

EFFECT OF ABSCISIC ACID ON THE EXUDATION PROCESS IN EXCISED ROOTS OF MAIZE, MILLET AND SORGHUM

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Abstract

The effect of abscisic acid (ABA) on the exudation process in excised roots of maize (*Zea mays* L.), pearl millet (*Pennisetum typhoideum* Rich.) and Sorghum (*Sorghum vulgare* Pers.) was investigated. ABA supplied to the growth as well as the bathing media stimulated volume flow (Jv) and to a lesser extent the potassium flux (Jk) from all three species. The major factor causing stimulation was an ABA induced increase in the hydraulic conductivity of the root (Lp).

Introduction

Abscisic acid has been shown to interact with the processes of ion transport in stomatal guard cells (Mansfield & Jones, 1971; Horton & Moran, 1972; Raschke, 1977, MacRobbie, 1981). But the action of ABA is not confined to leaves, it also acts on the ion transport in storage tissues and roots (Van Steveninck, 1972; Cram & Pitman, 1972). However, the data regarding effect of ABA on ion and water transport across the roots, reported by different workers, appears to be conflicting. Ion and water transport is stimulated in some cases (Collins & Kerrigan, 1974; Glinka, 1977, 1980) while on the other hand it is inhibited by ABA (Cram & Pitman, 1972; Pitman *et al.* 1974). The inhibiting action of ABA is found to be dependent on the type of plant species and culture conditions in which plants are grown (Pitman *et al.* 1974). Thus the effect of ABA on ion and water transport from excised roots of three crop species was investigated to attempt to resolve these differences.

Materials and Methods

Dark grown plants: Seeds of maize (*Zea mays* L. Cv Anjou 210) were soaked in tap water for about 1–2h and then were set to germinate on moist tissue at 25°C in the

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dark for a period of 48 hours. After germination these seeds were transferred to plastic containers containing 1/10 Long Ashton culture solution and were grown for a period of 72 hours. To facilitate root growth, the culture solution was aerated continuously. The seedlings were then removed from the culture solution for exudation experiments. Measurements of exudate rate and exudate ion concentration were carried out as described by Collins & Kerrigan (1974).

Light grown plants: After germination in the dark the seedlings of pearl millet (*Pennisetum typhoideum* Rich) and sorghum (*Sorghum vulgare* Pers.) were transferred to a growth chamber and maintained under continuous illumination. Illumination was from fluorescent tubes and incandescent lamps with a ratio of 3:1 providing a total light intensity of about 15,000 to 16,000 lux at the surface of the medium. The temperature was $22 \pm 1^\circ\text{C}$ and the relative humidity was 65-75%. Growth medium and experimental condition for exudation studies were identical to those for dark grown plants. For millet and sorghum plants, whole root systems were used in exudation studies.

Stock solution of ABA (10^{-2}M) was prepared by dissolving in dimethyl sulphoxide (DMSO), the final concentration of ABA used was 10^{-6}M in all experiments. DMSO was added to control roots at the same v/v percentage. The ABA used was 2-cis-4-trans abscisic acid, which is a synthetic crystalline product and is approximately 95% pure supplied by Sigma Chemical Company U.S.A.

Ion analyses: Ion analyses were made on a Unicam SP 90A Series 2 Atomic Absorption Spectrometer. K^+ and Na^+ were measured by emission spectrometry and Ca^+ and Mg^+ by absorption spectrometry.

Ion fluxes into the exudate: The net flux of ions across the root from outside to inside was estimated using the following:

$$J_s = J_v \cdot C_s^x \quad \text{equation 1.}$$

where,

J_s , is the net flux of ion "s" into the xylem exudate in $\text{mol m}^{-2}\text{s}^{-1}$.

J_v , is the water flow (strictly volume flow) in $\text{m}^3\text{m}^{-2}\text{s}^{-1}$.

C_s^x , is the concentration of ion "s" into the xylem exudate in mol m^{-3} .

Hydraulic conductivity (L_p): The values for L_p were calculated by using the equation 2. (House & Findlay, 1966).

$$J_v = L_p \sigma RT (C_s^x - C_s^o) + \phi_w^o \quad \text{equation 2.}$$

where,

L_p , is the hydraulic conductivity of the root in $\text{m s}^{-1} \text{Pa}^{-1}$

σ' is the reflection coefficient of solute dimensionless (assumed to be unity)

R , is the gas constant $\text{m}^3 \text{Pa mol}^{-1} \text{K}^{-1}$

T , is the temperature in degree absolute K.

