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Manipulation of oleander growth, development and foliage colour by paclobutrazol and ethephon

Einfluß von Paclobutrazol und Ethephon auf das Wachstum, die Entwicklung und die Blattfarbe von Oleander

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Summary

The effect of different doses of paclobutrazol (1, 5, 10, 20, 40 and 80 mg per plant) and ethephon (0.1, 0.5, 1, 2, 3 and 5 g per plant) on the growth, development and leaf colour of oleander seedlings (*Nerium oleander* L. cv. Ajauque) was studied in order to evaluate the use of these plant growth retardants in the pot cultivation of this plant. The length of time which the effect of such treatment lasted was evaluated by studying the above parameters, four months and one year after treatment. At the first evaluation paclobutrazol (5 mg) and ethephon (0.1 g) were seen to have inhibited plant growth since plant height, internode length and the aerial part dry weight were substantially reduced. Ethephon (0.5 g) significantly increased the number of shoots per plant compared with the control, while paclobutrazol did so at all the doses assayed. Paclobutrazol (20 mg) and ethephon (0.5 g) reduced the leaf blade area. Paclobutrazol (40 mg) gave the leaves a more elliptic shape, while ethephon had no effect in this respect. Paclobutrazol (5 mg) imparted a slightly darker green colour to the leaves while ethephon (2 g) had the opposite effect. The latter compound at a dose 3 mg imparted a slightly yellowish colour to the leaves. At the second assessment, the plants treated with both paclobutrazol and ethephon continued to show restricted growth but any effect on foliage colour had disappeared. The degree to which plants were affected depended on which parameter was being analysed (internode length was the most affected by paclobutrazol and leaf blade area by ethephon) and ¥on the particular doses (the higher doses had a stronger effect).

Zusammenfassung

Um die Verwendung von Pflanzenwachstumsregulatoren (PGRs) in der Kultivierung von Oleander-Topfpflanzen (*Nerium oleander* L. cv. Ajauque) bewerten zu können, ist die Auswirkung unterschiedlicher Mengen Paclobutrazol (1, 5, 10, 20, 40 und 80 mg pro Pflanze) und Ethephon (0.1, 0.5, 1, 2, 3 und 5 g pro Pflanze) auf Wachstum, Entwicklung und Blattfarbe von Oleander-Sämlingen untersucht worden. Indem die obengenannten Parameter vier Monate und ein Jahr nach der Behandlung untersucht worden sind, ist bewertet worden, wie lange eine solche Behandlung anhält. Bei der ersten Untersuchung ist festgestellt worden, dass Paclobutrazol (5 mg) und Ethephon (0.1 g) das Pflanzenwachstum hemmen, wobei Pflanzenhöhe, Internodienlänge und das Trockengewicht des Sprosses erheblich reduziert sind. Ethephon (0.5 g) erhöhte die Zahl der Triebe pro Pflanze im Vergleich zur Kontrolle erheblich, während Paclobutrazol dieses bei allen angewendeten Mengen tat. Paclobutrazol (20 mg) und Ethephon (0.5 g) verringerten die Blattgrösse. Paclobutrazol (40 mg) gab den Blättern eine elliptischere Form, während Ethephon in dieser Hinsicht keine Wirkung hatte. Paclobutrazol (5 mg) führte zu einer etwas dunkelgrüneren Färbung der Blätter, während Ethephon (2 g) die gegensätzliche Wirkung zeigte. Ethephon (3 mg) gab den Blättern eine leicht gelbe Färbung. In der zweiten Untersuchung haben sowohl die mit Paclobutrazol als auch die mit Ethephon behandelten Pflanzen weiterhin ein begrenztes Wachstum gezeigt, aber die Auswirkung auf die Laubfarbe war verschwunden. Der Grad, in dem die Pflanzen betroffen worden sind, hängt sowohl von den Parametern, die analysiert worden sind, (die Länge der Internodien war am meisten von Paclobutrazol und die Blattgrösse am meisten von Ethephon betroffen) als auch von den unterschiedlichen Mengen ab (höhere Mengen hatten eine grössere Auswirkung).

Introduction

Oleander is a native of the Mediterranean area and much appreciated for its ornamental value. However, its ornamental use is restricted to gardens and landscaping and it is less used as a potted plant because of its strong vertical growth. It is well known that among the most important qualities looked for in the production of oleander for pot growth is a small, compact form, as has been demonstrated by the good commercial acceptance of dwarf oleander. Paclobutrazol is a strong growth retardant used in many plants to control their growth and development (RUTER 1996, WHIPKER and HAMMER 1997, HAMID and WILLIAMS 1997, GONZÁLEZ et al. 1999). Ethephon, an ethylene releasing

compound, has also demonstrated a capacity to modify plant growth (MUSE and HOLCOMB 1997, BAÑÓN et al. 1998, CARDOSO et al. 1998). Both paclobutrazol (ANDERSEN et al. 1998, KIM et al. 1999) and ethephon (ENDRES et al. 1999) have been shown to have an additional effect of modifying the colour of the leaves of some plants, another aspect which may increase their ornamental value.

The experiment described in this paper analyses the effect of different doses since this factor is know to play an important role in hormonal treatment. However, besides a knowledge of the effects which growth regulators have on plants, it is also important to ascertain how long such effects last so that the behaviour of plants can be foreseen. Such information is scant compared with the knowledge we have of the effects themselves. We therefore set out to determine the effect of different doses of paclobutrazol and ethephon on plant growth and development and on the leaf colour of oleander at two stages of growth in the nursery in order to improve the suitability of this species as a potted plant.

Materials and Methods

Plant material

Nerium oleander cultivar Ajauque (native of Región de Murcia, SE Spain) seedlings were used. The most homogeneous selection of seedlings possible (14–16 cm height) was made avoiding, to the greatest extent possible, variations resulting from sexual propagation.

Greenhouse growth conditions

Seedlings were transplanted on 21 May 1999 into plastic pots (1 plant per 14 cm pot) filled with a mixture of peat, perlite and clay soil (2:1:1, by volume) amended with Osmocote plus 5-11-22 (2 g/l of substrate), Metalaxyl (5 g per 10 l of substrate) and Oxamyl (5 g per 10 l of substrate). They were pot-grown in an unheated greenhouse covered with polycarbonate. The weather conditions during greenhouse culture were: 14-22 °C (minimum) and 24-44 °C (maximum); minimum relative humidity 19-70 % and a maximum 68-99 %. Plants were irrigated by a computer-controlled drip irrigation system, twice per week to 100 % water holding capacity (leaching 20–25 % of the applied water). Acrinathrin and Imidacloprid were applied as foliar spray to control twospotted spider mite and whitefly, respectively. Copper and Quinosol were applied to prevent bacteriosis. All plants were pinched 30 days after planting, leaving six nodes.

