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ORIGINAL ARTICLE

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The effects of propagation environment and foliar area on the rooting physiology of *Cordia alliodora* (Ruiz & Pavon) Oken cuttings

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Abstract The effects of propagation microclimate and foliar area on the rooting of *Cordia alliodora* (Ruiz & Pavon) Oken cuttings were investigated using non-mist propagators with and without shade. Photosynthetic rates (P_n), stomatal conductance (g_s) and chlorophyll fluorescence ratio (*Fv/Fm*) of the cuttings were assessed during propagation. Pronounced differences in microclimate were recorded between treatments, with lower temperatures and vapour pressure deficit (VPD) under shade. During the first 8 days after insertion, P_n varied between 2.21 and 4.96 and 0.47–2.54 μmol CO_2 $m^{-2}s^{-1}$ in the shaded and unshaded propagators, respectively. In the unshaded propagator, *Fv/Fm* decreased to a minimum of 0.72 2 days after insertion, recovering thereafter. In two separate rooting experiments, rooting percentage was reduced by high irradiance in the 20 and 30 cm^2 leaf area treatments, but not in the 10 cm^2 treatment. P_n decreased with an increase in leaf area in both shaded and unshaded propagators. *Fv/Fm* also declined with increasing leaf area in the high irradiance treatment. PAR and P_n were positively correlated under shade ($r^2 = 0.51$) but negatively correlated in the unshaded treatment ($r^2 = 0.49$); maximum P_n values were recorded at a PAR of 400 μmol m^{-2} s^{-1}. No significant differences in g_s were found between treatments, values ranging between 130 and 194 mmol H_2O m^{-2} s^{-1}. Positive correlations were found between rooting percentage and mean *Fv/Fm*. These results indicate that rooting of *C. alliodora* cuttings is related to photosynthetic activity during propagation, which is itself influenced both by propagator microclimate and cutting leaf area.

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Introduction

Adventitious root development in leafy stem cuttings is influenced by a range of physiological processes, and by their interaction with environmental variables (Leakey et al. 1994). In particular, successful rooting has been attributed to the provision of an appropriate propagation environment to permit the cuttings to photosynthesize during propagation (Eliasson and Brunes 1980). However, there is surprisingly little evidence to indicate whether or not photosynthesis by cuttings actually takes place, or whether it positively infuences rooting (Davis 1988). Evidence from tropical tree species is primarily based on the effects of variation in foliar area on rooting. For example, in *Triplochiton scleroxylon*, *Lovoa trichilioides* and *Khaya ivorensis*, optimum leaf areas for rooting of 50 cm^2, 200 cm^2 and 20 cm^2 have been identified, respectively (Leakey 1985; Leakey et al. 1994). These results have been attributed to a reduction in photosynthesis above an optimum leaf size, resulting from increased water deficits (Leakey and Coutts 1989). An increase in dry mass of *Triplochiton scleroxylon* cuttings during propagation is further evidence that photosynthesis has taken place (Leakey and Coutts 1989). However, few measurements of photosynthesis have actually been made in cuttings during propagation.

The fact that a number of species can root successfully in the dark or as leafless hardwood cuttings (Davis and Potter 1981) indicates that post-severance photosynthesis is not an absolute requirement for rooting in all species. Such observations do not negate the possibility that photosynthesis may be important for adventitious root development in those species where it occurs (Davis 1988). However, as noted by Davis (1988), it is difficult to test this hypothesis, primarily because photosynthetic rate cannot be varied independently of other factors which influence rooting.

For example, although photosynthetic rate may be influenced by altering the ambient CO_2 concentration, water relations and auxin content of the cutting may also be affected (Morison and Gifford 1984).

In the current investigation, three experiments were undertaken to investigate the relationship between photosynthetic activity and rooting of leafy cuttings of *Cordia alliodora* (Ruiz & Pavon) Oken (Boraginaceae). Each of the experiments involved manipulation of both foliar area and the irradiance regime during propagation, in order to assess the interaction between these factors. In this investigation, a low technology non-mist propagator was used, which facilitates physiological measurement *in situ* as the cuttings are not continuously moistened by misting sprays (Leakey et al. 1990). *Cordia alliodora* is an important timber species native to continental tropical America, distributed from central Mexico to northern Argentina (Greaves and McCarter 1990). A genetic improvement programme with this species is currently in progress at CATIE, Costa Rica (Mesén et al. 1994).

