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Paclobutrazol as an Aid to Reduce some Effects of Salt Stress in Oleander Seedlings

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Summary

Nerium oleander seedlings were irrigated with untreated locally available water and 70 mM salt water in greenhouse conditions to study the effect of the drench application of 30 mg paclobutrazol (PBZ) per plant on the salt stress response. PBZ improved the development of oleander in the saline irrigation condition by reducing leaf salt symptoms, reducing defoliation and limiting, in relative terms, the reduction of root dry weight and leaf area that normally results from salinity. However, the retardant effects of salinity and PBZ on development were additive, and the joint application produced a substantial reduction in plant development. Although salinity reduced the relative chlorophyll content of leaves, PBZ maintained it in the same conditions. PBZ application reduced the Na^+ content in stem and roots, and Cl^- content in leaves, stem, and roots. In the saline treatment, the salt content of the leaching water increased when plants were treated with PBZ, because it reduced the availability of saline ions in the medium. These findings suggest that PBZ reduced saline stress symptoms in stressed oleander plants by reducing the uptake and accumulation of harmful Na^+ and Cl^- ions by plant tissues. Salinity reduced both the relative water content and water potential, but maintained turgor potential in PBZ-treated and non-treated plants as a consequence of osmotic adjustment. In saline conditions, PBZ also promoted a process of osmotic adjustment, probably through the accumulation of organic osmotic compounds.

Zusammenfassung

Paclobutrazol als Hilfsmittel zur Reduzierung von Salzstressreaktionen bei Oleandersämlingen. *Nerium oleander* Sämlinge wurden entweder mit unbehandeltem, lokal verfügbarem Wasser oder mit 70 mM Salzwasser unter Gewächshausbedingungen bewässert, um den Effekt einer Tropfbewässerung mit 30 mg Paclobutrazol (PBZ) pro Pflanze auf die Salzstressreaktion zu untersuchen. PBZ verbesserte die Entwicklung von Oleander unter Salzwasserbedingungen durch eine Verminderung der Salzsymptome an Blättern, einer Reduktion des Laubverlusts sowie eine relative Begrenzung der Verminderung des Wurzeltrockengewichts und der Blattfläche, welche normalerweise bei hohem Salzgehalt auftritt. Dennoch wirkten die Salz- und PBZ bedingten Hemmungseffekte additiv, so dass insgesamt eine beträchtliche Reduktion der Pflanzenentwicklung resultierte. Obwohl der erhöhte Salzgehalt den relativen Chlorophyllgehalt der Blätter herabsetzte, wurde dieser durch die PBZ-Behandlung auf gleichem Niveau gehalten. Die PBZ-Behandlung reduzierte den Na^+-Gehalt in Stamm und Wurzeln und den Cl^--Gehalt in Blättern, Stamm und Wurzeln. Im Falle der Salzbewässerung erhöhte sich der Salzgehalt des Dränagewassers in PBZ behandelten Pflanzen auf Grund einer Herabsetzung der Ionenverfügbarkeit im Medium. Unsere Beobachtungen deuten darauf hin, dass PBZ die Salzstresssymptome in gestressten Oleanderpflanzen durch eine Reduktion der Aufnahme und Akkumulation von schädigenden Na^+ und Cl^- Ionen durch das Pflanzengewebe vermindert. Der erhöhte Salzgehalt reduzierte sowohl den relativen Wassergehalt als auch das Wasserpotential, während das Turgorpotential in PBZ behandelten und -unbehandelten Pflanzen als Folge eines osmotischen Ausgleichs konstant blieb. Unter Salzwasserbedingungen förderte PBZ auch den Prozess des osmotischen Ausgleichs, möglicherweise durch die Akkumulation von organischen osmotischen Bestandteilen.