Shading screen growth conditions

On 20 Oct 1999 greenhouse culture was concluded, and then 18 plants per treatment were transplanted into plastic pots (1 plant per 22 cm pot) filled with a mixture of peat, perlite and clay soil (1:1:2, by volume) amended in the same way as for greenhouse cultivation. The plants were pot-grown in a metallic structure (2.8 m in height) covered with a plastic black screen (50 % transmissivity). The weather conditions during shading screen culture were 4-21 °C (minimum) and 12-32 °C (maximum); minimum relative humidity 12-60 % and a maximum 48-98 %. Irrigation was similar that to used during greenhouse culture.

Experimental design

The two plant growth retardants studied (paclobutrazol and ethephon) were used in two independent experiments. In the greenhouse, each treatment consisted of 36 randomly distributed plants, while during the shading screen culture each treatment consisted of 18 randomly distributed plants

Treatments

Paclobutrazol (CULTAR 25 %) at a rate of 1, 5, 10, 20, 40 and 80 mg per plant, or ethephon (FRUITEL 48 %) at 0.1, 0.5, 1, 2, 3 and 5 g per plant, were applied to the substrate surface as a single 45 ml liquid drench on the morning of 9 July 1999, two hours after the last watering. Untreated plants acted as control.

Measurements of plant parameters

The following parameters were recorded at the end of greenhouse culture: 1) plant height, 2) the number of shoots per plant, 3) principal stem diameter [measured 2 cm from the substrate surface], 4) internode length, 5) leaf blade area, 6) foliar area, 7) leaf width/length ratio, 8) dry weight of the aerial part, 9) colour coordinates and characteristics of leaves, 10) the relative amount of chlorophyll in leaves. Foliar area was measured with a DELTA T leaf area meter (Device Ltd., Cambridge, UK). Leaf width/length ratio measurements, the relative amount of chlorophyll and colour coordinates were determined in mature leaves at the first node from the apex in each shoot. Colour coordinates were measured with a MINOLTA CR 10 colorimeter, and the relative amount of chlorophyll with a MINOLTA SPAD-502 chlorophyll meter, both at the midpoint of the leaves. Leaf colour measurements used CIELAB L* a* b* coordinates. The hue angle was calculated as $\tan^{-1}$ b*/a* (in degrees), and chroma was calculated as $(a^{*2} + b^{*2})^{1/2}$ (MCGUIRE 1992). The presence of phytotoxicity symptoms was measured using a rating of 0 to 3: (0) none, (1) low, (2) average, (3) high. The percentage of dead plants was also determined.

At the end of shading screen culture (on 26 June 2000) the following measurements were determined in the new shoots: 1) internode length, 2) leaf width/length ratio, 3) leaf blade area, 4) aerial part dry weight, 5) number of flower buds per plant, 6) colour coordinates and characteristics of leaves, 7) the relative amount of chlorophyll in leaves.

Statistical analysis

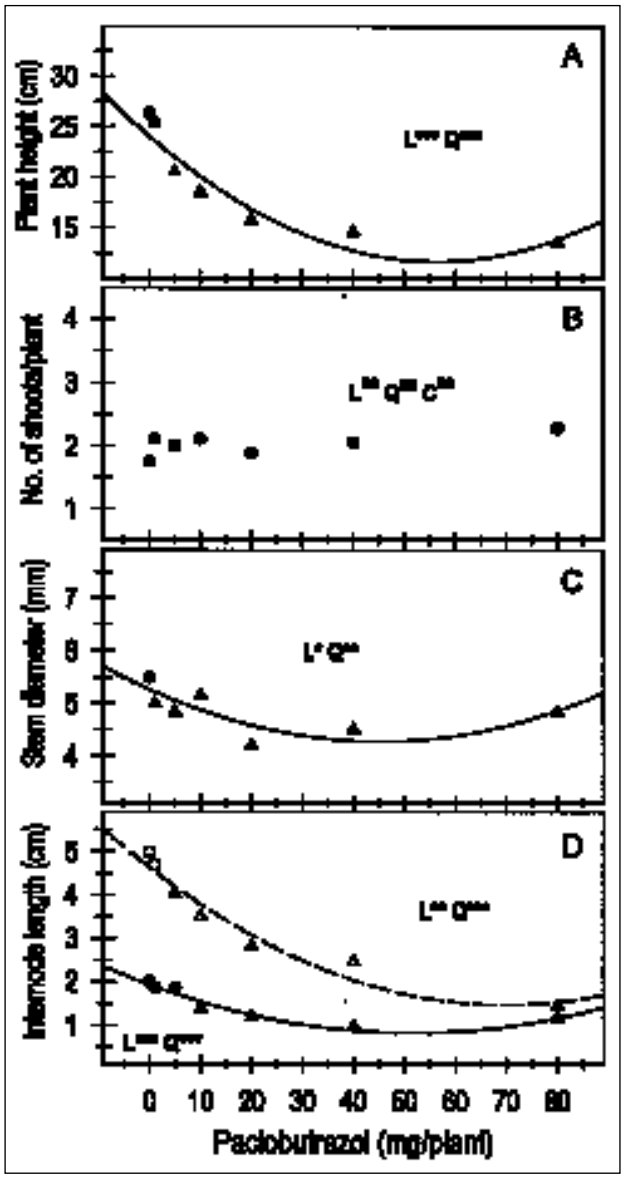
The significance of the effects of both plant growth regulators (paclobutrazol and ethephon) were analysed by ANOVA using STATGRAPHICS PLUS for Windows. Treatment means were separated with Duncan's Multiple Range Test (P 0.01). A regression analysis was made using SIGMAPLOT 2.0 for Windows.

Results

Greenhouse culture

Paclobutrazol

Plant height showed a significant quadratic response to paclobutrazol doses, the height of plants decreasing as the dose of paclobutrazol increased. Compared with the height of control plants. The height of plants treated with 5 mg was significantly reduced (by 22 % to 49 %.) (Fig. 1A). The effect of paclobutrazol on the number of shoots per plant was not significant (Fig. 1B). Stem diameter showed a significantly quadratic relationship with paclobutrazol doses. All the paclobutrazol doses reduced stem diameter by a statistically significant degree compared with the control (6 % to 24 % reduction) (Fig. 1C). Internode length showed a significantly high quadratic relationship with paclobutrazol doses of 10 mg, this parameter being significantly reduced by 31 % to 52 % compared with the control (Fig. 1D)



The response of leaf blade area, leaf area, leaf width/length ratio and aerial part dry weight to paclobutrazol doses showed significant quadratic regression models (Fig. 2). Doses of 20 mg significantly reduced the leaf blade area by 41 % to 49 % compared with the control (Fig. 2A). Paclobutrazol at 5, 40 and 80 mg significantly reduced the foliar area compared with the control (by 28 %, 57 % and 52 %, respectively) (Fig. 2B). Plants treated with 40 or 80 mg of paclobutrazol significantly increased the leaf width/length ratio from 0.19 (control) to 0.26 (Fig. 2C). Doses of 5 mg reduced the aerial part dry weight by 22 % to 53 % compared with the control (Fig. 2D).

