

Short Communication

Rapid Action of Abscisic Acid on Photosynthesis and Stomatal Resistance

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Summary. (RS)-Abscisic acid, a natural plant hormone, has been found to inhibit photosynthesis in both detached and attached primary wheat leaves. The action occurs rapidly and is accompanied by large increases in stomatal diffusive resistance.

The plant hormones gibberellin, auxin and kinetin have been shown to promote the rate of photosynthesis in various plants (Coulombe and Paquin, 1965; Turner and Bidwell, 1965; Meidner, 1967, 1969; Wareing *et al.*, 1968). It has been shown previously that ABA inhibits transpiration (Little and Eidt, 1968; Mittelheuser and Van Steveninck, 1969) and induces stomatal closure (Mittelheuser and Van Steveninck, 1969) as measured by silicone-rubber impressions of leaf surfaces after 6 h treatment. Recently Imber and Tal (1970) have extended these findings to show that foliar applications of ABA caused the morphological reversion of a wilted tomato mutant to the normally transpiring phenotype by inducing stomatal closure and lowering the rate of transpiration. It now will be shown that transpiration-supplied ABA causes a rapid increase in wheat leaf stomatal resistance and decrease in CO₂ assimilation.

Wheat plants (*Triticum aestivum* L. cv. Mendos) were grown in vermiculite and nutrient solution. When the plants were 10 days old they were transferred to conditions for photosynthesis or stomatal resistance measurement (light intensity of 30,000 lux supplied by a Quartz-iodide lamp with water filter, $26 \pm 1^\circ$) for 1 h before further experimental treatment. In experiments using detached leaves, primary leaves were placed in individual vials containing either deionized water or 3.8×10^{-6} M ABA and the rates of photosynthesis and stomatal resistance measured at the times indicated. Intact plants were carefully washed free of vermiculite under running water and the roots placed in small beakers containing either deionized water or 3.8×10^{-6} M ABA. The ABA solution was protected from light. After detachment and area measurement 4 leaves (2 ABA and 2 control) were allowed to equilibrate with medical air for 45 sec in a water-jacketed glass photosynthesis chamber before exposure to ¹⁴CO₂-air (300 ppm) for 60 sec followed by flushing with medical air for 20 sec. Assimilated ¹⁴CO₂ was assayed by liquid scintillation at 76 %

Table 1. *Effect of 3.8×10^{-6} M ABA on the rate of photosynthesis and on stomatal resistance (Rs) of detached primary wheat leaves*

Control leaves were treated with deionized water (W). Measurements of rates of photosynthesis and of stomatal resistance were made on different leaves.

Time (min)	Photosynthesis			Rs (sec cm ⁻¹)	
	mg CO ₂ dm ⁻² h ⁻¹		% in- hibition	W	ABA
	W	ABA			
0	11.0 ± 0.7 ^a	—	—	—	—
5	—	—	—	2.8	2.1
10	6.2 ± 0.4	6.0 ± 0.9	—	1.8	10.3
15	—	—	—	6.7	15.7
18	8.4 ± 0.6	4.0 ± 0.9	48	—	—
20	—	—	—	6.0	28.8
21	12.2 ± 0.6	2.8 ± 0.6	23	—	—
25	—	—	—	4.0	14.3
26	14.4 ± 3.6	2.8 ± 0.3	19	—	—
30	10.8 ± 0.3	3.4 ± 0.6	32	2.8	15.7
40	7.0 ± 0.7	2.2 ± 0.1	31	3.5	34.5
45	8.4 ± 0.1	3.0 ± 0.1	36	—	—
50	10.8 ± 0.6	3.2 ± 0.6	30	—	—
60	—	—	—	2.6	22.8
90	11.4 ± 0.7	4.4 ± 0.4	39	—	—

^a Standard error of the mean.

efficiency using the method of Shimshi (1969) modified to correct for fluorescence. Stomatal resistance was measured with a water-vapour-diffusion porometer (Kanemasu *et al.*, 1969). Single measurements were made on different leaves.

Rates of photosynthesis and stomatal resistance of detached leaves treated with ABA or deionized water *via* the transpiration stream are presented in Table 1. ABA rapidly caused a marked inhibition of ¹⁴CO₂ assimilation and enhancement of stomatal resistance. In 4 experiments, stomatal resistance was significantly higher in ABA-treated leaves after only 10 min treatment, and in 6 experiments after 15 min treatment. Results of photosynthetic rates of attached leaves of plants supplied with ABA or deionized water *via* the root system are presented in Table 2. In intact plants photosynthesis was also inhibited strongly by ABA, but short-term experiments showed that this effect was less rapid than in the case of detached leaves.

