$  ( $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>), photosynthetic rate at 1200  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup> PPF;  $Q_r$  (mol mol<sup>-1</sup>), quantum requirement (photon required for assimilation of one mole of  $CO_2$ );  $Q_y$  ( $10^{-2}$  mol mol<sup>-1</sup>), quantum yield, the reciprocal of  $Q_r$ ,  $CO_2$  mole assimilated by one mole of photon.

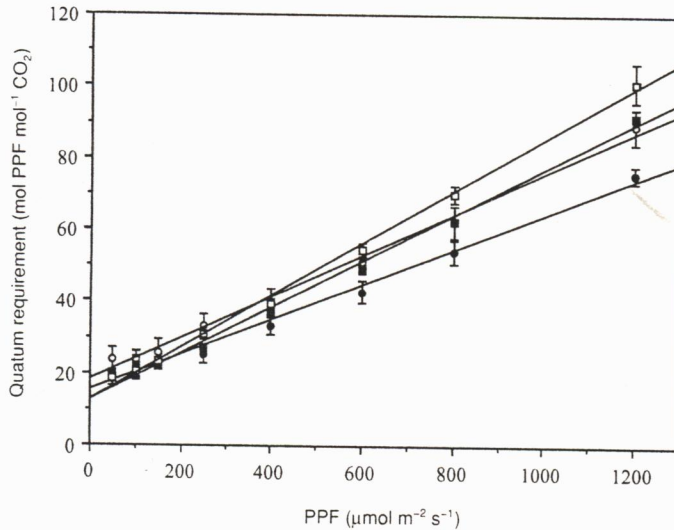


FIG. 2

Quantum requirement (PPF/photosynthesis ratio) in tomato plants treated with different nutrient solution electric conductivities and soil water contents. ● - LEC/HWC, low EC and high soil water content; ○ - LEC/LWC, low EC and low soil water content; ■ - HEC/HWC, high EC and high soil water content; □ - HEC/LWC, high EC and low soil water content. Data at each PPF level are the mean values of three measurements with a standard error shown by the vertical bars.

In our research, the increases in Pn at low PPF or the initial slope by high EC might be due to the increased chlorophyll content as described in the later paragraphs. Photochemical activity is positively related to chlorophyll content and nutrients, especially to the levels of N, Mg and Fe (Peterson and Onken, 1992). In the present work, the nutrient solution with high EC had high levels of these elements (Table I). The concentrations of nutrients in the substrate were also higher in high-EC treatment (Table II). Therefore, it is not clear whether high quantum use efficiency is attributed to high nutrient levels or to high EC. High EC decreased photosynthesis at high PPF to a significant level in the present work. There also have been reports on other species, in which photosynthesis is depressed by high EC or salt stress (Greenway and Munns, 1980; Cheesman, 1988; Norrie *et al.*, 1994). However, in other work (Xu *et al.*, 1994), where the substrates were overwatered every week to wash out the accumulated salts, a high EC of 4.0 to 5.5  $\text{mS cm}^{-1}$  did not decrease photosynthesis. Recent research by Adams (1991) has shown that tomato plants can tolerate high ECs when grown in hydroponic systems where there is no salt accumulation.

Therefore, effect of high EC on photosynthesis in the present work might be attributed to salt accumulation in the substrate (Table II).

Dark respiration ( $r$ ) was also increased by high EC under both well watered and water stressed conditions (Figure 1 and Table III). The effect of environmental stress on plant respiration varied according to plant growth stages and the extent of stress. Usually, a mild stress promotes respiration and a severe stress depresses respiration activity (Jones, 1983). However, even under soil water deficits, respiration was still higher in high EC treated plants. Therefore, it is not clear whether the respiration increase is related to high EC stress or some factors like nutrient levels other than stresses *per se*. Under mild or slowly imposed water or salt stress, osmotic adjustment can occur to avoid further tissue water loss and retain partial turgor maintenance. This process requires more energy expenditure by maintenance respiration (McCree, 1986). This might account for the increase in respiration under high EC conditions.