C_s^x and C_s^o are solute concentrations in mol m^{-3}

(x refer to the xylem and o to the outside medium).

ϕ_w^o , is the non-osmotic water flow $\text{m}^3 \text{m}^{-2} \text{s}^{-1}$.

Many of the above units are inconveniently large when applied to the exudation process. Accordingly the following units, which arise naturally in the generation of data have been used, unless stated otherwise.

J_v in $\mu\text{l cm}^{-2} \text{h}^{-1}$	(= $2.8 \times 10^{-9} \text{m}^{-3} \text{m}^{-2} \text{s}^{-1}$).
C_s^x and C_s^o mM	(mM = Mol m^{-3}).
J_s in $\text{nmol cm}^{-2} \text{h}^{-1}$	(= $2.8 \times 10^{-9} \text{m}^{-2} \text{s}^{-1}$).
L_p in $\text{cms}^{-1} \text{Pa}^{-1}$	(= 10^{-2}m s^{-1}).
ϕ_w^o has been set to zero.	

Results and Discussion

For most plant species, ABA has been shown to inhibit both shoot growth (Quarrie & Jones, 1977) and root elongation (Phillipson & Coutts, 1979). Such responses however, did not appear in these experiments which may be due to the different concentrations of ABA used (Pilet & Rivier, 1980), the duration of treatment (Chanson & Pilet, 1981) or the lack of penetration of ABA into the plant (Watt *et al.* 1981).

ABA stimulated J_v in all three species (Table 1), but to different degrees, stimulation was most marked in millet and maize (Fig. 1). These differences could be explained by a number of factors; endogenous hormone levels in the plants, growth conditions, nature of root system used (whether excised root or whole root system), and species and cultivar variations (Glass & Perley, 1980). It is also noteworthy that stimulation of J_v in sorghum was much slower than in the other two species (Fig. 1).

The stimulation of J_v by ABA supplied to the growth medium reported here agrees well with the report of Collins & Morgan (1980) where it was found that when plants were grown in ABA the influence of ABA on J_v was more pronounced. The concentration of K^+ in the exudate was not significantly altered by ABA treatment in any of the three species (Table 1), this is in agreement with the previous findings for maize (Collins & Kerrigan, 1974).

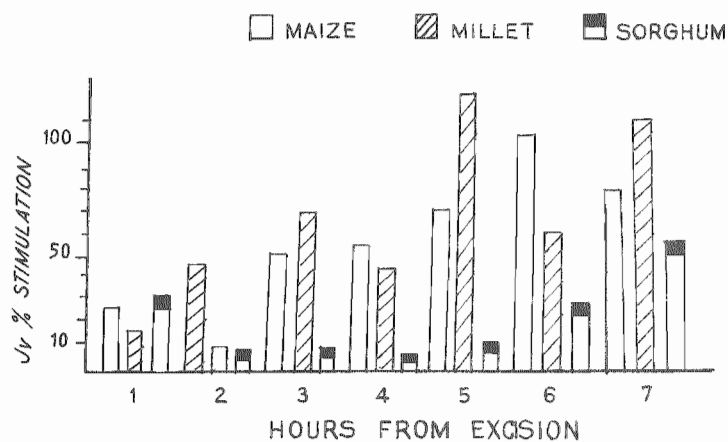


Fig. 1. Percentage stimulation of Jv by ABA.

Table 1. Effect of ABA, supplied to the growth medium on Jv and Ion concentration into the exudate

	Control			ABA 10 ⁻⁶ M		
	Maize	Millet*	Sorghum*	Maize	Millet	Sorghum
Jv	2.87 ± 0.24	127.2 ± 2.22	205.2 ± 1.12	4.60 ± 0.70	186.6 ± 1.56	235.5 ± 2.50
K ⁺	14.4 ± 0.5	39.1 ± 2.45	24.2 ± 1.02	12.4 ± 0.7	34.5 ± 1.6	23.5 ± 1.0
Na ⁺	0.59 ± 0.01	0.85 ± 0.05	1.29 ± 0.81	0.75 ± 0.06	0.95 ± 0.03	0.82 ± 0.05
Ca ⁺⁺	1.10 ± 0.06	1.04 ± 0.20	1.47 ± 0.83	0.89 ± 0.06	0.47 ± 0.02	0.84 ± 0.05
Mg ⁺⁺	0.88 ± 0.07	1.80 ± 0.81	0.87 ± 0.81	0.77 ± 0.03	0.53 ± 0.02	0.99 ± 0.06

* Jv $\mu\text{l (gfw)}^{-1} \text{h}^{-1}$; Jv mean of 20 roots. Ion concentration (mM) mean of 5 samples.