The response of chroma to paclobutrazol showed a significantly cubic relationship, and the doses at 5 and 80 mg statistically reduced its value by 10 % compared with the control (Fig. 3A). The effect of the paclobutrazol doses on hue angle showed a significant quadratic relationship, the 10 and 80 mg doses significantly increasing this parameter (Fig. 3B). The regression analysis showed how clarity (L*) was reduced with increasing doses of paclobutrazol (see parabola of figure 3C). Doses of 5 mg significantly reduced this parameter by 4 % to 6.3 % compared with untreated plants. There was a significant quadratic relationship between the relative amount of chlorophyll in leaves and paclobutrazol doses. Doses of 5 mg significantly increased the relative amount of chloro-

Fig. 1. Influence of paclobutrazol doses on plant height, number of shoots per plant, stem diameter and internode length. Continuous lines and filled symbols indicate measurements made four months after paclobutrazol application (greenhouse measurements). Broken lines and empty symbols indicate measurements made one year after treatments (shading screen measurements). Triangles indicate significant differences with the control according to Duncan's Multiple Range Test (P 0.01). L = linear, Q = quadratic, C = cubic. ^{ns}, *,**,***, indicant not significant, statistically significant at P 0.05, 0.01 or 0.001, respectively. Regression lines were generated from the means of the treatments, and the points are means for each treatment (n = 36; n = 18 for destructive or shading screen measurements). Regression equations and adjusted r^2 are: A) $y = 0.004x^2 - 0.44x + 24.07$, $r^2 = 0.60$; C) $y = 0.0005x^2 - 0.04x + 5.27$, $r^2 = 0.21$; D) $y = 0.0004x^2 - 0.04x + 1.93$, $r^2 = 0.60$ (continuous line); $y = 0.0006x^2 - 0.09x + 4.6$, $r^2 = 0.55$ (broken line)

Einfluss der Paclobutrazoldosis auf Pflanzenhöhe, Sprosszahl pro Pflanze, Stammdurchmesser und Internodienlänge. Durchgehende Linien und ausgefüllte Symbole bezeichnen Messungen, die vier Monate nach der Paclobutrazolgabe durchgeführt wurde (Gewächshausmessungen). Unterbrochene Linien und leere Symbole bezeichnen Messungen, die ein Jahr nach der Behandlung erhoben wurden (Messungen unter beschattendem Netz). Dreiecke bezeichnen signifikante Unterschiede zur Kontrolle gemäss des Duncan Multiple Range Tests (P 0.01). L = linear, Q = quadratisch, C = dritten Grades. ^{ns},,**, ***, bedeutet jeweils: nicht signifikant, statistisch signifikant bei P 0.05, 0.01 oder 0.001. Regressionslinien wurden von den Mittelwerten der Messungen abgeleitet und die Punkte stellen die Mittelwerte jeder Behandlung dar (n = 36; n = 18 für "Pflanzen zerstörende" Messungen und Messungen unter beschattendem Netz). Die Regressionsgleichungen und angepassten r^2-Werte lauten: A) $y = 0.004x^2 - 0.44x + 24.07$, $r^2 = 0.60$; C) $y = 0.0005x^2 - 0.04x + 5.27$, $r^2 = 0.21$; D) $y = 0.0004x^2 - 0.04x + 1.93$, $r^2 = 0.60$ (durchgehende Linie); $y = 0.0006x^2 - 0.09x + 4.6$, $r^2 = 0.55$ (unterbrochene Linie)*

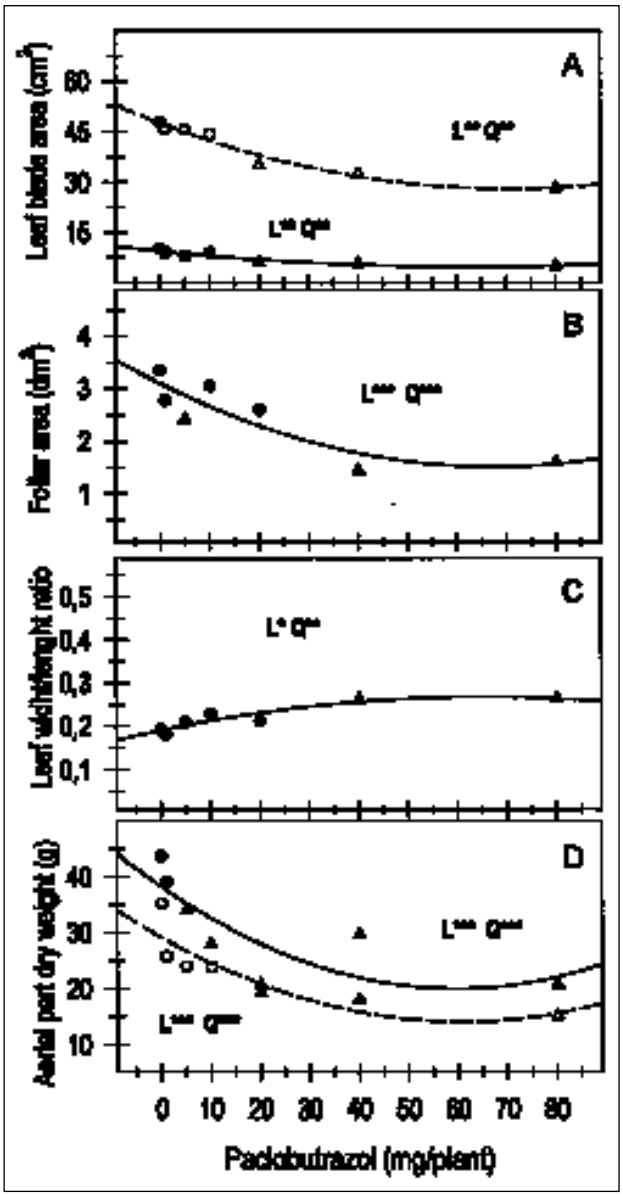


Fig. 2. Influence of paclobutrazol doses on leaf blade area, foliar area, leaf width/length ratio and the aerial part dry weight (or dry weight of shoots in shading screen measurements). For legend see figure 1. Regression equations and adjusted r^2 are: A) $y = 0.0012x^2 - 0.14x + 9.43$, $r^2 = 0.22$ (continuous line); $y = 0.0039x^2 - 0.56x + 47.67$, $r^2 = 0.46$ (broken line); B) $y = 0.00034x^2 - 0.05x + 3.10$, $r^2 = 0.23$; C) $y = -0.000014x^2 - 0.0028x + 0.36$, $r^2 = 0.11$; D) $y = 0.0051x^2 - 0.62x + 38.21$, $r^2 = 0.41$ (continuous line); $y = 0.0041x^2 - 0.58x + 29.15$, $r^2 = 0.59$ (broken line)