*Effect of low soil water content:* The effect of soil water deficit differed from that of high EC. Soil water deficit changed the light-response

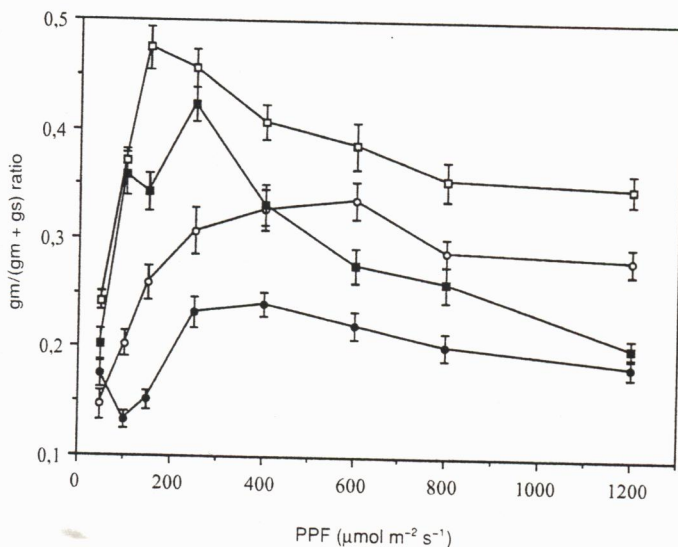


FIG. 3  
The ratio of  $g_m/(g_m+g_s)$  in tomato plants treated with different nutrient solution electric conductivities and soil water contents. -●- LEC/HWC, low EC and high soil water content; -○- LEC/LWC, low EC and low soil water content; -■- HEC/HWC, high EC and high soil water content; -□- HEC/LWC, high EC and low soil water content. Data at each PPF level are the mean value of three measurements with a standard error shown by the vertical bars.

curve by decreasing both initial slope and Pn at high PPF levels (Figure 1); The curve-fitting analysis showed that photosynthetic capacity (shown by  $P_{1200}$  and  $P_{max}$ ) was decreased significantly by soil water deficit under both low and high EC conditions (Table III). Soil water deficit also slightly decreased the maximum quantum use efficiency or quantum yield (shown by  $P_{max}K$  or  $Q_y$ ) under low EC conditions. The quantum requirement was higher for water stressed plants at all PPF levels (Figure 2). The results suggested that soil water deficit depressed activities of both photochemical process and carboxylation, but the effect on photochemical activity was small.

Dark respiration ( $r$ ) was hardly decreased by soil water deficit under both low and high EC conditions (Figure 1 and Table III). In the present experiment, plants with low soil water treatment showed no wilting even at midday. The extent of water stress was not sufficient to depress respiration.

#### Conductances and water use efficiency

*Effect of high EC:* From the response of photosynthetic capacity, we suggest that high EC decreases carboxylation and/or stomatal con-

ductance. The analysis of stomatal ( $g_s$ ) and mesophyll ( $g_m$ ) conductances helps to explain whether or not both carboxylation activity and stomatal conductance were depressed and which was more depressed. Stomatal conductance was decreased significantly by high EC at all PPF levels for both well watered and water stressed plants (Figure 1). However,  $g_m$  showed different light response from  $g_s$ . At low PPF,  $g_m$  was significantly higher in high EC treated plants, while at high PPF levels,  $g_m$  was lower in high EC treated plants. This phenomenon was consistent with, but clearer than, that of photosynthetic light-response. The result of  $g_s$  and  $g_m$  supported the result of photosynthetic capacity and showed that carboxylation activity and  $\text{CO}_2$  supply process were both affected by high EC although the extent of high EC effect on these two processes was different.