Key words. cultar – native species – nursery – plant growth retardant – salinity

Introduction

Nerium oleander L. is a native plant of the Mediterranean area that is widely used for revegetation and landscaping purposes because of its ornamental value and its capacity to adapt to adverse environmental conditions. In the nursery production of plants in salt affected regions, such as SE Spain, the use of saline waters is fre-

quent, which may result in growth and development problems in the cultivated species. The most common plant salinity symptoms are leaf chlorosis and necrosis, a reduction in growth and defoliation, symptoms which are the consequence of the physiological and biochemical effects induced by the salts. Although extensive information exists describing these effects, the effect that saline stress has on vegetal development is mainly due to two factors: osmotic and ionic. Today, the most common practice for alleviating salt stress is to increase leaching. However, because in arid or semiarid areas leaching is an expense option, one possible short term solution may be the application of plant growth retardants, since some studies have demonstrated their usefulness in alleviating environmental stresses. SAHA and GUPTA (1998) found that PBZ partially reduced the saline effect induced by the accumulation of sodium and chloride in *Phaseolus vulgaris* plants. SHEN et al. (1996) concluded that PBZ diminished the saline stress and promoted salinity tolerance in *Gossypium* sp. seedlings. EL-KHASHAB et al. (1997) suggested that PBZ promoted mechanisms of saline stress avoidance in *Prunus persica* and EL-KHEIR et al. (2000) indicated that PBZ increased the quality and yield of *Helianthus annuus* seeds under saline conditions. Furthermore PBZ reduced some negative effects of salinity on root development in several citrus rootstocks (EL-DESOUKY and ATAWIA 1998). Based on these suggestions, in the present experiment the ability of PBZ to improve the response of *Nerium oleander* seedlings to salt stress and its effect on salt resistance mechanisms were studied.

Materials and Methods

Plant material and cultivation conditions

Nerium oleander L. (native of Región de Murcia, SE Spain) seedlings were used. The most homogeneous selection of seedlings possible (16–18 cm in height) was made avoiding, to the greatest extent possible, variations resulting from sexual production. Seedlings were grown in pots 20 cm high and measuring 15x15 cm at the top and 11x11 cm at the base. The pots were filled with a substrate of black peat, perlite and a clay-loam soil (1:1:2 volume) and placed in a greenhouse (100 m^2 area, 3.5 m height at the side and 5.5 at the centre) in the Campo de Cartagena (lat. 37 °45 'N; long. 0 °59 'W). Transplantation was made on 15 May 2001 and all plants were irrigated (one day per week and leaching 15 % of the applied water) until the application of the chemical and saline treatments. Weather conditions during screen culture were 8.1 °C (minimum), 25.2 °C (average) and 42.9 °C (maximum); minimum relative humidity was 22.2 %, and the maximum 100 %, with a mean of 67.3 %.

PBZ and saline treatments

30 mg of PBZ (Cultar 25 %) were applied to the substrate surface as a single 45 ml liquid drench on the morning of 4 June 2001, two hours after the last watering. Untreated plants acted as control. The saline treatments began on 15 June 2001, and consisted on the irrigation of the half of the PBZ treated and untreated plants with local water, and the other half with a solution of 70 mM of NaCl. These produced four different treatments: PBZ-treated plants irrigated with local water; PBZ-treated plants irrigated with 70 mM of NaCl; PBZ-untreated plants irrigated with local water and PBZ-untreated plants irrigated with 70 mM of NaCl. The irrigation volume was the same for all treatments, 600 ml per plant and week. Local irrigation water (pH 7.66; electric conductivity 1 dS/m) was used in plants as a saline control. The saline treatments continued until 20 November 2001 (end of the experimental period). During this period, plants were leached with local water after every fourth irrigation to prevent excessive salt accumulation.

Parameter measurements

The following were recorded at the end of the experimental period:

1. Shoot and root development [plant height, leaf area, aerial dry weight, root dry weight, root/shoot ratio, relative chlorophyll content (RCC)],
2. leaf abscission, presence of phytotoxicity symptoms and the percentage of dead plants,
3. sodium and chloride content of tissues,
4. average percentage of leachate and salt totals of the leaching water,
5. leaf water status [leaf water potential (Ψ_l), leaf osmotic potential (Ψ_s), leaf osmotic potential at full turgor (Ψ_{os}), leaf turgor potential (Ψ_p), relative water content (RWC), stomatal conductance (g_s)].

Soil was gently washed from roots, and the plants were divided into leaves, stems and roots. Leaves, stems and roots were oven dried at 70 °C until they reached a constant weight. Total leaf area was measured using a Delta-T leaf area meter (Device Ltd., Cambridge, UK). Abscised leaves were collected and their area measured as they fell during the growth period and the cumulative area of abscised leaves per plant was recorded. RCC was measured at the midpoint of six mature leaves per plant with a Minolta SPAD-502 chlorophyll meter.