Einfluss der Paclobutrazoldosis auf Blattfläche, Gesamtlaubfläche, Verhältniss Blattlänge/Blattweite und Trockengewicht der oberirdischen Pflanzenteile (oder Trockengewicht der Sprosse in Messungen unter beschattendem Netz). Zu den Erläuterungen siehe Abb. 1. Die Regressionsgleichungen und angepassten r^2-Werte lauten: A) $y = 0.0012x^2 - 0.14x + 9.43$, $r^2 = 0.22$ (durchgehende Linie); $y = 0.0039x^2 - 0.56x + 47.67$, $r^2 = 0.46$ (unterbrochene Linie); B) $y = 0.00034x^2 - 0.05x + 3.10$, $r^2 = 0.23$; C) $y = -0.000014x^2 - 0.0028x + 0.36$, $r^2 = 0.11$; D) $y = 0.0051x^2 - 0.62x + 38.21$, $r^2 = 0.41$ (durchgehende Linie); $y = 0.0041x^2 - 0.58x + 29.15$, $r^2 = 0.59$ (unterbrochene Linie)

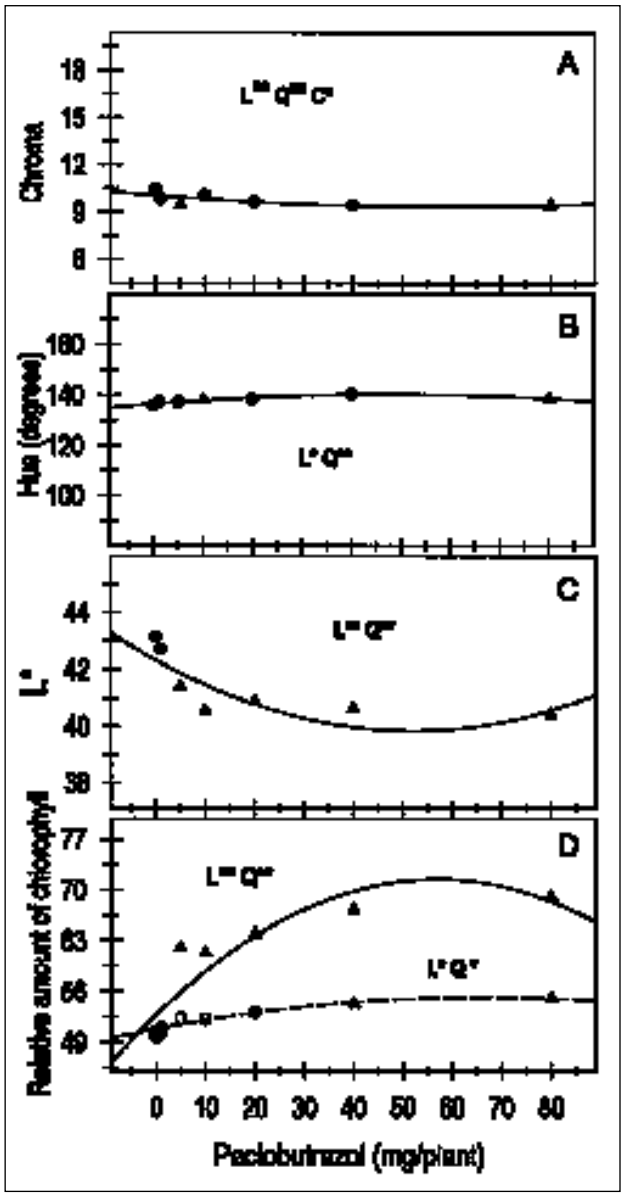


Fig. 3. Influence of paclobutrazol doses on colour characteristics and the relative amount of chlorophyll in leaves. For legend see figure 1. Regression equations and adjusted r^2 are: A) $y = -4.03x^3 + 0.31x^2 + 0.00007x^2 + -0.04x + 10.1$, $r^2 = 0.11$; B) $y = -0.0017x^2 + 0.16x + 136.54$, $r^2 = 0.22$; C) $y = 0.0093x^2 - 0.09x + 42.34$, $r^2 = 0.25$; D) $y = -0.0058x^2 + 0.65x + 52.76$, $r^2 = 0.42$ (continuous line); $y = -0.00097x^2 + 0.13x + 50.98$, $r^2 = 0.24$ (broken line)

Einfluss der Paclobutrazoldosis auf Farbmerkmale und relativen Chlorophyllgehalt in den Blättern. Zu den Erläuterungen siehe Abb. 1. Die Regressionsgleichungen und angepassten r^2-Werte lauten: A) $y = -4.03x^3 + 0.31x^2 + 0.00007x^2 + -0.04x + 10.1$, $r^2 = 0.11$; B) $y = -0.0017x^2 + 0.16x + 136.54$, $r^2 = 0.22$; C) $y = 0.0093x^2 - 0.09x + 42.34$, $r^2 = 0.25$; D) $y = = -0.0058x^2 + 0.65x + 52.76$, $r^2 = 0.42$ (durchgehende Linie); $y = -0.00097x^2 + 0.13x + 50.98$, $r^2 = 0.24$ (unterbrochene Linie)