The mode of action of ABA in reducing the rate of photosynthesis and increasing stomatal resistance is unknown. The effect occurs very rapidly. Also it should be considered that introduction into the transpiration stream and transport to the active site is required. However, stomata

Table 2. *Effect of 3.8×10^{-6} M ABA on the photosynthetic rate of attached primary wheat leaves*

Control leaves treated with deionized water (W).			
Time (h)	Photosynthesis (mg CO ₂ dm ⁻² h ⁻¹)		% of control (ABA inhibition)
	W	ABA	
0	14.6 ± 0.7 ^a		
0.5	6.0 ± 0.7	7.0 ± 1.1	—
1	10.8 ± 0.6	3.2 ± 0.1	30
1.5	15.6 ± 1.0	7.0 ± 1.1	45
2	12.6 ± 1.6	7.8 ± 0.7	62
4	11.2 ± 1.4	3.4 ± 0.7	30
24	7.0 ± 0.6	1.2 ± 0.4	17

^a Standard error of the mean.

are known to respond almost instantaneously to changes in hydrostatic pressure in the water supply of leaves (Raschke, 1970).

Although stomatal resistances and rates of photosynthesis were not measured on the same leaves, repeated experiments showed the stomatal response to occur earlier than the effect on CO₂ incorporation. The influence of varying CO₂ concentrations on the ABA-induced effect on photosynthesis and stomatal resistance has not been investigated but, because of the differences in time of response, it seem unlikely that the ABA effect on wheat leaf stomata is an immediate result of an effect on CO₂ concentration in the intercellular spaces and in the guard cells. It would seem, rather, that the ABA-induced increase in stomatal resistance is the cause of the decrease in rates of photosynthesis in treated leaves.

It has been shown that stomatal opening in tobacco leaves is correlated with the potassium-ion concentration in guard cells, and it is considered that an active ion-pump mechanism necessitating ATP synthesis is involved (Sawhney and Zelitch, 1969). Again the effect of ABA is unknown and warrants further investigation. Of interest is the recent report by Tillberg (1970) that the ATP level in the algae *Scenedesmus* is increased by ABA. The rapidity of ABA action on stomatal resistance might indicate a mechanism involving a change in the activity of an ion pump or in membrane permeability but this can only be conjecture at this stage.

It has been suggested previously that hormones may be involved in the regulation of photosynthetic rates (Turner and Bidwell, 1965; Wareing *et al.*, 1968). On the basis of the above findings and also those of Imber and Tal (1970), a detailed investigation into the role of endo-

genous ABA in the regulation of stomatal apertures and rates of photosynthesis is warranted.

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References

- Coulombe, L.-J., Paquin, R.: Effets de l'acide gibbérellique sur le métabolisme des plantes. *Canad. J. Bot.* **37**, 897-901 (1959).
- Imber, D., Tal, M.: Phenotypic reversion of *flacca*, a wilted mutant of tomato, by abscisic acid. *Science* **169**, 592-593 (1970).
- Kanemasu, E. T., Thurtell, G. W., Tanner, C. B.: Design, calibration and field use of a stomatal diffusion porometer. *Plant Physiol.* **44**, 881-885 (1969).
- Little, C. H. A., Eidt, D. C.: Effect of abscisic acid on budbreak and transpiration in woody species. *Nature (Lond.)* **220**, 498-499 (1968).
- Meidner, H.: The effect of kinetin on stomatal opening and the rate of intake of CO₂ in mature primary leaves of barley. *J. exp. Bot.* **18**, 556-561 (1967).
- "Rate-limiting" resistances and photosynthesis. *Nature (Lond.)* **222**, 876-877 (1969).
- Mittelheuser, C. J., Steveninck, R. F. M., Van: Stomatal closure and inhibition of transpiration induced by (RS)-abscisic acid. *Nature (Lond.)* **221**, 281-282 (1969).
- Raschke, K.: Stomatal responses to pressure changes and interruptions in the water supply of detached leaves of *Zea mays* L. *Plant Physiol.* **45**, 415-423 (1970).
- Sawhney, B. L., Zelitch, I.: Direct determination of potassium ion accumulation in guard cells in relation to stomatal opening in light. *Plant Physiol.* **44**, 1350-1354 (1969).
- Shimshi, D.: A rapid field method for measuring photosynthesis with labelled carbon dioxide. *J. exp. Bot.* **20**, 381-401 (1969).
- Tillberg, J.-E.: Effects of abscisic acid, salicylic acid and trans-cinnamic acid on phosphate uptake, ATP-level and oxygen evolution in *Scenedesmus*. *Physiol. Plantarum (Cph.)* **23**, 647-653 (1970).
- Turner, W. B., Bidwell, R. G. S.: Rates of photosynthesis in attached and detached bean leaves, and the effect of spraying with indoleacetic acid solution. *Plant Physiol.* **40**, 446-451 (1965).
- Wareing, P. F., Khalifa, M. M., Treharne, K. J.: Rate-limiting processes in photosynthesis at saturating light intensities. *Nature (Lond.)* **220**, 453-457 (1968).

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