

**Short Communication****Kinetin Reversal of NaCl Effects**

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*Department of Biology, Ben Gurion University of the Negev, Beer Sheva, Israel***ABSTRACT**

Leaf discs of *Nicotiana rustica* L. were floated on NaCl in the presence of kinetin or abscisic acid. On the 5th day  $^{14}\text{CO}_2$  fixation, [ $^3\text{H}$ ]leucine incorporation, stomatal conductance, and chlorophyll content were determined. Kinetin either partially or completely reversed the inhibitory effects of NaCl while ABA had no effect.

The endogenous levels of cytokinins (CK) and abscisic acid (ABA) are altered in response to water stress whether imposed by drought, salinity, or high transpirational demand. It is possible that these changes play a regulative role and are an essential part of the metabolic and developmental processes involved in the adaptation of plants to water stress (5). Attempts to prove this by reversal of the stress responses with kinetin application have so far failed (8–10). This failure is probably the result of the decrease in stomatal resistance due to kinetin application thereby further enhancing the stress. When kinetin was applied to tissue excised from stressed plants, it reversed stress effects by retarding Chl degradation (5) and enhancing protein synthesis (3, 7). Further, ABA treatment of stressed plants reduced RNAase activity enhanced by water stress (1). It is not clear if this is a direct metabolic effect of ABA or rather a result of a shift in water balance of the plants due to stomatal closure and/or reduction in root resistance. The outcome of experiments in which hormonal balance is manipulated could support the hypothesis that the endogenous changes are regulating the metabolic response directly during water stress. In this work, such an attempt was made by treating excised tobacco leaf discs floating on NaCl solution with either kinetin or ABA.

**MATERIALS AND METHODS**

Leaf discs, 15 mm in diameter, from fully expanded leaves of tobacco (*Nicotiana rustica* L.) were floated on a 0.1% NaCl (w/v) solution for 5 days. Control discs were incubated in water. Kinetin or ABA was added to make 1  $\mu\text{g}/\text{ml}$  solutions. On the 5th day, the discs were recut to 12-mm diameter. Four separate evaluations of treatment effects were made: (a) Chl content leaf discs were placed one each in 4 ml of dimethylformamide in the dark. After 2 days at 4 C the optical density of the solution was determined at 665 nm. (b) [ $^3\text{H}$ ]Leucine incorporation into proteins, and (c)  $^{14}\text{CO}_2$  fixation in light were evaluated as described previously (2, 3); and (d) stomatal conductance was determined with a viscous flow porometer (designed and contracted by D. Shimshi).

**RESULTS AND DISCUSSION**

The results are given in Table I and indicate that the response of discs floating on NaCl solution is similar to the known response

Table I. Effects of NaCl, ABA and kinetin on  $^{14}\text{CO}_2$  fixation,  $^3\text{H}$  leucine incorporation, stomatal conductance, and chlorophyll content in tobacco leaf discs. Responses are presented as percentages of the water control values. In each treatment 8 discs were used.

Response	$^{14}\text{CO}_2$ Fixation	$^3\text{H}$ leucine incorporation	Stomatal conductance	Chlorophyll content
No. of Experiments	3	3	1	3
Absolute value control	45,288 cpm/disc	14,822 cpm/disc	1.67 $\text{cm}^2/\text{s}$	0.64 O.D./disc
Control	100.0	100.0	100.0	100
Kinetin	133.6	86.9	111.0	114
ABA	84.3	90.6	41.0	99
NaCl	53.5	47.3	47.7	75
NaCl + Kinetin	105.2	78.3	59.8	90
NaCl + ABA	48.0	41.2	45.0	74

of leaves from water-stressed plants (4). Kinetin applied simultaneously with NaCl to those discs altered the stress response. The four processes studied were differentially affected by salinity and the extent of the kinetin reversal was also different. ABA application, on the other hand, manifested the stress conditions. This is in accordance with the assigned role of ABA in the adaptation process to environmental stress. Comparing the changes in  $^{14}\text{CO}_2$  fixation and stomatal conductance reveals that the latter cannot account by itself for the increased capacity of kinetin-treated stressed leaf discs, to fix  $^{14}\text{CO}_2$ . In this preliminary experiment, no attempt was made to optimize the kinetin concentration, an approach which might result in additional relevant information. The enhancement of three processes by kinetin in the absence of stress condition points toward a possibility that the effect of kinetin on stressed leaf discs is not specific for the stress situation. Careful comparison will reveal, however, that stressed tissue responded to kinetin more than did unstressed tissue. This together with the reported reduction in cytokinins during stress (6) point toward the possibility that kinetin indeed is a limiting factor under stress conditions and plays a role in plant responses to them. Similarly it can be argued that the effect of ABA treatment to stressed tissue is negligible due to the rise of the endogenous level of ABA, hence no reversal of the response to water stress due to ABA treatment is evident.

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