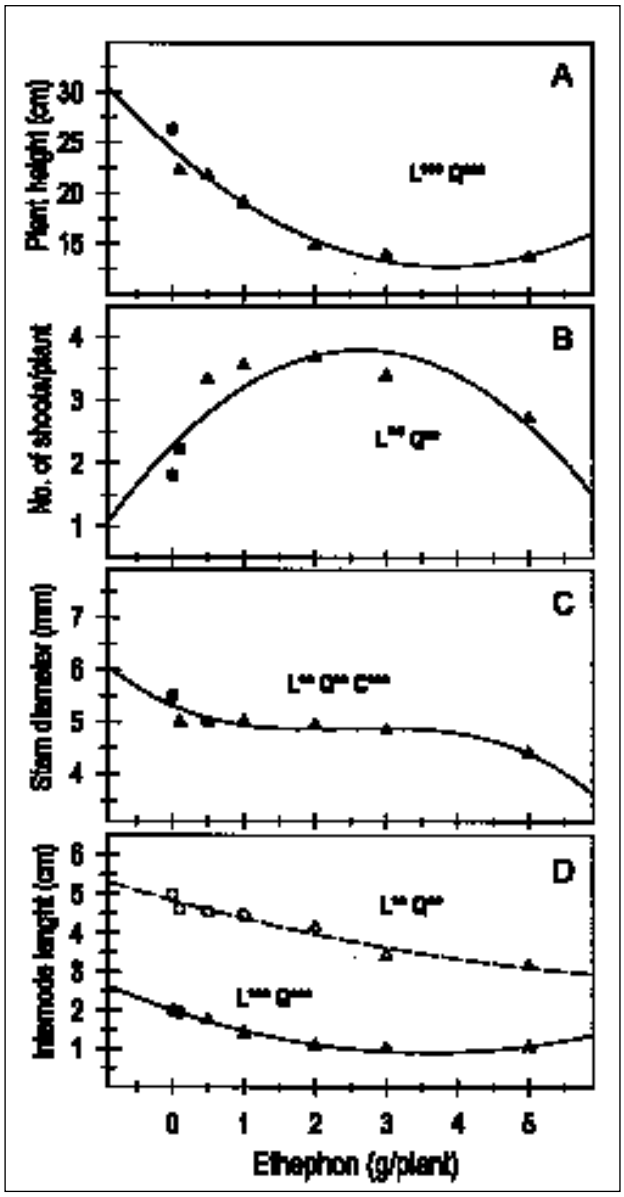


Fig. 4. Influence of ethephon doses on plant height, number of shoots per plant, stem diameter and internode length. For legend see figure 1. Regression equations and adjusted r^2 are: A) $y = 0.79x^2 - 6.05x + 24.36$, $r^2 = 0.59$; B) $y = -0.22x^2 + 1.14x + 2.28$, $r^2 = 0.14$; C) $y = -0.03x^3 + 0.24x^2 - 0.58x + 5.31$, $r^2 = 0.21$; D) $y = 0.08x^2 - 0.62x + 2.01$, $r^2 = 0.66$ (continuous line); $y = 0.03x^2 - 0.49x + 4.83$, $r^2 = 0.26$ (broken line).

Einfluss der Ethephondosis auf Pflanzenhöhe, Sprosszahl pro Pflanze, Stammdurchmesser und Internodienlänge. Zu den Erläuterungen siehe Abb. 1. Die Regressionsgleichungen und angepassten r^2-Werte lauten: A) $y = 0.79x^2 - 6.05x + 24.36$, $r^2 = 0.59$; B) $y = -0.22x^2 + 1.14x + 2.28$, $r^2 = 0.14$; C) $y = -0.03x^3 + 0.24x^2 - 0.58x + 5.31$, $r^2 = 0.21$; D) $y = 0.08x^2 - 0.62x + 2.01$, $r^2 = 0.66$ (durchgehende Linie); $y = 0.03x^2 - 0.49x + 4.83$, $r^2 = 0.26$ (unterbrochene Linie)

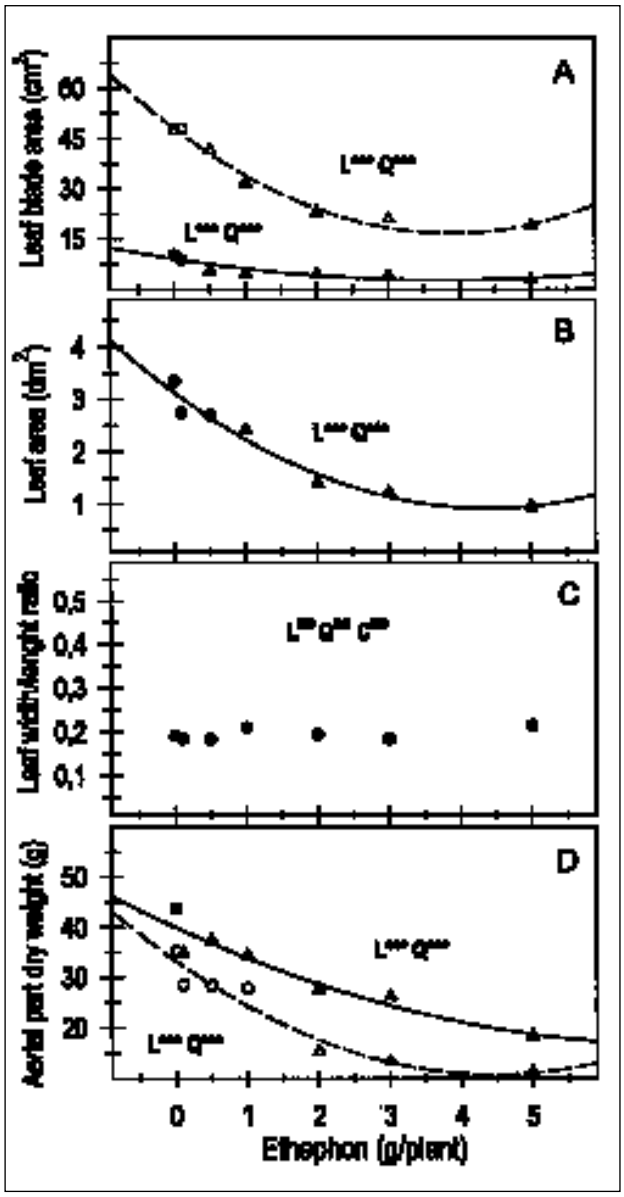


Fig. 5. Influence of ethephon doses on leaf blade area, foliar area, leaf width/length ratio and the aerial part dry weight (or dry weight of shoots in shading screen measurements). For legend see figure 1. Regression equations and adjusted r^2 are: A) $y = 0.43x^2 - 3.31x + 9.06$, $r^2 = 0.43$ (continuous line); $y = 2.08x^2 - 16.17x + 48.01$, $r^2 = 0.64$ (broken line); B) $y = 0.11x^2 - 1.01x + 3.18$, $r^2 = 0.38$; D) $y = 0.46x^2 - 6.25x + 39.91$, $r^2 = 0.69$ (continuous line); $y = 1.12x^2 - 9.98x + 33.17$, $r^2 = 0.82$ (broken line)

Einfluss der Ethephondosis auf Blattfläche, Gesamtlaubfläche, Verhältniss Blattlänge/Blattweite und Trockengewicht der oberirdischen Pflanzenteile (oder Trockengewicht der Sprosse in Messungen unter beschattendem Netz). Zu den Erläuterungen siehe Abb. 1. Die Regressionsgleichungen und angepassten r^2-Werte lauten: A) $y = 0.43x^2 - 3.31x + 9.06$, $r^2 = 0.43$ (durchgehende Linie); $y = 2.08x^2 - 16.17x + 48.01$, $r^2 = 0.64$ (unterbrochene Linie); B) $y = 0.11x^2 - 1.01x + 3.18$, $r^2 = 0.38$; D) $y = 0.46x^2 - 6.25x + 39.91$, $r^2 = 0.69$ (durchgehende Linie); $y = 1.12x^2 - 9.98x + 33.17$, $r^2 = 0.82$ (unterbrochene Linie)

phyll in leaves by 23.4 % to 38.9 % compared with the control (Fig. 3D). None of the doses applied resulted in plant death or produced phytotoxicity symptoms in leaves.

