

## DIFFERENT SENSITIVITY OF PHOTOSYNTHESIS AND TRANSPIRATION TO SOME HERBICIDES AND TRANSPIRATION INHIBITORS

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### SUMMARY

Rates of transpiration and net photosynthesis in intact sugar beet and bean plants were measured during 14 hours' exposure to some herbicides, to transpiration inhibitors, and to high osmotic pressure.

Transpiration and photosynthesis were inhibited to the same extent or transpiration slightly stronger, by treatment of the roots with ioxynil, by application of PMA and DSA either to the rooting medium or to the foliage, and by high osmotic stress. Photosynthesis, however, was inhibited to a greater degree than transpiration by simeton and isocil applied both to the roots and the leaves, and by a foliar spray of ioxynil. Similar, but somewhat smaller differences were found when DNOC was applied to the nutrient solution or to the leaves.

### 1. INTRODUCTION

In previous studies we found that herbicides interfering with light reactions in chloroplasts reduced photosynthesis much more than they did transpiration when applied to the nutrient solution of various plant species (e.g. VAN OORSCHOT 1965). This was ascribed to partial closure of the stomata due to inhibition of CO<sub>2</sub> assimilation. Similar differences between photosynthesis and transpiration were observed when diquat was applied to the nutrient solution of bean plants, but also with leaf sprays (VAN OORSCHOT 1966).

To compare the effects of various herbicides tests were made with sugar beet plants. The effects of specific inhibitors of photosynthesis like simeton<sup>1</sup> and isocil, and those of other herbicides like ioxynil and DNOC were compared with the effects of transpiration inhibitors like PMA and DSA, and conditions of high osmotic stress. These experiments were repeated with bean plants which had their leaves fixed in a horizontal position. Some preliminary results were published earlier (VAN OORSCHOT 1970).

### 2. MATERIAL AND METHODS

Details of the continuous recording of CO<sub>2</sub> uptake and transpiration of intact

<sup>1</sup>Abbreviations of the compounds used— DNOC: 2-methyl-4,6-dinitrophenol; DSA: *n*-2-decenylsuccinic acid; ioxynil: 4-hydroxy-3,5-di-iodobenzonitrile; isocil: 5-bromo-3-isopropyl-6-methyluracil; PMA: phenylmercuric acetic acid; simeton: 4,6-bisethylamino-2-methoxy-1,3,5-triazine.

plants were described by LOUWERSE & VAN OORSCHOT (1969). The experiments were made at 24–26°C. Pure, analytical material of the herbicides was used. Before applying the compounds, gas exchange and transpiration were measured during a short dark period followed by 2–3 hours light of around  $140 \text{ J}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  ( $\lambda < 700 \text{ nm}$ ). The roots were outside the plant chamber and could be treated while the measurements continued, the normal nutrient solution being replaced by one containing the compound. For leaf application the area of each leaf was determined at the beginning of the experiment. After dissolving the compound in de-mineralized water a measured volume of aqueous solution was sprayed on each leaf with a microsprayer (VAN OORSCHOT 1966), usually equivalent to 200 l per ha leaf area. In case of DNOC and DSA a volume of 400 l per ha was used. In these experiments the measurements were discontinued for the 15–20 minutes required for opening the plant chamber, spraying the leaves, closing and re-equilibrating the air flow. The measurements continued during 10–15 hours in light of  $140 \text{ J}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  and a subsequent dark period of 1–2 hours. The rates of transpiration and net photosynthesis were expressed and plotted as a percentage of the initial rates of the same plants before treatment. In the graphs dark respiration and dark transpiration at the beginning and end of the experiment were plotted as short horizontal bars.

In each experiment with sugar beet (*Beta vulgaris* L. cv. Klein Wanzleben) 4 plants with 2 developed leaves were placed in a plant chamber. Their leaves normally were about horizontal, but some treatments resulted in drooping of the leaves by a gradual loss of turgor during the first hours. Drooping leaves are less efficient in light interception than horizontal ones. Therefore, the experiments were repeated with beans (*Phaseolus vulgaris* L. cv. Berna) with the primary leaves forced into a horizontal position. All plants were germinated in moist gravel, placed on nutrient solution, and grown in a climate room at 20°C at a light intensity of  $70 \text{ J}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  prior to the experiments.

### 3. RESULTS AND DISCUSSION

Transpiration was inhibited to the same extent as or somewhat greater extent than photosynthesis by high osmotic pressure in the rooting medium, treatments with transpiration inhibitors, and root treatment with ioxynil. The results of these treatments are given in part A of table 1, and some of them in more detail in fig. 1 and 2.