The ratio of  $g_m/(g_m+g_s)$  is a measure of the relative control of Pn by the two conductances. It was much higher in high EC than in low EC treated plants. This showed that photosynthetic depression caused by high EC was attributed more to  $g_s$  reduction than to  $g_m$  depression. Our data suggest that  $\text{CO}_2$  diffusion through stomata was more affected by the high EC treatment



than the biochemical processes in photosynthetic mesophyll. This also suggested that water use efficiency was high in high EC treated plants since more reduction of  $g_s$  means more depression of transpiration. Our results supported the hypothesis that water use efficiency usually increases as stomata close (Jones, 1980).

*Effect of soil water deficit:* Both stomatal ( $g_s$ ) and mesophyll ( $g_m$ ) conductances were decreased significantly by soil water deficit under low and high EC conditions (Figure 1). The  $g_m/(g_m + g_s)$  ratio was much higher in water stressed than in well-watered plants (Figure 3). This suggested that soil water deficit decreased  $g_m$  less than  $g_s$ . This also suggested that water use efficiency was higher for the water deficit treatment. Plants under water deficit conditions usually minimize transpirational water loss and maximize photosynthesis and show a high water use efficiency as a consequence (Jones, 1983). This is one kind of adaptation mechanism that allows plants to survive water deficit conditions.

#### *Chlorophyll content and water potential*

*Effect of high EC:* Chlorophyll a and b were significantly increased by high EC treatment under both well watered and water stressed conditions (Figure 4). High EC also increased the ratio of chlorophyll a/chlorophyll b. This result is consistent with that of quantum yield. In a given range, quantum yield is positively related to chlorophyll content as chlorophyll is important in collecting and transferring radiant energy in the process of light reaction (Rüdiger and Schoch, 1988). However, chlorophyll content was not positively related to photosynthetic capacity at high PPF. Our data suggest that chlorophyll content is a weak light factor for photosynthesis, i.e. chlorophyll content is positively related to photosynthesis only at low PPF. Usually, chlorophyll content and PPF during growth affect quantum yield (Givnish, 1988).

Leaf water potential was decreased by high EC under high and low soil water contents. The difference in leaf water potential between low EC and high EC treated plants was larger at midday than at predawn (Figure 5) and larger at high PPF than at low PPF (Figure 6). Usually, potential evapotranspiration is high at midday

or under high PPF. Under high transpiration demand, water uptake cannot balance the water consumption because high EC impedes water uptake (Awang *et al.*, 1993). The measurement of leaf water potential supported this hypothesis.

*Effect of soil water deficit:* Chlorophyll content was significantly higher in water stressed plants under the high EC regime. In the greenhouse, water stressed plants with the treatment of high EC had a deeper green colour, consistent with the analysis of chlorophyll content. However, under low EC conditions, water stressed plants showed only a slightly higher chlorophyll content and no deeper a green colour than well-watered plants.

Leaf water potential was decreased significantly by soil water deficit under low and high EC conditions. The difference in leaf water potential between well watered and water stressed plants was larger at midday than at predawn (Figure 5) and larger at high PPF than at low PPF (Figure 6). This suggested that the water uptake limitation caused by soil water deficit decreased leaf water potential, especially at midday and under high PPF. This result was consistent with that of photosynthetic depression, and also suggests that substrate water deficit affects leaf water potential additively with potential evapotranspiration.

#### *Additive effect of soil water deficit and high EC treatments*

In the tomato plants subjected to high EC combined with low soil water content,  $P_n$  was not significantly lower at low PPF, but it became significantly lower at high PPF than in other two individual stress treatments (Figure 1C).  $P_{max}$  obtained from the model analysis was significantly lower in this treatment (Table III). Our data suggested that high EC and substrate water deficit decreased photosynthetic capacity additively (effect of water deficit (19.7–16.5) + effect of high EC (19.7–15.4)  $\approx$  the effect of combined stress (19.7–13.3)). On the contrary, quantum use efficiency ( $P_{max}K$ ) or quantum yield ( $Q_y$ ) was increased by this treatment (Table III). This effect was synergistic between high EC and substrate water deficit.