Six plants per treatment were harvested and separated into leaves, stems and roots. These were washed with distilled water, dried at 70 °C and stored at room temperature for inorganic solute analysis. Cl^- content was analysed by a chloride analyser (Chloride analyser mod. 926, Sherwood Scientific Ltd.) on the aqueous extracts obtained after mixing 100 mg dried and powdered plant material with 40 ml water, and stirring for 30 minutes before filtering. The sodium content was determined in a digestion extract with $HNO_3/HClO_4$ (2:1, v/v) by atomic absorption spectrometry (Shimadzu AA-6701, Shimadzu Co.).

Ψ_l, Ψ_s and Ψ_{os} were measured at midday in six plants of each treatment. Ψ_l was determined by enclosing the leaves in a plastic bag (TURNER 1988), which was immediately placed in a pressure chamber (Soil Moisture Equipment Co; Santa Barbara, CA, USA) according to SCHOLANDER et al. (1965), and raising the pressure at a rate of 0.03 Mpa s^{-1} using nitrogen gas. Ψ_s was estimated using a vapour pressure osmometer (Wescor 5500, Wescor Inc., Logan, Ut, Usa) in excised leaves harvested at midday and immediately frozen and stored at –30 °C. Before the measurements, samples

were thawed and leaf sap was extracted for immediate Ψ_s determination, according to GUCCI et al. (1991). The leaves used for determining Ψ_{os} were placed in plastic bags and allowed to reach full saturation by dipping the petioles in distilled water overnight. After that, leaves were dried by filter paper to eliminate surface water, wrapped in aluminum foil and immersed in liquid nitrogen for analysis by the method described above. Ψ_p was estimated as the difference between Ψ_l and Ψ_s. Leaf conductance was determined in the same number of samples as used to determine water potential measurements, using a Steady-state porometer (LI-1600, Li-cor Inc., Lincoln, NE, USA) based on measuring the flow of dry air necessary to balance the water transpired by the leaf in the measuring tray. Measurements were made at midday (the midpoint of the light period). RWC was calculated as: (fresh weight – dry weight)/(saturated weight – dry weight) x 100.

Statistical analysis

The two factors and its interaction were analysed by two-way ANOVA using Statgraphics 2.1 Plus for Windows. Treatment means were separated with Duncan´s Multiple Range Test ($P \leq 0.05$). Thirty randomly distributed plants per treatment were considered.

Results and Discussion

Salinity reduced the dry weight of the aerial part to a statistically significant degree (52 % of the corresponding control, see Table 1). The effect of salinity on biomass is well documented and the level of growth has been used in several studies to measure resistance to saline conditions (SÁNCHEZ-BLANCO et al. 1991). PBZ significantly reduced the aerial dry weight of the control by 32 %. However, the interaction between salinity and PBZ was not significant, meaning that the reduction in the aerial dry weight of the control caused by salinity was similar in plants treated with PBZ and those not so treated. This suggests that the PBZ did not diminish the tendency of salinity to reduce the aerial dry weight. Similarly, the root dry weight was reduced by salinity and PBZ (Table 1). However, the reduction of root dry weight by salinity was more intense than in the aerial part, while the opposite was true for PBZ. The ANOVA showed that the interaction salinity-PBZ was statistically significant in the case of root dry weight (Table 1). Salinity had a lower effect on PBZ-treated plants than on non-treated plants (Fig. 1A), suggesting that PBZ improves the resistance of roots to salinity. Both factors studied and their interaction had a highly significant effect on the root/shoot ratio (Table 1). While salinity reduced this ratio as a consequence of the greater sensitivity of roots to salt, PBZ increased it since the aerial part was more sensitive to this retardant than the roots. Analysis of their interaction (Fig. 1B) suggests that PBZ attenuates the fall in this ratio produced by salinity, enabling treated plants to minimise the loss of water through transpiration (ALARCÓN et al. 1999).