If at zero time the nutrient solution of sugar beets was replaced by one containing sodium chloride in a concentration corresponding to an osmotic pressure of 2.5 atmospheres, the degree of inhibition of transpiration and photosynthesis was about the same. This result is shown in fig. 1. After some fluctuation during the first few hours, transpiration and photosynthesis were constant at about 65% of their initial rates. Similar experiments with sugar beets and beans at the same osmotic pressure and at 5 atmospheres gave similar results (table 1A), although in two cases transpiration was somewhat more reduced than photosynthesis. At 5 atmospheres the leaves wilted at first, but gradually re-

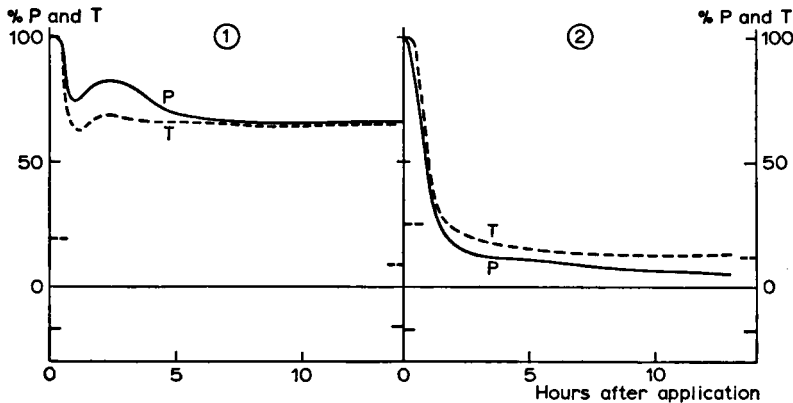


Fig. 1, 2. Relative rates of transpiration (T, ---) and net photosynthesis (P, —) of sugar beets after the application of 2.5 atmospheres osmotic pressure to the rooting medium (fig. 1), or after the application of  $10^{-4}$ M ioxynil to the rooting medium (fig. 2).

gained full turgor after a few hours. Transpiration and photosynthesis of maize were equally reduced by the application of high osmotic pressure (2.5 atm). If, in other experiments, the water supply of sugar beet and maize was discontinued by suspending the roots in the air, again inhibition of photosynthesis closely followed that of transpiration.

This coordinate inhibition suggests that the falloff in transpiration caused by water stress is due to a mechanism that equally affects photosynthesis. It is known that water stress reduces stomatal aperture, but the equal effect on photosynthesis and transpiration suggests that it also reduces the diffusion of carbon dioxide from the stomata to the chloroplasts in the mesophyll cells. Otherwise photosynthesis would be less sensitive to water stress than transpiration. MAKINK & VAN HEEMST (1972) also observed an equal degree of inhibition of transpiration and photosynthesis in drying of bean leaves, while BOYER (1970) found a more or less equal response of transpiration and photosynthesis during desiccation of maize and soybean.

Strictly comparable results were obtained when ioxynil or PMA were added to the rooting medium (table 1A, fig. 2). Again, equal inhibition was effected within the first few hours, transpiration and photosynthesis fell to low rates, and loss of leaf turgor occurred in sugar beets. Although the leaves gradually regained full turgor, both rates remained at a low level throughout the experiment. Such wilting was not observed with beans, but the treatments were also somewhat less effective. The degree of correlation suggests that ioxynil and PMA, when applied to the rooting medium, affected water transfer across the medium-root interface in such a measure as to cause a water deficit comparable to that caused by sodium chloride.

Foliar application of PMA to sugar beet resulted in a lower effect at the dose applied. There was no effect of the same and higher rates (up to 1.5 mole/ha) on

Table 1. Rates of transpiration (T) and net photosynthesis (P) after 7 hours of treatment with the various compounds, expressed in percentages of the initial, uninhibited rates. Data under A for equal or greater inhibition of transpiration, those under B for greater inhibition of photosynthesis.

Treatment	Sugar beet						Bean			
	1		2		3		1		2	
	T	P	T	P	T	P	T	P	T	P
<b>A</b>										
sodium chloride, 2.5 atm.	65	67	60	80	55	68	54	56	73	76
sodium chloride, 5 atm.	18	20	15	19						
ioxynil, $10^{-4}$ M, roots	13	9	11	20	17	20	79	68	71	55
PMA, $2 \times 10^{-5}$ M, roots	14	17	20	24	33	37	51	44	22	15
PMA, 0.3 mole/ha leaves	71	61	98	85					no inhibition	
DSA, $10^{-4}$ M, roots	34	54	31	62	39	53			no inhibition	
DSA, 0.6 mole/ha leaves	72	86	76	91	71	87			no inhibition	
<b>B<sup>1</sup></b>										
simeton, $2 \times 10^{-5}$ M, roots	56	-20	28	-10	52	-2	85	-2	86	21
simeton, 0.3 mole/ha leaves	61	3	53	-5	58	10	96	43	100	31
isocil, $2 \times 10^{-5}$ M, roots	63	-20	33	-15			88	-15	62	-3
isocil, 0.3 mole/ha leaves	47	-2	73	27	55	-6	100	22		
ioxynil, 0.3 mole/ha leaves	64	21	90	20	71	47	64	9	79	2
DNOC, 0.3 mole/ha leaves	68	30	82	54	76	46	99	79	100	80
DNOC, $10^{-4}$ M, roots	36	20	33	-2	23	26	76	43	53	27

<sup>1</sup>Beans:  $10^{-5}$ M, roots and 0.1 mole/ha leaves of simeton and isocil; 0.6 mole/ha leaves and  $2.5 \times 10^{-4}$ M, roots of DNOC.

beans. As with root treatments the differences between transpiration and photosynthesis of sugar beets were small. In this respect these results are in contrast with those obtained with other plants cultivated in soil, where transpiration was somewhat more inhibited (e.g. SLATYER & BIERHUIZEN 1964). Foliar application of ioxynil gave completely different results, which will be discussed later.