Ethephon

There was a highly significant quadratic relationship between ethephon doses and plant height, all doses producing a statistically significant reduction in this parameter compared with the control (15-48 % reduction) (Fig. 4A). The number of shoots per plant reflected the significant quadratic effect of ethephon, doses of 0.5 g significantly increasing the number of shoots from 1.8 (control) to 3.8 (2 g) (Fig. 4B). There was a significant cubic relationship between stem diameter and ethephon dose, the 0.1, 1, 2, 3 and 5 g doses produced reductions of 12, 11, 14, 15 and 34 %, respectively (Fig. 4C). The effect of ethephon on internode length is demonstrated by the parabola shown in figure 4D, doses of 0.5 g significantly reducing this parameter by between 12 and 50.8%.

Leaf blade area, leaf area and aerial part dry weight showed a significant quadratic relationship with the doses applied (Figs. 5A, 5B and 5D). Doses of 0.5 g significantly reduced leaf blade area by between 46 and 71 % compared with the control (Fig. 5A). Doses of 1 g had a similar effect on leaf area (reduced by 28-72 %) (Fig. 5B). However, ethephon had no significant effect on the leaf width/length ratio (Fig. 5C). All the doses assayed significantly reduced by 15-58 % the dry weight of the aerial part compared with the control value (Fig. 5D).

Hue angle and the relative chlorophyll content of the leaves showed a statistically significant linear relationship with the ethephon doses applied (Figs. 6B and 6D). Chroma and L* exhibited a statistically significant quadratic response to paclobutrazol, and doses of 2 g increased these parameters significantly compared to the control values (Figs. 6A and 6C). These same doses reduced the hue angle by with 11.9 and 20.4 degrees (Fig. 6B). All ethephon doses significantly reduced (by 11.7 % to 46.7 %) the relative chlorophyll content of leaves compared with the control (Fig. 6D). Chlorotic symptoms were detected in the leaves of plants treated with 2 g ethephon, especially in those treated with 5 g, which besides losing leaves, showed 17 % mortality (Tab. 1).

Table 1. Chlorosis, defoliation and mortality in ethephon-treated oleander. Greenhouse measurements
Chlorose, Laubverlust und Sterblichkeit in Oleander, behandelt mit Ethephon. Gewächshausdaten.

Treatments mg/plant	Chlorosis	Defoliation	Mortality %
Control	-	-	0
0.1	-	-	0
0.5	-	-	0
1	-	-	0
2	1	-	0
3	2	-	0
5	3	2	17

Rating: (0) none, (1) low, (2) average, (3) high
Bewertung: (0) keine, (1) niedrig, (2) mittel, (3) hoch

Shading screen culture

Paclobutrazol

Internode length, leaf blade area, aerial part dry weight and the relative amount of chlorophyll in leaves showed a significant quadratic response to paclobutra-

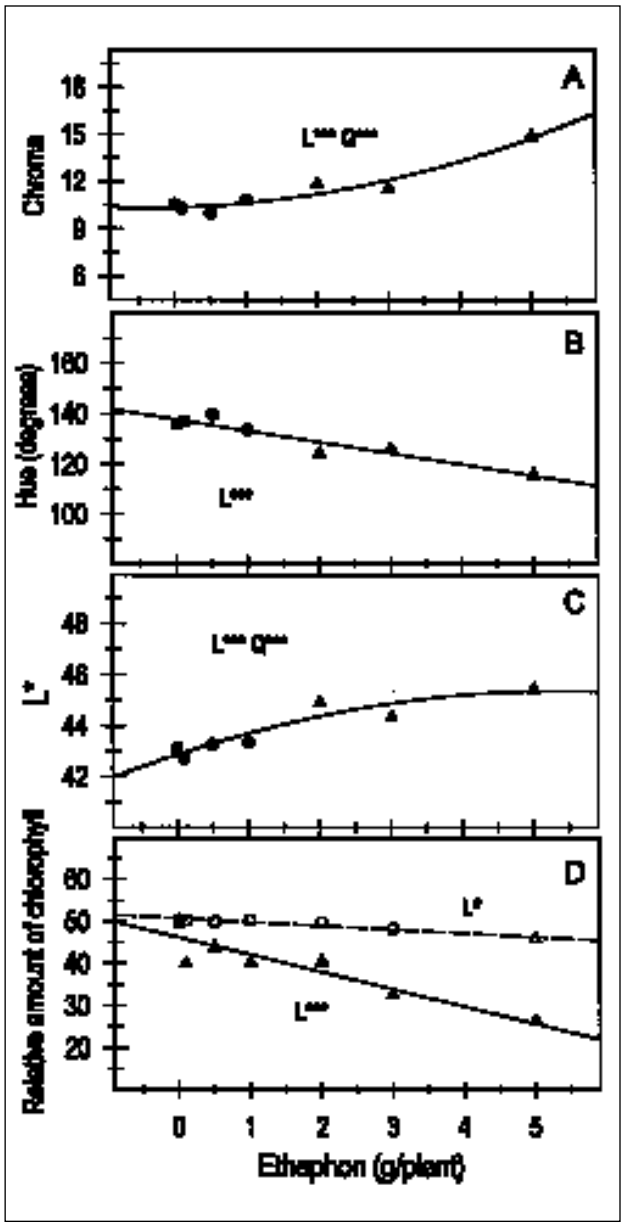


Fig. 6. Influence of ethephon doses on colour characteristics and the relative amount of chlorophyll in leaves. For legend see figure 1. Regression equations and adjusted r^2 are: A) $y = 0.15x^2 + 0.13x + 10.35$, $r^2 = 0.63$; B) $y = -4.47x + 137.76$, $r^2 = 0.56$; C) $y = -0.08x^2 + 0.93x + 42.88$, $r^2 = 0.39$; D) $y = -4.08x + 46.17$, $r^2 = 0.58$ (continuous line); $y = -0.88x + 50.69$, $r^2 = 0.26$ (broken line)
Einfluss der Ethephondosis auf Farbmerkmale und relativen Chlorophyllgehalt in den Blättern. Zu den Erläuterungen siehe Abb. 1. Die Regressionsgleichungen und angepassten r^2-Werte lauten A) $y = 0.15x^2 + 0.13x + 10.35$, $r^2 = 0.63$; B) $y = -4.47x + 137.76$, $r^2 = 0.56$; C) $y = -0.08x^2 + 0.93x + 42.88$, $r^2 = 0.39$; D) $y = -4.08x + 46.17$, $r^2 = 0.58$ (durchgehende Linie); $y = -0.88x + 50.69$, $r^2 = 0.26$ (unterbrochene Linie)

