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# EFFECTS OF $O_2$ IN AIR AND NaCl IN MEDIUM ON PHOTOSYNTHESIS AND PHOTORESPIRATION IN TWO COTTON CULTIVARS

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MERT H. H. *Effects of $O_2$ in air and NaCl in medium on photosynthesis and photorespiration in two cotton cultivars.* BIOTRONICS **15**, 1–7, 1986. The measurements of photosynthesis and photorespiration in the two cultivars of cotton plant under different salt and oxygen concentrations showed that, *Gossypium hirsutum* cv. Coker 100 A/2 was less tolerant than *G. hirsutum* cv. 2421-A. There was a decrease in the photosynthesis with an increase in the salt concentrations at both 21 and 2% oxygen concentrations, however, the inhibition was higher at the former as compared to the latter oxygen concentration. The photorespiration too was low in both the cultivars but, significantly higher in normal air (21% oxygen) than in an air of low oxygen (2%) content.

**Key words:** *Gossypium hirsutum* L.; cotton; oxygen; salt; sodium chloride; photosynthesis; photorespiration.

## INTRODUCTION

Several investigations have been carried out on the effects of salts on the photosynthetic behaviour of plants. According to Tiku (*13*), in the species of *Salicornia* and *Distichlis*, the $CO_2$ uptake increases in all the salt concentrations, however, it was observed that, in different cultivars of cotton grown from the seeds pretreated with different concentrations of NaCl for different periods, photosynthesis of these plants decreases gradually in relation to the NaCl content of the medium (*8*), which could be a characteristic for the glycophytes. The findings concerning the respiratory behaviour of the plants in relation to the salt content too are contradictory. Nieman (*9*) has observed an increase in the respiration of the plants grown in a nutrient medium with salt. On the other hand, Boyer (*3*) reported that salt has an inhibitory effect on this process.

The photorespiratory behaviour of the plants in relation to the oxygen, carbon dioxide, light and temperature has also been studied by many workers, such as Forrester *et al.* (*5*), Bjorkman *et al.* (*2*), Ludvig and Canvin (*7*) and Laing (*6*), however, not much work has been done on the effects of salt on this phenomenon. From this view point, the studies were carried out on the photosynthesis and photorespiration in two cultivars of cotton, designated as a semihalophyte by Waisel (*16*), using different concentrations of salt and oxygen.

## MATERIAL AND METHODS

The two cultivars of cotton, *Gossypium hirsutum* L., cv. Coker 100 A/2 and 2421-A were used in the present studies: the former being less tolerant to the salt and the latter more tolerant (*1, 4*). The seeds were sown in rockwool supplied with 0.1 percent superba nutrient solution shown in Table 1 (*10*).

After the plant growing for 15 days, 0.3, 0.6 and 1.0 percent solutions of NaCl were supplied to seedlings in different pots. The salt solutions were applied 3 times on different days in a week to these seedlings. The experiments were carried out in a biotron at 30°C with a photoperiod of 16 h and 70% RH. The light source was 400 W Phillips HPI/T fluorescent tubes lying at a distance of 65 cm from the pots.

Table 1. Composition and percentage distribution of elements in superba

| Elements | mg per g superba |
|---|---|
| N | 130 |
| P | 40 |
| K | 190 |
| S | 53 |
| Mg | 15 |
| Fe | 1 |
| Mn | 0.4 |
| Cu | 0.4 |
| B | 0.1 |
| Zn | 0.2 |
| Mo | 0.03 |
| Co | 0.01 |