Materials and methods

The experiments were carried out at the nursery of the Centro Agronómico Tropical de Investigación y Enseñanza (CATIE) in Turrialba, Costa Rica (9°54′ N Lat., 83°40′ W Long., 600 m above sea level). Non-mist propagators were constructed following the design of Leakey et al. (1990). In this propagation system, air humidity is maintained by the provision of a water table beneath the rooting medium, and by enclosing the cuttings in a sealed polythene box. Plants of *C. alliodora* were derived from open pollinated progeny of plus trees selected by the CATIE Tree Improvement Project in Costa Rica (Mesén et al. 1994). Original seedlings were grown 1 m apart in beds at the CATIE nursery, cut back regularly (when shoots reached approx. 0.5 m height) to maintain a supply of coppice shoots, which were used as a source of cuttings to build up clonal populations. Rooted cuttings were potted into black polythene bags (600 cm^3) containing a 1:1:1 mixture of forest soil, sand and organic compost, then weaned under shade and decreasing watering (from automatic mist irrigation to once daily watering) during a 2–3 week period. After this weaning period, cuttings were planted in beds beside the original stockplant, at a spacing of 20 cm×20 cm. The clonal plants were given fortnightly soil applications of a powder fertilizer (FERTICA, Puntarenas, Costa Rica) containing 10%N, 30%P and 10%K, at a rate of approximately 30 g per plant. The beds themselves were made up of the potting mixture described above. Mean annual rainfall in Turrialba is 2600 mm, with no month below 50 mm. In consequence, watering was not usually necessary, but the plants were watered to field capacity when there was no rain for 2 consecutive days (typically in January and February).

Experiment 1

Twenty single-node cuttings were collected from clone 53, five from each shoot. The soft apical nodes were discarded and the leaves at all other nodes trimmed to 30 cm^2, using paper templates. The cutting base was treated with 10 µl indole-3-butyric acid (IBA) in methanol solution at a concentration of 1.6% using a microsyringe, before inserting the cuttings in cleaned river-sand in non-mist propagators. Prior to inserting the cuttings, holes were made in the medium to a depth of 2 cm using a board with wooden pegs at regular spacing (5 cm×5 cm) and the medium pressed firmly around the cutting. The cuttings were assigned randomly to one of two propagators with or without shade. The shade treatment was produced by covering one of the propagators with a single layer of black plastic netting, which was in addition to the standard layer of netting located 2 m above the propagators. The unshaded propagator was placed under full sunlight, without either of the two layers of netting.

Measurements of net photosynthetic rate (P_n), stomatal conductance (g_s)and chlorophyll fluorescence (Fv/Fm) were taken daily *in situ* in all ten cuttings from each propagator, between 0900 hours and 1200 hours. For assessments of P_n and g_s an infrared gas analyser was used (LCA-3, Analytical Development, Hoddesdon, UK). Measurements of chlorophyll fluorescence were taken using a fluorescence meter (PSM Mark II, Bio Monitor S.C.I. AB, Sweden), set at a run-time of 25 s and a dark-adaptation period of 20 min. Measurements of leaf, air and substrate temperatures, relative humidity and irradiance inside the propagators were recorded for the duration of the experiment using a 21X Micrologger (Campbell Scientific, Loughborough, England). Air temperature was measured using thermocouples (Type K chromel-alumel; T.C., Uxbridge, UK), humidity using a thermistor probe (MP. 100 Rotronic probe, Campbell Scientific, Loughborough, UK), substrate temperature using a 107-thermistor probe (Campbell Scientific, Loughborough, UK) and irradiance using quantum sensors (Skye Instruments, Llandrindod Wells, UK, supplied by Campbell Scientific, Loughborough, UK). The logger was programmed to record each sensor every 10 s, and to calculate and store mean readings every 15 min. Sensors were cross-calibrated prior to use.