Salt stress tolerance can also determine the height of plants (CHEESEMAN 1998). In our experiment plant height was significantly inhibited by salinity (reaching 28 % of the height of control plants) and by PBZ (24 % of control) (Table 1). BAÑÓN et al. (2003) and EL-KHASHAB et al. (1997) observed a similar response in *Rhamnus alaternus* and *Prunus persica*, respectively. The absence of any interaction for this parameter suggests that the reduction caused by salinity was similar in both untreated and PBZ-treated plants, revealing the lack of effectiveness of the retardant in counterbalancing the reduction in height caused by salinity. Furthermore, the effects of salinity and PBZ on plant height were cumulative since there was a reduction from 40.3 cm (plants not treated by salinity or PBZ) to 21.6 cm when the plants were treated by both (Fig. 1H). However, this reduction in height is not a serious problem in this oleander since its great vigour means that it is best controlled in this respect, especially when intended for use as a potted plant (BAÑÓN et al. 2001).

Salinity significantly reduced leaf area by 15 % compared with the control (Table 1), while PBZ caused a reduction of 54 %. EL-KHEIR et al. (2000) also observed a reduction in the leaf area of *Helianthus annuus* as a result of salinity and PBZ. An analysis of leaf area in Fig. 1C shows how this parameter was only reduced in plants that had not been treated with PBZ, that is, the retardant lessened the reduction caused by salinity, although the accumulative effect of both factors strongly inhibited leaf development. This suggests that PBZ attenuates the water stress provoked by salinity

Table 1. Effect of salinity (S) and paclobutrazol (P) on growth and development in oleander seedlings. Relative chlorophyll content (RCC). Control-1 represents paclobutrazol-treated (and untreated) plants in non-saline conditions, while Control-2 represents salt-treated (and untreated) plants not treated with paclobutrazol. Average values are given.

Parameters	Control-1	S	Control-2	P	S	P	S x P
Aerial dry weight (g)	15.35	7.35	13.55	9.15	*	**	ns
Root dry weight (g)	15.35	4.75	11.35	8.75	***	*	**
Root/shoot ratio	1.05	0.68	0.76	0.98	***	***	***
Plant height (cm)	36.00	25.80	35.15	26.65	***	***	ns
Leaf area (dm^2)	3.75	3.20	4.75	2.20	***	**	*
Leaf salt symptoms (%)	0.00	12.40	11.85	0.55	***	***	***
Defoliation (%)	0.00	9.95	8.80	1.20	***	***	***
RCC	64.35	59.70	59.30	64.75	***	**	*

$P < 0.05$*, $P < 0.01$**, $P < 0.001$***, ns (not significant)

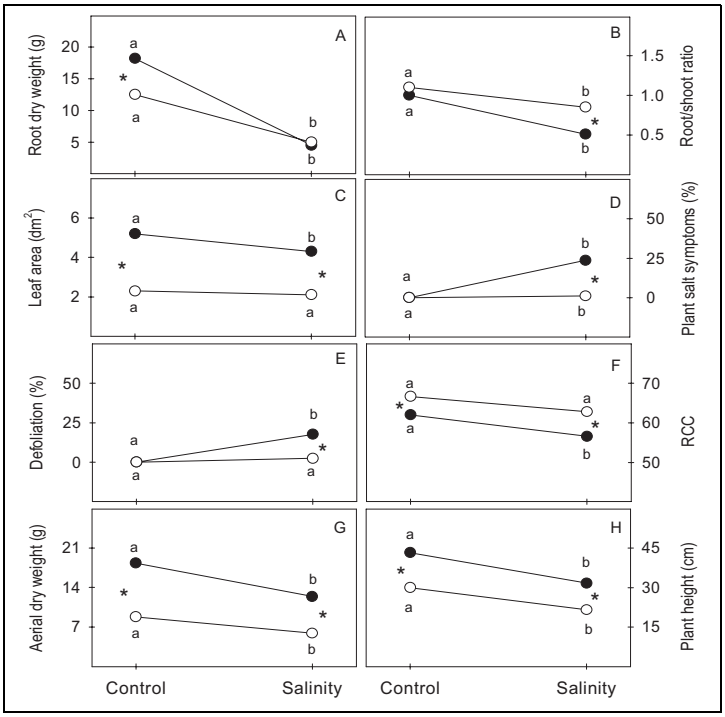


Fig. 1. Study of salinity x paclobutrazol interaction on the following parameters: root dry weight (A), root/shoot ratio (B), leaf area (C) leaf salt symptoms (D), defoliation (E), relative chlorophyll content (F). Mean values within a plot followed by the same letter are not significantly different ($p \leq 0.05$, Duncan's multiple range test). For both the saline treatment (70 mM) and control, an asterisk indicates significant differences ($p \leq 0.05$, Duncan's multiple range test) between PBZ-nontreated (filled symbols) and treated plants (empty symbols). Relative chlorophyll content (RCC).

since leaf development is normally reduced by a water deficit (BRADFORD and HSIAO 1982). Several authors have justified the drought resistance of PBZ-treated plants by reference to the lower transpiration caused by lower leaf area (JENKS et al. 2001).