zol doses (Figs. 1D, 2A, 2D and 3D). The regression analysis was not significant when the colour characteristics and leaf width/length ratio in the new shoots were studied (data not shown). Doses of 5 mg reduced internode length by 19 % to 71.1 % and the aerial part (shoots) dry weight by 45.3 % to 57.3 % compared to the control (Figs. 1D and 2D, respectively). Doses of 20 mg reduced by a statistically significant extent the leaf blade area (by 26.7 to 30.6 %) compared with the control. Doses of 40 and 80 mg significantly increased the relative amount of chlorophyll (by 9.2 and 10.8 %, respectively) compared with the control (Fig. 3D). No flower buds were observed in either treated or controls plants.

Ethephon

As in the case of paclobutrazol, ethephon had no significant effect on the leaf width/length ratio or colour coordinates and characteristics (data not shown). Internode length showed a significant quadratic response to ethephon doses, with the 2, 3 and 5 g doses significantly reducing the values of this parameters by 24 %, 32 % and 37 %, respectively (Fig. 4D). The relationship between ethephon treatment and leaf blade area was also quadratic, doses of 0.5 g reducing this latter parameter by 9 to 64 % (Fig. 5A). There was a highly significant quadratic relationship between shoot dry weight and ethephon doses, the (2, 3 and 5 g) doses producing a 56 %, 62 % and 68 % reduction compared with the control. None of the treated plants showed flower buds. The relative chlorophyll content of leaves was significantly related with ethephon as shown by the gently sloping straight line. Only the 5 g dose reduced this parameter by a statistically significant degree (Fig. 6D).

Discussion

Effects of paclobutrazol and ethephon in greenhouse culture

Both paclobutrazol and ethephon were effective in reducing the height of oleander at the doses studied, which makes them suitable for dwarfing plants for prospective use as potted plant. The fact that plant height and internode length were reduced by similar percentages suggests that the first was a result of the second. Paclobutrazol has been used to reduce the height of other ornamental plants (GAD et al. 1997, WHIPKER and HAMMER 1997, GONZÁLEZ et al. 1999), and it has also been seen to reduce internode length (MALORGIO et al. 1993, GAD et al. 1997). Ethephon, too, has been seen to reduce the plant height on other species (MUSE and HOLCOMB 1997, BAÑÓN et al. 1998).

The production of lateral shoots was not modified by paclobutrazol but was increased by ethephon. This is of interest for adapting oleander to pot growth since a greater volume of lateral growth is needed to balance the natural vertical growth of this plant. The ethephon doses of greatest interest were these between 0.5 and 2 since higher doses tended to reduce this beneficial effect (Fig. 4B). Such a response is probably the consequence of ethephon´s known ability to reduce apical dominance (YEANG and HILLMAN 1984) and is in agreement with the findings of MUSE and HOLCOMB (1997) en Pelargonium peltatum and Vinca major. The lack of any effect on the number of shoots per plant limits the usefulness of paclobutrazol for the purposes outlined in this study. GONZÁLEZ et al. (1999) found similar results in Zantedeschia aethiopica, although HAMID and WILLIAMS (1997) and KARAGUZEL (1999) found that paclobutrazol increased this parameter in other species.

Principal stem diameter was reduced by both paclobutrazol and ethephon, particularly the former, which is of great interest when trying to reduce plant size. YEWALE et al. (1998) found that paclobutrazol reduced the stem diameter of chrysanthemum. Among other reasons, this would be due to the reducing effect paclobutrazol has on xylem development (PROIETTI et al. 1998). The cubic function relating ethephon doses and main stem diameter (Fig. 4C) suggests that high doses (in excess of 3 g) are needed to obtain any meaningful reduction. Studies carried out in Liatris spicata showed that the diameter of the flowering stem could only be reduced by using high concentrations of ethephon (BAÑÓN et al. 1998).

Both plant growth regulators reduce the leaf blade area, ethephon being the more effective. The presence of smaller leaves gives the impression of a small plant and imparts a finer texture. The reduction of leaf blade size by paclobutrazol has been mentioned by LIU et al. (1995) in several species and ethephon has been seen to reduce foliar area in Curcubita moschata (CARDOSO et al. 1998). Paclobutrazol produced a slightly elliptic shape in the naturally linear form of the leaves, an effect which helped give a different look to the plant after hormonal treatment. TONKINSON et al. (1995) suggested that paclobutrazol shortened the leaves of wheat.

Both paclobutrazol and ethephon were very effective in reducing the aerial growth of oleander and for minimising its size. The reduction noted in the dry weight of the aerial part after treatment with paclobutrazol agrees with the findings of RUTER (1996) in New Gold Lantana. A similar reduction in the dry weight of corn was suggested by KASELE et al. (1995) after the application of ethephon.

The inhibitory effects of both paclobutrazol and ethephon on shoot growth are well known. In some plants, the inhibitory effects of ethylene on shoot growth has been attributed to a reduction in auxin levels either through modification of their metabolism (RIOV et al. 1982) or inhibition of their transport (KOCH and MOORE 1990). According to the above, ethephon probably decreased auxin levels in oleander and reduced stem growth by inhibiting cell enlargement. Indirect evidence of this was the defoliation seen in the plants treated with the higher doses, which can be attributed to a sharp reduction in auxin levels caused by ethephon (MORGAN and DURHAM 1972). However, the physiological mechanism of ethylene's action on shoot growth is little understood (ABELES et al. 1992). It is also possible that the greater acidity of higher doses of ethephon affected the growing regions resulting in reduced growth (HAN and NOBEL 1995). Paclobutrazol reduces shoot extension by inhibiting the biosynthesis of gibberellins (MARTIN et al. 1994), a plant hormone that promotes plant growth by stimulating cell division and enlargement. It is likely that a decrease in endogenous gibberellins caused by pa-

clobutrazol reduced the height and internode length of the oleander plants treated with this compound.