Consistent with photosynthesis, both  $g_s$  and  $g_m$  were decreased by the combined treatment, but  $g_s$  was more depressed than  $g_m$  (Figure 1).

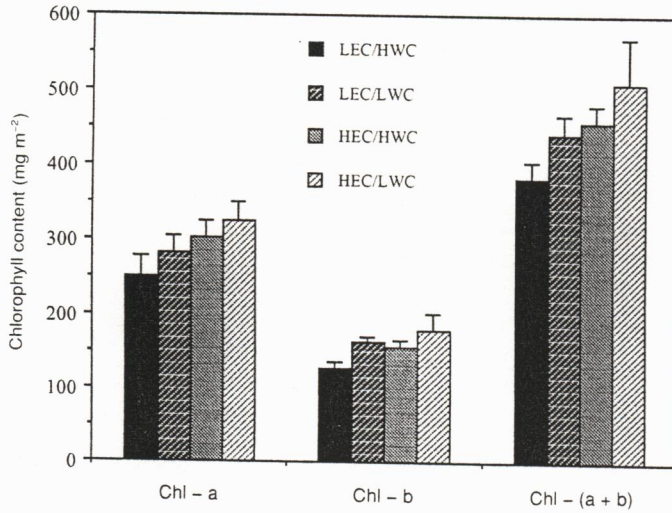


FIG. 4

Chlorophyll content in tomato plants treated with different nutrient solution electric conductivities and soil water contents. LEC/HWC. (low EC and high soil water content); LEC/LWC. (low EC and low soil water content); HEC/HWC. (high EC and high soil water content); HEC/LWC. (high EC and low soil water content). Data are the mean values of three measurements with a standard error shown by the vertical bars.

The result of  $g_m/(g_m + g_s)$  showed that photosynthetic depression in this treatment was much more contributed by  $g_s$  reduction than by  $g_m$  depression and that water use efficiency was

the highest in this treatment (Figure 3). This effect was also additive between high EC and substrate water deficit.

Chlorophyll content, whether shown by chlo-

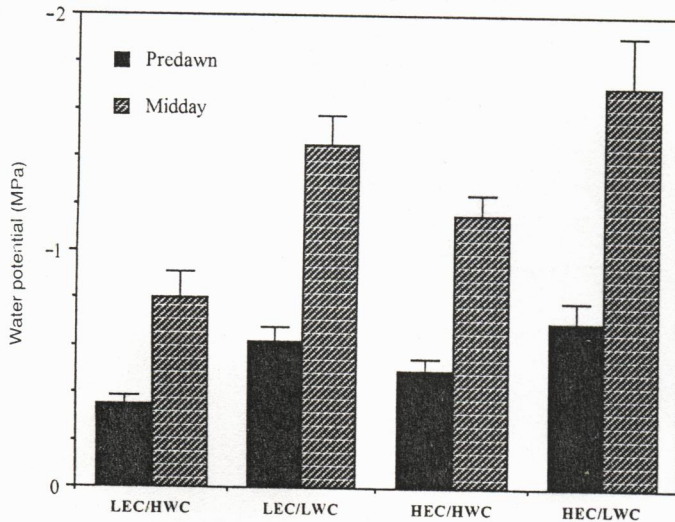


FIG. 5

Predawn and midday leaf water potentials in tomato plants treated with different nutrient solution electric conductivities and soil water contents. LEC/HWC, low EC and high soil water content; LEC/LWC, low EC and low soil water content; HEC/HWC, high EC and high soil water content; HEC/LWC, high EC and low soil water content. Data are the mean values of three measurements with a standard error shown by the vertical bar.