Salinity was responsible for leaf damage symptoms, the most obvious being the appearance of necrosis at the tips, which extended along the limbus towards the petiole, finally causing the leaf to fall. Chlorotic symptoms were also seen in the limbus. The accumulation of salt ions in leaves of plants lacking the capacity to regulate them internally may be harmful (SÁNCHEZ-BLANCO et al. 1998). Salinity caused damage symptoms to appear on 12.4 % of the leaf area in oleander, while the application of PBZ strongly reduced these symptoms (Table 1). The interaction between salinity and the retardant clearly showed how PBZ reduced symptoms in plants treated with 70 mM NaCl (Fig. 1D). Defoliation behaved in a similar way to the leaf salt symptoms for the two factors analysed (Table 1 and Fig. 1E). Salinity did not result in plant death. The presence of salinity-induced damage is one of the principal indicators of salt stress in plants. BERNSTEIN et al. (1972) suggested that the presence of salt damage is a more important indicator than reductions in growth for assessing the salt-tolerance of ornamental plants, while FRANCOIS (1982), also referring to ornamentals, suggested that such symptoms should be taken as the principal criterion in determining the degree of salt tolerance. Based on these assertions and our results, we suggest that PBZ induces tolerance to salt stress in oleander. Other authors have shown that PBZ is capable of reducing symptoms of damage in leaves caused by different types of stress in other species (KRAUS et al. 1995; GILLEY and FLETCHER 1997).

RCC was significantly reduced by salinity and increased by PBZ (Table 1). A reduction in the leaf chlorophyll content as a result of salinity has been described by several authors (EL-KEHIR et al. 2000), while its increase is considered a typical effect of this retardant. A

Table 2. Effect of salinity (S) and paclobutrazol (P) on sodium and chloride contents (mg g^{-1} dry weight) in oleander seedlings at the end of cultivation phase. Control-1 represents paclobutrazol-treated (and untreated) plants in non-saline conditions, while Control-2 represents salt-treated (and untreated) plants not treated with paclobutrazol. Average values are given.

Parameters		Control-1	S	Control-2	P	S	P	S x P
Leaf	Cl^-	6.51	7.29	7.30	6.50	**	*	**
	Na^+	1.14	1.71	1.55	1.69	**	ns	ns
Stem	Cl^-	5.21	11.18	9.55	6.84	***	*	**
	Na^+	1.93	2.12	2.29	1.77	*	*	**
Root	Cl^-	7.74	17.46	13.48	11.73	**	*	*
	Na^+	5.83	26.37	17.36	14.84	***	**	*

P < 0.05*, P < 0.01**, P < 0.001***, ns (not significant)

Table 3. Effect of salinity (S) and paclobutrazol (P) on leachate and leached salt at the end of cultivation phase in oleander. Control-1 represents paclobutrazol-treated (and untreated) plants in non-saline conditions, while Control-2 represents salt-treated (and untreated) plants not treated with paclobutrazol. Average values are given.

Parameters	Control-1	S	Control-2	P	S	P	S x P
Leachate (%)	21.05	25.19	19.71	26.51	***	***	ns
Leached salt (g)	4.76	13.88	7.32	10.05	***	***	ns

P < 0.05*, P < 0.01**, P < 0.001***, ns (not significant)

Table 4. Effect of salinity (S) and paclobutrazol (P) on leaf water status in oleander seedlings at the end of the cultivation phase. Leaf water potential (Ψ_l, MPa), leaf turgor potential (Ψ_p, MPa), leaf osmotic potential at full turgor (Ψ_{os}, MPa), relative water content (RWC, %) and stomatal conductance (g_s). Control-1 represents paclobutrazol-treated (and untreated) plants in non-saline conditions, while Control-2 represents salt-treated (and untreated) plants not treated with paclobutrazol. Average values are given.