* Plants grown in the light. Light intensity 15,000 to 16,000 lux. Temperature 22±1°C. Experimental temperature 25°C; Culture and bathing medium 1/10 Long Ashton solution. All values ± st. error of the mean. ABA has been supplied in both the growth medium and the bathing medium.

These data using equation 1 and 2, can be used to estimate values for the hydraulic conductivity (L_p) and potassium flux (J_k) of the roots (Table 2). It can be seen that in both millet and maize there is a pronounced increase in L_p in the presence of ABA, and to a lesser extent in sorghum. These observations point that even though maize was grown in dark it did not behave differently. It should be stressed though that this analysis assumes that a potassium salt is the major osmotic component in the exudate, obviously this needs to be checked in subsequent work.

Table 2. Effect of ABA, supplied to the growth medium on Hydraulic conductivity (L_p) and Potassium Flux (J_k).

	J_k ($\text{nmol cm}^{-2} \text{ h}^{-1}$)		L_p ($\text{cm s}^{-1} \text{ Pa}^{-1}$) $\times 10^{12}$		J_k ($\mu\text{mol g}^{-1} \text{ h}^{-1}$)		L_p ($\mu\text{l g}^{-1} \text{ h}^{-1}/\text{bar}$) $\times 10^{-2}$	
	Maize		Millet	Sorghum	Millet	Sorghum		
Control	41.0	20.0	4.97	4.97	0.60	1.50		
ABA 10^{-6}M	57.0	32.3	6.44	5.53	1.10	1.85		

Pitman *et al.* (1974) reported that ABA can inhibit or stimulate water permeability and ion transport depending on temperature and nutrient status of the plant. However, in a detailed study it was found that the stimulatory effect of ABA on J_v and L_p occurred over a wide temperature range and at several KCl/NaCl concentrations in the medium (Channa, 1982). Earlier, increased J_v and J_s due to ABA as a function of different salt concentrations in the medium (Karmoker & Van Steveninck, 1978) and temperature (Collins & Morgan, 1980) have also been reported for bean and maize roots. The contradictions in the published reports (Table 3) might be due to the varying concentrations of ABA used by various workers. At concentrations above 10^{-5}M ABA may become inhibitory, markedly so at 10^{-4}M in maize (Collins & Channa, 1983).

In this preliminary work we have shown that ABA appear to act on both millet and sorghum in the same way as it does on maize roots. Thus ABA has been shown to play a role in the wilting response of millet (Henson & Quarrie, 1981), and it may be that an alteration of the root permeability to water, induced by endogenous ABA, is a cause of this response.

Table 3. Reported effects of ABA on ion and water transport of various plant roots.

Material	Temp. °C	Conc. ABA (M)	Reported effect	Authority
Excised barley and maize roots	25	10 ⁻⁶	Inhibition of Jv and Js	Cram & Pitman (1972)
Excised maize roots	25	10 ⁻⁶	Stimulation of Lp, Jv and Js	Collins & Kerrigan (1974)
Excised barley roots	25	2,10,50x10 ⁻⁶	Strong reduction in Jv and Js	Pitman <i>et al.</i> (1974)
Sunflower root system	25	4x10 ⁻⁶	Stimulated Jv	Glinka (1977)
Excised barley roots	22	5x10 ⁻⁶	Inhibition of Na transport	Behl & Jeschke (1979)
Excised maize roots	25	10 ⁻⁶	Stimulation of Lp, Js and activation energy of water permeability	Collins & Morgan (1980)
Excised sunflower root system	25	10 ⁻⁶	Stimulation of Lp and Js	Glinka (1980)
Excised maize roots	20–25	10 ⁻¹⁰ , 10 ⁻⁹ , 10 ⁻⁸ , 10 ⁻⁷ and 10 ⁻⁶	Stimulated Jv and Lp	Collins & Channa (1983)

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