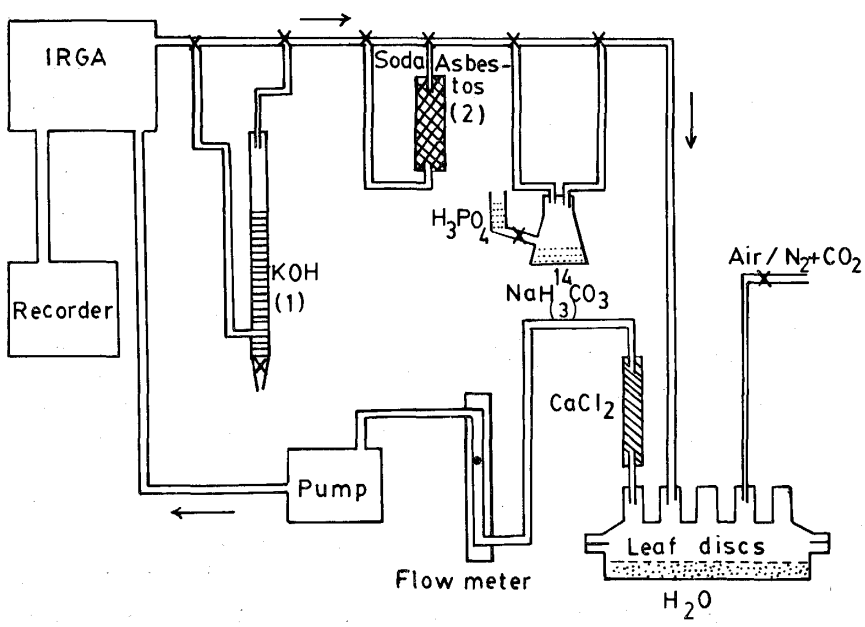


Fig. 1. Diagram of the closed system assimilation chamber.

The experiments were performed in a closed system assimilation chamber (Fig. 1). The reaction vessel was 75 mm in diameter and 20 mm deep. The cover of the vessel was fitted with a valve combination of the quickfit MAF I/75 with five outlets through which the gas could be directed. The gas leaving the chamber passed through a column of $CaCl_2$ for dehydration and then through an infrared $CO_2$ gas analyzer model Beckman 865, calibrated with $N_2$ and $N_2$ +378 ppm $CO_2$. The signal from the gas analyzer was recorded with a Watanabe milivolt recorder, and the gas returned to the reaction vessel through a glass tubing. The gas could be directed alternately by manipulating valves in to one of the three loops: leading either to a solution of 10 ml of 0.5 M KOH (1), where $CO_2$ was absorbed and stored for scintillation measurements; to a container of soda asbestos (2), when $CO_2$ free air was desired; or to a third loop connecting the system to a flask (3), where $^{14}CO_2$ was produced from $NaH^{14}CO_3$. The total volume of the assimilation chamber with tube connections was 0.76 l.

Two slide projectors with quantum flux densities of 1500 $\mu$E $m^{-2}$ $s^{-1}$ from the top and 2500 $\mu$E $m^{-2}$ $s^{-1}$ from below were used for irradiation and $^{14}CO_2$ fixation and photorespiration were measured under white light. The flux densities were measured with a Li-cor-modell Li-185.

About 40 days old plants were used and the leaf discs were taken from the third youngest leaf for the measurement. Thirty leaf discs (11 mm in diameter and 0.28 $dm^2$ in the total leaf area) were floated with the upper epidermis up, on 75 ml distilled water in the reaction vessel. After placing the material in the vessel, the system was flushed for 3 min with normal air, or with $N_2$+$CO_2$ gas mixture for obtaining $O_2$ free air. For the treatment of 2% $O_2$, 15 ml of $O_2$ was injected to the chamber by a syringe. The measurement was performed as follows: After the measurement of dark respiration for 6 min, the $CO_2$ was removed from the system through the column of soda asbestos for 6 min. It was then opened to the $CO_2$ reservoir tube containing 1 ml unlabeled 0.02 M $NaHCO_3$ solution and 25 $\mu$l of $NaH^{14}CO_3$ (0.587 mCi $\mu mol^{-1}$), to which 4 ml of 20% $H_3PO_4$ was injected just prior to opening. The $^{14}CO_2$ supply to the system was stopped after a short period of equilibrium (6 min) and the leaf discs then irradiated with white light for measurement of photosynthesis and photorespiration. Any remaining $^{14}CO_2$ was removed from the system by absorption on soda asbestos. The assimilation chamber was flushed with normal air (approx. 350 ppm $CO_2$) or to $N_2$+$CO_2$ gas (378 ppm $CO_2$). The chamber was again irradiated and the system immediately opened to the KOH column in order to recover the $^{14}CO_2$ released from the leaf discs during photorespiration. After 0, 2, 6 and 8 min, 1.5 ml samples were taken from the KOH column. The system was then cleared of all $CO_2$ by opening it to the soda asbestos.