Parameters	Control-1	S	Control-2	P	S	P	S x P
RWC (%)	93.10	91.07	92.71	91.47	**	ns	ns
Ψ_l (MPa)	–1.00	–1.58	–1.27	–1.31	***	ns	ns
Ψ_p (MPa)	0.48	0.94	0.67	0.76	***	*	**
Ψ_{os} (MPa	–1.12	–1.41	–1.27	–1.30	***	ns	**
g_s (mmol m^{-2} s^{-1})	59.91	41.43	56.49	44.91	***	*	**

P < 0.05*, P < 0.01**, P < 0.001***, ns (not significant)

study of the interaction of both parameters showed that RCC was only reduced by salinity in plants not treated with PBZ (Fig. 1F) since the RCC values in treated plants were statistically equal in both the saline treatment and control. Such an effect may be mediated by the cytokinins, hormones that stimulate chlorophyll synthesis and the differentiation of chloroplasts (VISSER et al. 1992). An increase in cytokinin levels by PBZ has been suggested by GILLEY and FLETCHER (1997), while the same hormones retard senescence and prevent leaf fall (FLETCHER 1969). This suggests that the protective effect of PBZ against leaf salt damage and leaf loss in oleander may be related with the promotion of cytokinin production. Salinity increased the chloride and sodium content of leaves, stems and roots to a statistically significant degree (Table 2). PBZ reduced the content of these ions in all the organs studied, except sodium in leaves, whose levels did not change to a statistically significant extent. A study of the interaction of salinity and PBZ and the chloride content of the same organs showed how PBZ only reduced its level in saline conditions (Figs. 2A, B and D). The same was observed for the sodium content of stems and roots (Figs. 2C and E). The effectiveness of PBZ in reducing salt stress by reducing the accumulation of Na^+ and Cl^- in leaves was suggested by EL-KHASHAB et al. (1997) in peach and by SAHA and GUPTA (1999) in sunflower. The sodium concentration was higher in roots than in stems and leaves (Table 2), suggesting that this retention by roots may play an important part in oleander's adaptation to saline conditions (GREENWAY and MUNNS 1980).

Salt accumulation in the leaching water in this experiment indicated the lower availability of saline ions in PBZ-treated plants because the leached salt from PBZ-treated plants was statistically greater than from non-treated plants in the studied saline treatment (Table 3). This greater leaching action in PBZ-treated plants is a consequence of their lower growth compared with the non-treated plants (Table 1), which would also explain the greater salt content leached in the former (Table 3). This would explain the reduction in the uptake of sodium and chloride ions by PBZ-treated plants with a subsequent decrease in salt symptoms and defoliation.

The effect of salinity was significant on all the water stress-related parameters studied. However, PBZ application only significantly altered leaf turgor potential (Ψ_p) and stomatal conductance (g_s) (Table 4). Salinity resulted in a reduction of the relative water content of PBZ-treated plants, manifested by a slight degree of leaf dehydration. This is confirmed by the leaf water potential, which fell in saline conditions (Table 4). Such behavior has been observed in several Mediterranean species (SÁNCHEZ-BLANCO et al. 1998). PBZ, on the other hand, had no significant effect on either parameter, although it has been recorded that plants treated with this compound show higher water potentials (FLETCHER and HOFSTRA 1985). The Ψ_p was higher both with the salt treatment and PBZ, suggesting that both treatments increased leaf turgor with respect to the control. Analysis of the interaction suggests that PBZ increased Ψ_p in saline conditions but not in control conditions (Fig. 2F). Salinity significantly reduced leaf osmotic potential at full turgor (Ψ_{os}), while PBZ had no such effect (Table 4). However, analysis of the interaction showed that Ψ_{os} reduction caused by salt

