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SHORT COMMUNICATION

THE EFFECT OF SALINE MEDIA OF VARYING IONIC COMPOSITION ON STOMATAL RESISTANCE OF RICE *(Oryza sativa L.)* LEAVES

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Introduction

Increased stomatal resistance due to water and salt stress reduce transpiration.[1-4] The sensitivity of stomata to decreased leaf water potentials varies between species and is influenced by age, growth condition of the plant[3,5,6] and the age of the leaf.[7] Accumulation of abscisic acid (ABA) as a result of water and salt stress is the cause of stomatal closure.[2,3,5,8] Although salt stress may lead to stomatal closure, and therefore, to decreased transpiration, if salt is allowed to enter guard cells, it may reverse the osmotic gradient leading to stomatal opening and therefore increased transpiration.[9] Earlier studies, especially on rice, have generally used single salts to create saline environments. When salt mixtures were used, their composition did not mimic natural salinity types.[11-12] The objective of this study was to observe stomatal responses of rice leaves to salt mixtures with compositions similar to those in saline soils.

Materials and Methods

Plant material and seed germination

A salt sensitive variety, IR5929-12-3 (IR5929) (acc.no. 57016) and a salt tolerant variety, Pokkali (acc. no. 26869) of rice *(Oryza sativa* L.) were used. Seeds were surface sterilized by immersion for one minute with 0.1% mercuric chloride solution, and then washed with several changes of demineralized water. The seeds were soaked for 24 hours in demineralized water, and then spread on a moist filter paper in Petri dishes for germination.

Plant growth and salinization

Three days later seedlings were transferred to 3 *l* plastic pots filled with culture solution[10] (Table 1) and placed in a greenhouse. Seedling density was three per pot. The medium was salinized with one of three types of ionic composition, Chloride Sulphate (CS) (with sulphate dominance), Sulphate Chloride (SC) (with Chloride dominance) and Chloride (C) when the seedlings were two weeks old. These three compositions represented natural saline soil types.[11,12] The three types of ionic composition were prepared using the ionic ratios given by Strogonov (1964)[12] (Table 2).

Table 1: Composition of the culture solution.

Compound	mmol/liter	Compound	μmol/liter
$CaCl_2$	2	FeEDTA	100
$MgSO_4$	2	H_3PO_3	25
KCl	2	$MnSO_4$	5
K_2SO_4	1	$ZnSO_4$	2
NH_4NO_3	2	$CuSO_4$	0.5
$NH_4H_2PO_4$	1	$(NH_4)_6.Mo_7O_{24}$	0.1

In each type four different levels of salinity (expressed as electrical conductance) 2, 4, 6 and 8 dS/m were used. Electrical conductance of the control (growth medium alone) varied from 1.0 to 1.1 dS/m. Electrical conductance = 0 indicates the non-salinized control.

Measurement of stomatal resistance

Fifty days after salinization, stomatal resistance was measured on the abaxial epidermis of the first recently matured leaf of the main tiller using LI-700 Transien Porometer.

Statistical analysis

Each value reported for stomatal resistance was the mean of 9 plants. A test of significance for differences between the treatments was provided by Tukey multiple range test at P = 0.05. Coefficient of variation was 10%.

Results

The stomatal resistance of the leaves of Pokkali plants grown in SC and CS media increased with increasing salinity. However, in C medium stomatal resistance increased only up to 2 dS/m and then declined (Table 3).

The stomatal resistance of salt sensitive IR5929 plants grown in CS medium increased up to 6 dS/m and then declined at a salinity of 8 dS/m. In the other 2 saline media stomatal resistance increased only up to 4 dS/m and then declined. IR 5929 plants grown in C medium at 8 dS/m salinity however did not survive (Table 3).

Table 2. Ion composition ($g\ dm^{-3}$) of the salt mixtures

Ionic composition	Electrical Conductance ($dS\ m^{-1}$)			
	2	4	6	8
Chloride (C)				
Na^+	0.295	0.688	1.081	1.495
K^+	0.008	0.020	0.031	0.043
Mg^{2+}	0.035	0.083	0.131	0.181
Ca^{2+}	0.010	0.025	0.040	0.055
SO_4^{2-}	0.054	0.165	0.260	0.360
Cl^-	0.532	1.242	1.951	2.697
HCO_3^-	0.004	0.013	0.021	0.030
Sulphate-chloride (SC)				
Na^+	0.235	0.540	0.864	1.172
K^+	0.009	0.027	0.045	0.064
Mg^{2+}	0.061	0.147	0.235	0.319
Ca^{2+}	0.011	0.026	0.047	0.064
SO_4^{2-}	0.250	0.583	0.930	1.264
Cl^-	0.392	0.912	1.461	1.981
Chloride-sulphate (CS)				
Na^+	0.090	0.211	0.331	0.445
K^+	0.011	0.029	0.045	0.062
Mg^{2+}	0.144	0.334	0.52	0.704
Ca^{2+}	0.005	0.014	0.02	0.031
SO_4^{2-}	0.701	1.617	2.532	3.401
Cl^-	0.070	0.161	0.255	0.342

Discussion

Water stress stimulates ABA accumulation which leads to stomatal closure.[2-4] Mizrahi *et al.*[8] and Tal[13] report that salinity promotes the synthesis of ABA. Therefore, the increasing stomatal resistance with increasing salinity observed in the current study might be due to the accumulation of ABA.

Table 3 : Effect of varying the ionic composition on stomatal resistance of rice

Salinity type	EC ds m^{-1}	Stomatal resistance (s cm^{-1})	
		Pokkali	IR5929
Control	0	1.76i	1.58e
CS	2	1.88gh	1.92bc
	4	2.00ef	1.98ab
	6	2.02e	2.09a
	8	2.16d	1.92bc
SC	2	2.05e	1.91bcd
	4	2.26c	1.96
	6	2.50b	1.83cd
	8	2.71a	1.80d
C	2	2.10de	1.68e
	4	1.90fg	1.69e
	6	1.80hi	1.59e
	8	1.84ghi	-

Values within a column followed by the same letter are not significantly different.

Decreased stomatal resistance at high salinity (Table 2) was also observed in the current study. This may be due to salt entering guard cells, at high salinity (6,8 ds/m) and reversing the osmotic gradient resulting in to stomatal opening as suggested by Sen and Chawan.[9]

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