The KOH samples were prepared for scintillation counting, by placing 1.0 ml in a scintillation vial and adding 10 ml toluene solution, containing 5 g PPO (2,5-Diphenyloxazole), 0.05 g POPOP (1,4-bis(2-(5-Phenyloxazolyl))benzene) and 30% triton x-100 per liter. The vials were refrigerated overnight, and radioactivity was measured with Philips scintillation counter.

The results, were tested statistically using the method by Steel and Torrie (*12*).

## RESULTS AND DISCUSSION

Table 2 shows the rate of photosynthesis in Coker 100 A/2 as well as 2421-A. The photosynthetic rate decreased with increasing salt concentrations in both air conditions, but the decrease of photosynthesis in both cultivars at 2% $O_2$ is smaller than that in normal air. Although, in the case of Coker 100 A/2, the decrease rate of photosynthesis in the plants treated 1% salt as compared to that in the control plants is 34.5% in normal air, it is only 26.8% in 2% $O_2$. In the case of 2421-A, the decrease is 24.0% in normal air and 21.4% in 2% $O_2$. This fact shows that there is some relation between salt tolerance and the $O_2$ effect on photosynthesis. The rate of photosynthesis is higher in 2421-A cv. than in Coker 100 A/2 at 1.0% salt concentration in both air conditions, which is related to the other physiological factors of these cultivars, e.g., disintegration of chlorophyll and protein content in Coker 100 A/2. It is necessary to make further analysis to solve this problem.

Table 3 shows the photorespiration data measured as the $^{14}CO_2$ release after a period of $^{14}C$-labelling in white light. Here again, the photorespiration was significantly higher in normal air than in the air with the lower $O_2$-content. There

Table 2. Net photosynthetic rate ($\mu$g $CO_2$ $dm^{-2}$ $h^{-1}$) of the leaf discs, illuminated with white light under different air conditions (mean of eight experiments)

| Air | Salt conc. (%) | Coker 100 A/2 | 2421-A |
|---|---|---|---|
| Normal air | Control | 4202 | 3898 |
| | 0.3 | 3451 (17.9)* | 3284 (15.8) |
| | 0.6 | 3091 (26.4) | 3072 (21.2) |
| | 1.0 | 2757 (34.4) | 2962 (24.0) |
| 2% $O_2$ | Control | 7275 | 6956 |
| | 0.3 | 6318 (13.2) | 6294 ( 9.5) |
| | 0.6 | 6029 (17.1) | 5906 (15.1) |
| | 1.0 | 5324 (26.8) | 5465 (21.4) |

LSD (5%): 0.64 at Coker 100 A/2; 0.69 at 2421-A.
* Percent reduction of photosynthetic rate against the control.

Table 3. Photorespiration rate (CPM $^{14}CO_2$), released after a period of photosynthesis in white light in normal air and in 2% $O_2$ under different salt concentrations in two cotton cultivars (mean of eight experiments)

| Air | Salt con. (%) | Coker 100 A/2 | 2421-A |
|---|---|---|---|
| Normal air | Control | 1228 | 2041 |
| | 0.3 | 1129 | 1883 |
| | 0.6 | 895 | 1663 |
| | 1.0 | 559 | 1204 |
| 2% $O_2$ | Control | 522 | 798 |
| | 0.3 | 450 | 708 |
| | 0.6 | 398 | 665 |
| | 1.0 | 369 | 583 |

LSD (5%): 73 at Coker 100 A/2; 80 at 2421-A.