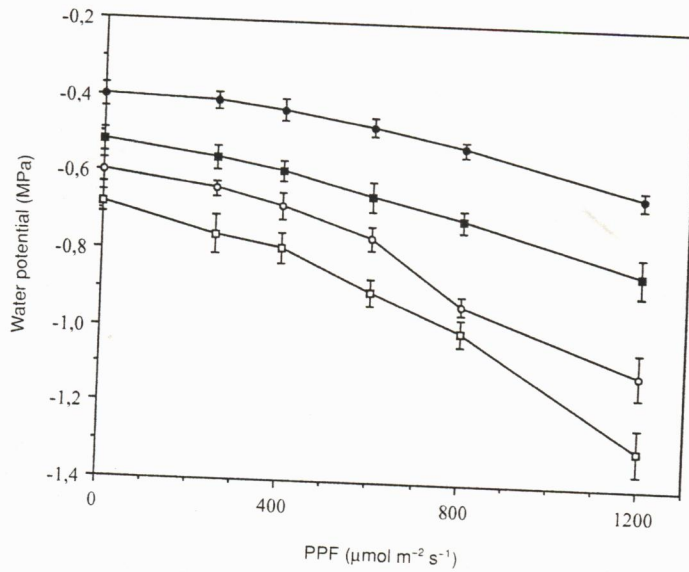


FIG. 6  
Response of leaf water potential to PPFD changes in tomato plants treated with different nutrient solution electric conductivities and soilwater contents. -●- LEC/HWC, low EC and high soil water content; -○- LEC/LWC, low EC and low soil water content; -■- HEC/HWC, high EC and high soil water content; -□- HEC/LWC, high EC and low soil water content. Data at each PPFD level are the mean values of three measurements with a standard error shown by the vertical bars.

rophyll a, chlorophyll b or chlorophyll a + b was highest in the treatment of high EC combined with substrate water deficit (Figure 4). This result was consistent with that of MQUE. The result of chlorophyll content supported the result of quantum use efficiency. The reason for the positive additive effect between high EC and soil water deficit on quantum use efficiency and chlorophyll content was not clear. However, it can be suggested that quantum use efficiency was quite stable to environmental stresses and that photosynthetic depression by high EC or substrate water deficit was attributed to the depression of stomatal conductance and/or inhibition of carboxylation activity instead of photochemical activity.

Leaf water potentials at both predawn and midday were the lowest in the treatment of high EC combined with substrate water deficit (Figure 5). At any PPFD, leaf water potential was the lowest in this treatment (Figure 6). The results showed clearly that high EC and soil water deficit decreased leaf water potential additively. Under low substrate water conditions, relative soil water volume is lower than under high substrate water conditions. The

osmotic potential in the soil water should be much lower than that in the irrigated nutrient solution. This low osmotic potential caused by the combination of low substrate water content and high EC impedes water uptake from soil to the roots (Awang *et al.*, 1993). This might be the reason for the additive effect between low substrate water content and high EC. The additive decrease in leaf water potential accounted for the photosynthetic depressions in an additive manner for the treatment of high EC combined with substrate water deficit. Photosynthetic performance under water stress is related not only to leaf water potential but also to turgor maintenance, which is more important for stomatal function. The detailed mechanism of water deficit effect on photosynthesis, especially in farm crops, is well documented (Close and Bray, 1993). However, so far there has been no report on the detailed explanations of the effect of high EC on photosynthesis; further studies and analyses are needed to elucidate the detailed mechanistic basis in terms of plant water relations.

#### CONCLUSION

The objective of the present work was to elu-

cidate the effect of high EC and low soil water content on photosynthesis and related processes in greenhouse tomato plants grown in a peat-moss based substrate. A rectangular hyperbolic model ( $P_n = P_{\max} KI(1 - KI)^{-1} - r$ ) fitted well the photosynthetic light-response curve. The photosynthetic capacity (the photosynthetic rate at saturation PPF) and the maximum quantum use efficiency (the initial slope of the curve) were analyzed from the curve fitting. High EC ( $4.5 \text{ mS cm}^{-1}$ ) treatment changed the curve shape by increasing initial slope (quantum use efficiency) and decreasing photosynthetic capacity at high PPF. The photosynthetic depression caused by high EC was attributed more to  $g_s$  reduction than to  $g_m$  depression. This suggested that  $\text{CO}_2$  diffusion through stomata was more affected than the biochemical processes. This also means that transpiration was more depressed than photosynthesis, i.e. water use efficiency (shown by  $g_m/(g_m + g_s)$  ratio) was increased by high EC. However, low soil water content not only decreased photosynthetic capacity at high PPF, but also slightly decreased quantum use efficiency. Low soil water content also increased water use efficiency. Chlorophyll content was increased by high EC treatment under both