The different action mechanism of the two compounds studied may explain the effects on the lateral sprouting of treated oleanders, ethephon reducing the levels of auxins and paclobutrazol increasing the same activity (?EBÀNEK et al. 1991). That auxin and cytokinin are capable of regulating apical dominance is well established (TAMAS 1995). If the level of auxin in the stem of oleander was reduced by ethephon, the apical dominance was hindered and lateral buds growth was encouraged. However, although other reports have indicated the involvement of ethylene in apical dominance, it is still not known whether endogenous ethylene is involved in the regulation of this process (RASKIN 1991).

The hue angle, clarity and chroma values registered in the control plants reflect the pale green colour and medium clarity of this oleander. From the results obtained (Figs. 3A and B) it is clear that paclobutrazol does not much alter the tone and saturation shown by the control plants. However, ethephon shifted the green colour of leaves towards yellow especially at the higher doses. This colorimetric shift would be related with the signs of chlorosis seen in leaves (Tab. 1), as observed by SHATTERS et al. (1998) in Cynodon dactylon. Paclobutrazol (doses of 5 mg) produced a slight darkening of the green colour of leaves, which may be regarded as a positive trait in the evaluation of oleander as an ornamental plant. This darkening effect caused by paclobutrazol has also been observed in the leaves of Lolium perenne (WANG et al. 1994) and Dicentra spectabilis (KIM et al. 1999), while PAN and LUO (1994) related the darkening of Cymbidium sinense leaves with the increased chlorophyll levels caused by paclobutrazol. The lighter colour observed in the leaves of plants treated with ethephon was probably due to the reduced chlorophyll level caused by this plant regulator. The acceleration of chlorophyll degradation by ethephon is well documented (ENDRES et al. 1999). None of the doses of paclobutrazol damaged the plants although there are references to chlorosis in leaves caused by this plant growth retardant (HAMID and WILLIAMS 1997). The 5 g doses of ethephon marked the threshold value for phytotoxicity in oleander.

Effect of paclobutrazol and ethephon in shading screen culture

The parabolas relating internode length and paclobutrazol doses at the two times studied (Fig. 1D) show that the inhibitive effect intensified with time, especially at the highest doses. The significance of the effect of the 5 mg dose of paclobutrazol on the shading screen measurements (in the greenhouse this dose produced no significant effect compared with the control) reflects this. This increasing effect of paclobutrazol on internode length with the passage of time may have due to its accumulation in tissues (LEHMAN et al. 1990) and its slow release to the plant (GROCHOWSKA and HODUN 1997). The response of internode length to ethephon was similar although not so pronounced (Fig. 4D). The lack of significance of the 0.5 g and 1 g doses of ethephon and the significance of 5 mg dose of paclobutrazol in the shading screen measurements compared with the greenhouse measurements suggest that the doses used plays an important role in whether or not the restrictive effect on internode length is maintained.

The plotting of leaf blade area versus growth retardant dosage parabolas in both the shading screen and greenhouse stages (Figs. 2A and 5A) points to the increasing effect of paclobutrazol and, especially, ethephon on this parameter with time, meaning that the differences in leaf size between treated and untreated plants were more pronounced at the second measuring time. However, the effect of paclobutrazol on the shape of the leaf limbo did not last the year.

One explanation for the increasing inhibitive effects of both products on internode length and leaf blade area can be found in the vigorous growth of the control plants during shading screen culture when the internode length increased from 2 cm (greenhouse measurements) to 5 cm and the leaf blade area changed from 11 to 47 cm^2 in untreated plants as a consequence of favourable climatic conditions and greater vigour (older plants).

Figures 2D and 5D show the parabolas relating aerial part dry weight and the doses of both growth regulators at the measurement times. Although the curves cannot be compared quantitatively since one refers to the whole plant (greenhouse measurements) and the other to new shoots (shading screen measurements), they show that the restrictive effect on aerial growth was maintained with paclobutrazol (doses of 20 mg) and intensified to a certain extent by ethephon (doses of 2 g). The loss of significance with respect to the control of the 5 and 10 mg doses of paclobutrazol and of the 0.1, 0.5 and 1 g doses of ethephon in the new shoots suggests that, as in the case of internode length, the dose plays an important role in maintaining the effects on this parameters.

The shading screen results also demonstrate the loss of any effect produced on leaf colour in the greenhouse, despite the differences seen between the relative amount of chlorophyll in plants treated with the highest doses and the amount seen in the control (Figs. 3D and 6D).

The fact that no flower buds appeared in any of the treatments with either growth retardant indicates that neither shortened the juvenile period (the plants were 16 months old when the measurements were made in the shade area), despite the well-known ability of ethephon to promote earlier flowering (ABELES et al. 1992).

The length of time which the effect of paclobutrazol last has been studied much more than the corresponding data for ethephon. RUTER (1994), for example noted that paclobutrazol was still effective 13 months after application in Pyracantha but not in Juniperus. MALORGIO et al. (1993) observed that some effects were still noticeable seven months after treatment in gardenia and WANG et al. (1996) found that the effect of paclobutrazol was still evident in the new growth observed 2 years after treatment in several trees. Clearly, in trying to adapt oleander to pot growth these positive effects are of great interest and the longer lasting they are, the better. However, when our objectives are other, such as the hardening of oleander in the nursery to facilitate its transplantation and establishment in its final plot, it may be of interest that any effect should be short-lived (during the establishment phase, for exam-

ple) so that natural growth patterns can resume when the plant has become established. The lasting effects observed in our experiment may have been due to the growth retardants being added to the substrate since KARAGUZEL (1999) noticed that the effect of spraying paclobutrazol on Bougainvillea spectabilis was more short-lived that when it was added to the substrate. Whatever the case, it is not clear whether the effects noticed during the shading screen phase were due to the products or to the poor vegetative state of the treated plants (compared with the control) when they were first put behind the screen. Very probably, both aspects played some part but it is difficult know which was more important.

We conclude that the application of paclobutrazol or ethephon can help in the adaptation of native oleander as a pot plant. One sole application of these growth regulators to the substrate had the desired effect on grown and development. The most suitable doses of paclobutrazol were 10 and 20 mg per plant and 1 and 2 g per plant of ethephon. One year after the treatment the effect of both ethephon and paclobutrazol was still manifest in internode growth, leaf blade area and aerial part dry weight. However, the alteration in leaf colour noticeable after four months did not endure to the following sprouting. Internode length was more sensitive to paclobutrazol that leaf growth was, while the opposite was the case with ethephon. The doses applied played an important part, the highest doses producing the longest lasting effect. We are at present engaged in attempting to ascertain whether the joint application of both growth retardants will produce a plant with the most interesting characteristics provide by paclobutrazol and ethephon individually: the increased lateral sprouting and reduced leaf size produced by ethephon, and the reduced height and darker leaves produced by paclobutrazol.

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