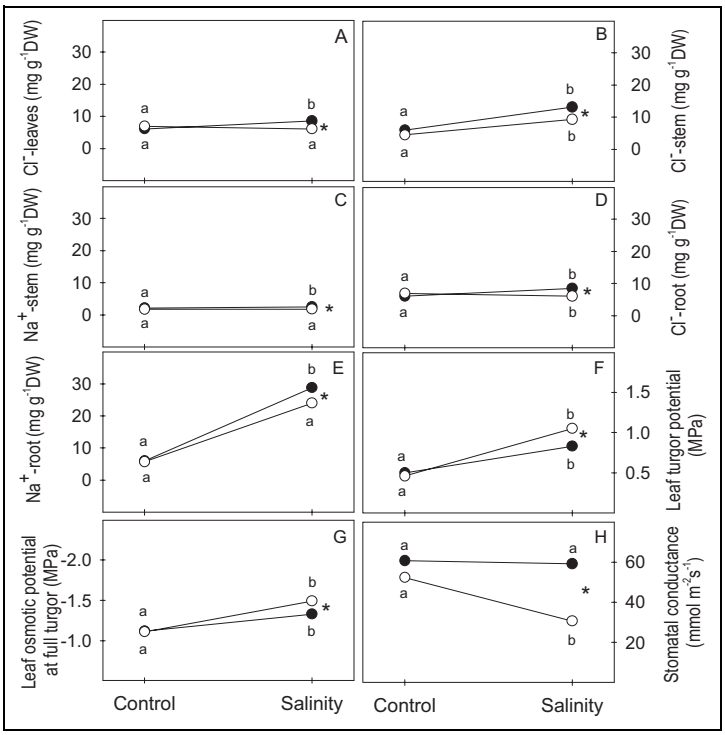


Fig. 2. Study of salinity x paclobutrazol interaction on the following parameters: leaves chloride content (A), stem chloride content (B), stem sodium content (C) root chloride content (D), root sodium content (E), leaf turgor potential (F), leaf osmotic potential at full turgor (G), stomatal conductance (H). Mean values within a plot followed by the same letter are not significantly different ($p \leq 0.05$, Duncan's multiple range test). For both the saline treatment (70 mM) and control, an asterisk indicates significant differences ($p \leq 0.05$, Duncan's multiple range test) between PBZ-nontreated (filled symbols) and treated plants (empty symbols).

was proportionally greater in PBZ-treated plants than in control plants (Fig. 2G). These results suggest that the osmotic effect induced by salinity (in both PBZ-treated and untreated plants) was alleviated by a process of osmotic adjustment (decrease of Ψ_{os}), which was sufficient to prevent any great loss of turgor. Furthermore, in saline conditions PBZ stimulates the accumulation of osmotically active compounds, probably osmoprotective organic compounds since the content of salt ions was reduced by the application of this retardant.

Stomatal conductance (g_s) was significantly reduced by salinity (Table 4), suggesting an effective stomatic regulation of the oleander seedlings as a mechanism to avoid salt-induced osmotic stress (De Herralde et al. 1998). PBZ reduced g_s, as was suggested by Fletcher and Hofstra (1985). However, when the interaction is analysed we see that the g_s was only reduced by salinity in PBZ-treated plants (Fig. 2H).

The study of the principal indicators of saline stress in plants, saline damage and the inhibition of vegetative growth (Wahome 2003), shows that, on the one hand, salinity induced the appearance of leaf necrosis and leaf fall, and, on the other, a reduction on plant dry weight, height and leaf development. PBZ considerably improved the first indicator in saline conditions and, to a certain extent, the second, since it diminished the reduction in root dry weight and leaf area. This protective effect of PBZ against saline stress in oleander may have several explanations. As regards ion accumulation, salinity increased the chloride and sodium content of leaves, stems and roots, while PBZ was effective in reducing the accumulation of both ions in leaf tissues, representing an important mechanism for reducing saline damage. As regards the water relations of plants irrigated with saline water, PBZ increased leaf turgor by favoring osmotic adjustment, probably through the promotion of organic solutes. Fletcher and Kraus (1995) suggested that plants may have an intrinsic stress protection potential and PBZ allows this to be expressed. The morphological modification of oleander by PBZ may be interpreted as the stimulation of avoidance mechanisms, such as the reduction of leaf area and the increase in the root/shoot ratio; the decrease in water loss through lower g_s should also be mentioned. However, these mechanisms seem to be more associated with the defense against water stress than against some ionic component and do not totally explain the reduction of saline damage by PBZ. The protection afforded to chlorophylls by PBZ, as observed in this study, suggests that the involvement of PBZ in the salt resistance mechanisms may also be related with modulation of the endogenous plant hormone balance (Fletcher and Hofstra 1985; El-Desouky and Atawia 1998).

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