high and low soil water content, and was consistent with the quantum use efficiency. Leaf water potential was decreased by high EC and low soil water content. Decreases in leaf water potential accounted for the  $P_n$  depressions. Our data also showed clear additive effect between high EC and soil water deficit on photosynthesis and most of its related processes. Photosynthetic capacity was depressed and the quantum use efficiency was increased to a larger extent in the combined treatment of high EC with low soil water content than in other two individual stress treatments. This result was supported by larger depressions in  $g_s$  and  $g_m$  as well as a larger increase in chlorophyll content. The effects of high EC on photosynthesis and related processes were large in the present experiment. Therefore, more detailed investigation and analysis are needed to elucidate the mechanisms in the aspects of salt accumulation and plant water relations.

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## Phenol induced browning and establishment of shoot-tip explants of 'Fuji' apple and 'Jinhua' pear cultured *in vitro*

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### SUMMARY

Several factors influencing phenol induced browning and establishment of apple (*Malus pumila* Mill. cv. Fuji) and pear (*Pyrus bretschneideri* Rehd., cv. Jinhua) were investigated by using shoot-tip explants cultured *in vitro*. Explants of 'Fuji' apple and 'Jinhua' pear collected from November to February produced low browning percentages. Browning percentages increased rapidly as the stock plants entered the growing season, reached a maximum during April and August and then decreased. Dark treatment of the stock plants reduced browning. Lowest incidence of browning and the highest surviving percentages of both 'Fuji' apple and 'Jinhua' pear were obtained after four weeks of dark treatment. However, bud-break percentage, fresh weight and the number of new leaves of the surviving explants decreased with increase in the length of dark treatment. When the dark treatment was used during the first phase of culture initiation, 5°C was better than 24°C for reducing browning and improving establishment of the explants. At 5°C, 6 d of dark treatment for 'Fuji' apple and 8 d for 'Jinhua' pear produced the least browning and the highest surviving percentages. However, bud break percentages, fresh weights and numbers of new leaves decreased as the length of the dark treatment increased. Addition of 2.5 g l<sup>-1</sup> activated charcoal to the initiation medium favoured establishment of 'Jinhua' pear explants and 100/150 mg l<sup>-1</sup> ascorbic/citric acid were effective with 'Fuji' apple.

BROWNING of explants cultured *in vitro* is a common problem which impairs the successful micropropagation of woody plants. Browning occurs through the action of polyphenoloxidase and tyrosinase on substrates such as chlorogenic acid and tyrosine (Lerch, 1981). High polyphenoloxidase activity and a high content of phenolic compounds results in much browning of *Cattleya* explants (Ichihashi and Kako, 1977; Ishii *et al.*, 1979).

Various methods have been developed for successful control of browning of explants in a number of plant genera and species other than apple and pear (George and Sherrington, 1984;

Zaid, 1987). By taking explants at right time of year, browning of explants can be reduced. In *Hamamelis*, browning intensity was greatest from explants taken in May and lowest in July and August (Christiansen and Fønnesbech, 1975). Vieitez and Vieitez (1980) reported that browning of chestnut was inhibited by washing explants for 2-3 h or soaking in sterile distilled water prior to *in vitro* initiation. Transfer of explants to fresh culture medium within 1-2 d of initial culture has been recommended to prevent browning in blackberry (Broome and Zimmerman, 1978). Browning was totally overcome by culturing *Dioscorea batatas* stem segments in an inverted position *in vitro* (Asahira and Nitch, 1969). Addition of absorbing agents such as activated charcoal (AC, Preil and

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