Experiment 2

Single-node cuttings (288) were collected from each of clones 22, 37 and 38, giving 864 cuttings in total, with 6 cuttings obtained per shoot. The soft apical nodes were discarded, then the leaf area of each cutting was trimmed to 10, 20 or 30 cm^2 using paper templates. Each cutting base was then treated immediately with IBA at a concentration of 1.6%, as described above. The cuttings were then set to root in sand in non-mist propagators, and were allocated in node order to one of two propagators with or without shade in eight randomized blocks. The shaded and unshaded treatments were as in experiment 1 above. During the course of the experiment, cuttings were finely sprayed twice a day to keep the leaves moist, at 0700 hours and 1500 hours. After 2 weeks the cutting length and midpoint diameter were measured, and each cutting lifted and assessed for number of roots formed, after which cuttings were returned to the propagators. Similar assessments were carried out for the next 7 consecutive weeks. The environmental conditions inside the propagators were monitored throughout the duration of the experiment using a 21X Micrologger (Campbell Scientific, Loughborough, England) and associated sensors, as described in experiment 1.

Experiment 3

Single-node cuttings (648) were collected from clones 2, 6 and 37, 216 cuttings from each clone, with 6 cuttings obtained per shoot, after discarding the soft apical node of each shoot. The cuttings were prepared as described above and were set to root in sand in two non-mist propagators, with or without shade, in six randomized blocks. The shaded and unshaded treatments were as described in experiment 1 above. Measurements of chlorophyll fluorescence were taken at weeks 1, 2 and 3 on 36 randomly selected cuttings, 6 from each irradiance-foliar area combination, using a fluorescence meter (PSM, Biomonitor S.C.I. AB, Sweden) as described above. Measurements of net photosynthetic rate (P_n) and stomatal conductance (g_s) were taken at weeks 2, 3, 4 and 5 on the same cuttings, using an infra-red gas analyser attached to a Parkinson leaf chamber (Analytical Development, Hoddesdon, UK). These measurements were taken between 0900 hours and 1200 hours.

Statistical analysis

Analyses of variance (ANOVA) were carried out on percentage rooting values, which were calculated by each environment-clone-leaf area

treatment combination, based on a total of 48 cuttings in experiment 2 and 36 cuttings in experiment 3. Percentage data were transformed by the formula arcsin √% prior to ANOVA. Treatment differences were examined by multiple range tests using SAS (1980). For comparison of clones, cuttings from all node positions within each treatment were pooled. Chlorophyll fluorescence data collected in experiment 3 were also subjected to ANOVA. Analyses of deviance for stepwise regression in GENSTAT 5 (Payne et al. 1987) were used to determine the influence of treatment, node position and morphological characteristics of the cuttings on their rooting ability. The influence of treatment and node position on the photosynthetic rate and stomatal conductance of the cuttings were analysed by stepwise analysis of variance in GENSTAT 5 (Payne et al. 1987). The relationship between a number of variables was assessed by correlation; where correlations were significant at $P < 0.05$, correlation coefficients (r^2) are given. Full results of statistical analyses, including ANOVA tables, are presented in Mesén (1993).

Results

Experiment 1

Pronounced differences in microclimate were recorded between the two propagators during the period of observation, with higher relative humidity and lower air, leaf and substrate temperatures, PAR and leaf-to-air vapour pressure deficit (VPD) in the shaded propagator (Fig. 1a,b; Table 1a). Strong positive correlations were recorded between PAR and VPD in both propagators ($r^2 = 0.64$ and 0.88 for shaded and unshaded propagators respectively).

Photosynthetic rates of 2.21–4.96 μmol CO_2 m^{-2} s^{-1} and 0.47–2.54 μmol CO_2 m^{-2} s^{-1} were recorded in the cuttings in the shaded and unshaded propagators respectively. Values were generally higher in the shaded propagator, except at day 5 when P_n was similar in both propagators (Fig. 2a). Stomatal conductance varied little from day to day in the unshaded propagator (Fig. 2b). Higher values of g_s were recorded in the shaded propagator, following a similar pattern to that of P_n. When values of PAR were correlated with P_n, clear differences were found between both propagators. A strong positive correlation was found in the shaded propagator ($r^2 = 0.68$), while a negative correlation was found in the unshaded propagator ($r^2 = 0.54$).