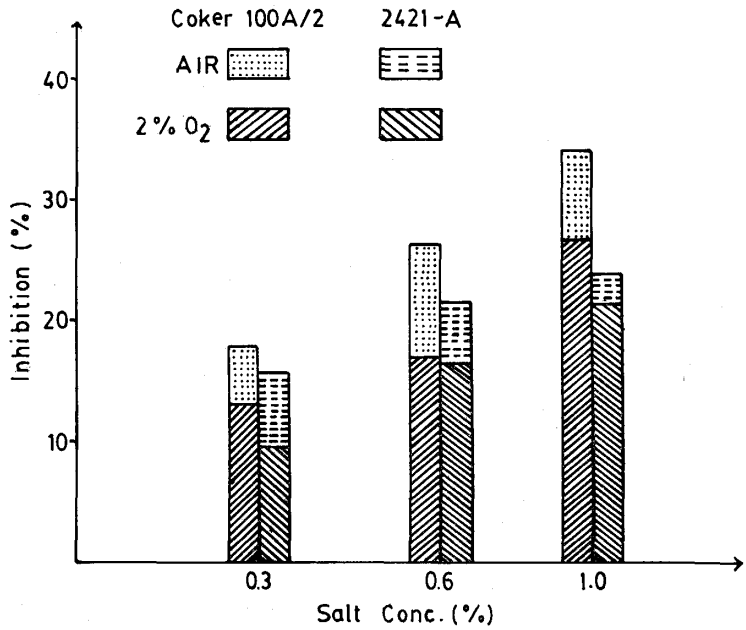


Fig. 2. The inhibitive effect of salt concentration on the photosynthesis of the two cotton cultivars as compared to the control.

is a great difference between Coker 100 A/2 and 2421-A in air. As is apparent from the table too, rates of photorespiration decreased gradually with an increase in the salt concentration of the medium in both cultivars.

The photorespiration rate in 2421-A was higher than that in Coker 100 A/2 in each salt concentration, and both in normal air and air of 2% $O_2$. However, photorespiration rate in air of 2% $O_2$ is almost half of that in normal air.

The photosynthesis in the cotton cultivars (Coker 100 A/2 and 2421-A), when measured as $CO_2$ uptake in the white light, seems to be dependent on oxygen as well as the salt concentrations. The percentage decrease in photosynthesis in relation to the salt in normal air is higher than that at 2% oxygen level (Table 2). No significant effect of NaCl on the photosynthesis of salt tolerant cultivar 2421-A was found in both air conditions.

This indicates that salt effect is dependent on $O_2$ concentrations in these different cultivars. These findings are in full agreement with those of Aydemir (*1*) and Emiroğlu (*4*). It is a well known fact that the glycolate pathway is saturated by 2% oxygen level (*15*) and glycolate production during photorespiration increases with oxygen concentration up to 100 percent level (*14*). Therefore, the effect of oxygen on the photosynthesis seems to be limited to the glycolate pathway rather than to the activity of RuDP carboxylase/oxygenease activity. According to Shomer-Ilan and Waisel (*11*), in the plants provided with a NaCl solution, the leaves show high activity of PEP carboxylase, as well as a significant $CO_2$ fixation via $C_4$-pathway. However, these findings are not parallel to those of ours (Table 2), which may be due to the fact that we used $C_3$ plants.

The photorespiration, when measured as the $^{14}CO_2$ release in air and air of low $O_2$, also shows an oxygen dependence (Table 3). These observations reveal that $CO_2$ release increases at higher oxygen concentrations, which stimulate glycolate production from the splitting of RuDP to PGA and P-glycolate. The production of glycolate in this way shows a linear correlation to the oxygen concentration between zero and 21 percent (*14*). Our results fully support the findings of Tolbert (*14*). The salt concentration also affected the photorespiration in the two cultivars of cotton plant (Table 3). The photorespiration rate is significantly higher in normal air than in the air of low $O_2$. Our findings show that, under such a condition the effect of the salt concentrations resembles that of the $O_2$ concentrations, as the salt concentrations stimulate glycolate production in the two cotton cultivars. But, the mechanism of the effect of the salt concentrations on the photorespiration of the plants still needs an explanation.

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