DSA applied to the rooting medium caused temporary wilting of sugar beet leaves and resulted in a substantial decrease in transpiration which was paralleled by a decrease in photosynthesis. Here, however, the degree of inhibition of photosynthesis was less than that of transpiration. The effect of a foliar spray was similar, but smaller owing to dose or slow penetration.

Those treatments with herbicides resulting in much less inhibition of transpiration than of photosynthesis are given in part B of table 1. Also included are root treatments with DNOC, although here the difference between both processes is smaller than with the other treatments.

Fig. 3 shows the effect of simeton applied to the rooting medium of bean plants. Here photosynthesis was completely inhibited within some hours, whereas transpiration was only partly suppressed. A similar difference was found with sugar beets. Much greater inhibition of photosynthesis than of transpiration was also observed after leaf treatment with simeton, and with isocil ap-

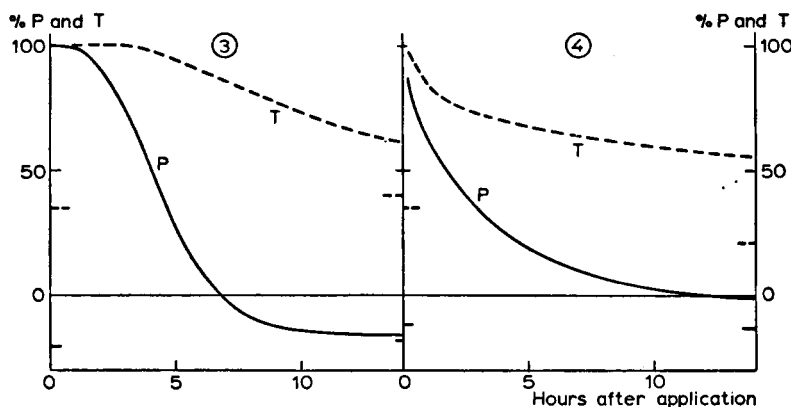


Fig. 3, 4. Relative rates of transpiration (T, ---) and net photosynthesis (P, —) of beans after the application of  $10^{-5}$ M simeton to the rooting medium (Fig. 3), or after foliar application of 0.3 mole ioxynil per ha leaves (Fig. 4).

plied either to the rooting medium or as a foliar spray (*table 1B*). In some of the sugar beet experiments the transpiration inhibition was partly relieved during the second half of the experimental period. These herbicides are specific inhibitors of light reactions in photosynthesis (e.g. MORELAND 1969; VAN RENSEN 1971), and various data indicate that they easily accumulate in the leaves when supplied to the roots (e.g. CRAFTS & ASHTON 1970). Inhibition of photosynthesis in the chloroplasts will probably lead to increased carbon dioxide concentration in the leaf, and this may result in a certain degree of stomatal closure. Closure of stomata induced by another photosynthesis inhibitor, 3-(4-chlorophenyl)-1,1-dimethylurea, could be relieved by flushing the intercellular spaces with air free of carbon dioxide (ALLAWAY & MANSFIELD 1967).

Leaf sprays with ioxynil on beans (*fig. 4*) and sugar beets (*table 1B*) also resulted in a much greater inhibition of photosynthesis than of transpiration. The correspondence with the effects of simeton and isocil suggests that ioxynil, when applied to the leaves, also primarily inhibits photosynthesis. KERR & WAIN (1964) have demonstrated that ioxynil and DNOC, known as uncouplers of oxidative phosphorylation, may also inhibit light reactions in chloroplasts. The completely different result of root treatment with ioxynil (*fig. 2, table 1A*) could possibly be explained by a limited mobility in the plant.

The result of foliar application of DNOC on sugar beet (*table 1B*) was rather similar to that of ioxynil, photosynthesis being much more suppressed than transpiration. The effect on bean was lower, but showed the same trend. As with ioxynil this result suggests that primarily photosynthesis is inhibited. When applied to the rooting medium DNOC reduced photosynthesis somewhat more than transpiration, but the differences are smaller than with leaf sprays. The temporary wilting of sugar beet leaves indicates that water stress effects may also play a role here. At similar concentration in the nutrient solution as

applied in these experiments such wilting has also been reported for winter rye (BRUINSMA 1967), and for tomato with the related 2-*sec*-butyl-4,6-dinitrophenol (SMITH & BUCHHOLTZ 1964). On the other hand, inhibition of photosynthesis in the chloroplasts may occur, because DNOC is translocated to the leaves when applied to the roots (BRUINSMA 1967).

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