The chlorophyll fluorescence ratio (*Fv/Fm*) of the cuttings was generally higher in the shaded propagator, and varied little from day to day (0.78–0.81). In the unshaded propagator, *Fv/Fm* decreased to the lowest values recorded (0.72) two days after insertion of the cuttings, but the values increased by day 5 and remained high until the end of the experiment, when there were no significant differences in *Fv/Fm* between both propagators (Fig. 2c).

Experiment 2

The use of shade above one of the propagators produced clear differences in microclimate, with reductions in irra-

Table 1 The propagator microclimate in shaded and non-shaded non-mist propagators in the nursery at CATIE, Costa Rica

(a) Experiment 1, over the 7 days after insertion of *C. alliodora* cuttings

	Shaded		Non-shaded	
	Mean	Range	Mean	Range
Relative humidity (%)	92.0	72.9 –100	87.5	51.4 – 100
Air temperature (°C)	24.5	19.2 – 33.6	26.5	17.8 – 41.9
Substrate temperature (°C)	24.7	21.6 – 28.8	26.5	21.4 – 33.1
PAR (μmol m^{-2} s^{-1})	53	0 –638	159	0 –1638
Leaf temperature (°C)	26.2	19.9 – 39.4	27.3	19.2 – 48.0
VPD (kPa)	1.01	0.19– 5.92	1.24	0.35– 9.85

(b) Experiment 2, over a 7 week period

	Shaded		Non-shaded	
	Mean	Range	Mean	Range
Relative humidity (%)	98.9	96.0 –100	94.7	71.0 – 100
Air temperature (°C)	21.9	19.9 – 26.3	24.3	20.4 – 37.0
Substrate temperature (°C)	22.3	21.2 – 23.9	23.9	20.8 – 34.2
PAR (μmol m^{-2} s^{-1})	6	0 – 88	99	0 –1278
Leaf temperature (°C)	23.5	21.2 – 27.9	25.1	21.2 – 41.0
VPD (kPa)	0.49	0.30– 0.95	0.60	0.18– 4.91

(c) Experiment 3, over a 7 week period

	Shaded		Non-shaded	
	Mean	Range	Mean	Range
Relative humidity (%)	99.9	92.4 –100	94.8	60.0– 100
Air temperature (°C)	23.4	19.0 – 31.8	25.3	18.5– 43.4
Substrate temperature (°C)	23.4	20.4 – 29.0	25.6	20.9– 38.9
PAR (μmol m^{-2} s^{-1})	24	0 –339	106	0 –1460
Leaf temperature (°C)	24.9	20.2 – 32.1	26.7	19.6– 47.5
VPD (kPa)	0.43	0.01– 1.47	0.91	0.14– 8.36

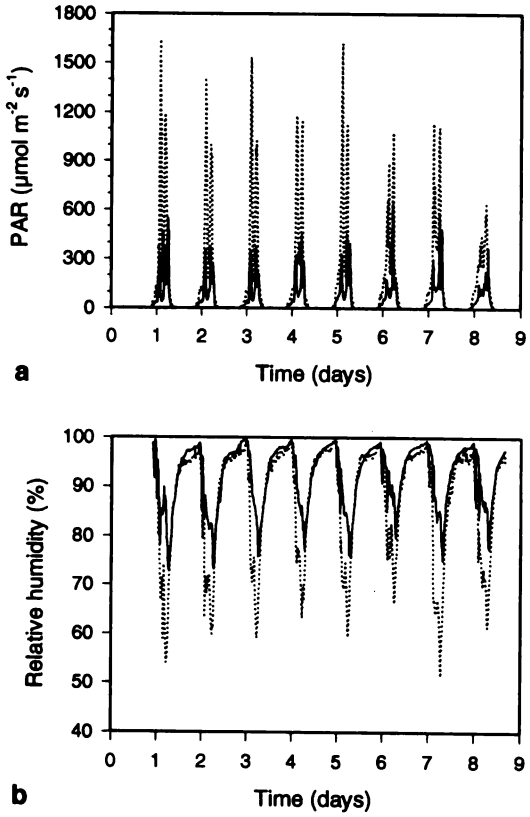

Fig. 1 Variations over an 8-day period in (**a**) photosynthetic active radiation (*PAR*), and (**b**) relative humidity inside non-mist propagators with shade (*solid line*) or without shade (*dotted line*) in experiment 1 (see text)

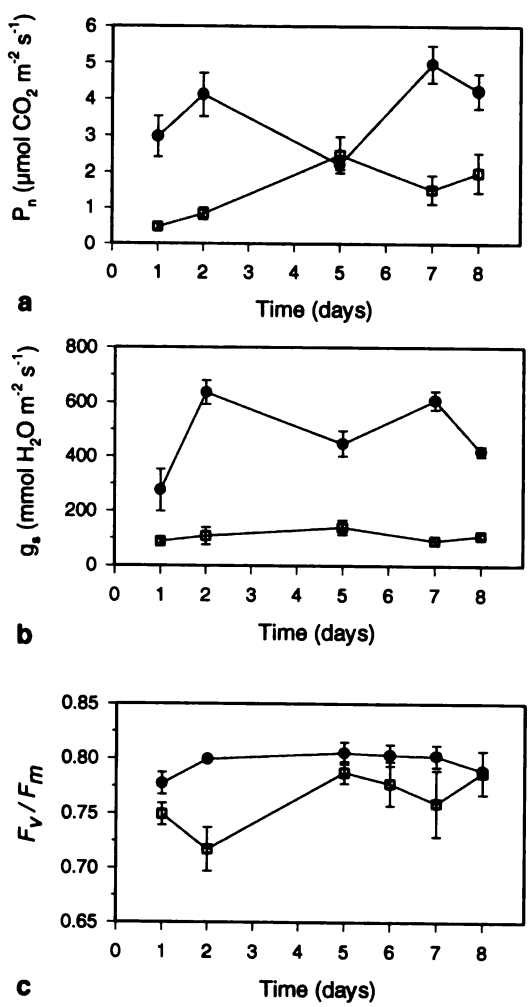

Fig. 2 The effects of two propagator environments on (**a**) the net photosynthetic rate (P_n), (**b**) the stomatal conductance (g_s) and (**c**) the chlorophyll fluorescence ratio (*Fv/Fm*) of single-node, leafy stem cuttings of *C. alliodora* set to root in non-mist propagators with shade (*filled circles*) or without shade (*empty squares*), in experiment 1. *Vertical bars*, SE

diance, VPD and air, foliar and substrate temperatures, and an increase in relative humidity in the shaded compared to the unshaded propagator (Table 1b). A strong positive correlation was found between PAR and VPD in the latter ($r^2 = 0.91$).

Rooting percentage was significantly ($P < 0.001$) affected by the interaction between propagation environment (PE) and leaf area, but not by these variables independently. In the shaded propagator, rooting after 7 weeks was significantly lower in the 10 cm² treatment, while cuttings with leaf areas of 20 cm² and 30 cm² rooted equally well. In the unshaded propagator, the highest rooting percentage was obtained in the 10 cm² treatment, which was significantly higher than in the 20 and 30 cm² treatments (Fig. 3a). There was less variation between treatments for mean number of roots per rooted cutting (Fig. 3b). Clones also displayed highly significant ($P < 0.001$) differences both in rooting percentage and in number of roots per rooted cutting at week 7 after insertion. Clone 22 displayed the highest mean rooting percentage (91.7%), followed by clone 38 (77.8%) and clone 37 (46.9%).

Highly significant ($P < 0.001$) differences were recorded both in rooting percentage and in number of roots per rooted cutting between node positions. Rooting percentage did not show any particular trend with respect to node position, varying from 63% for node 1 (apical) to 77% for node 3. The number of roots per rooted cutting showed a progressive increase with successive node positions down the stem, from 3.2 for node 1 (apical) to 4.7 for node 6.

When the results were analysed by stepwise regression, rooting was found to be significantly affected by clone, block, node position and cutting diameter, but was not significantly affected by propagation environment and the cutting length. Cuttings showed a progressive increase in diameter from the apical to the basal nodes, from 4.2 mm to 5.0 mm, while cutting length decreased from node 1 to node 2, then increased progressively up to node 6. A strong positive correlation was found between cutting diameter and mean number of roots per rooted cutting ($r^2 = 0.98$).

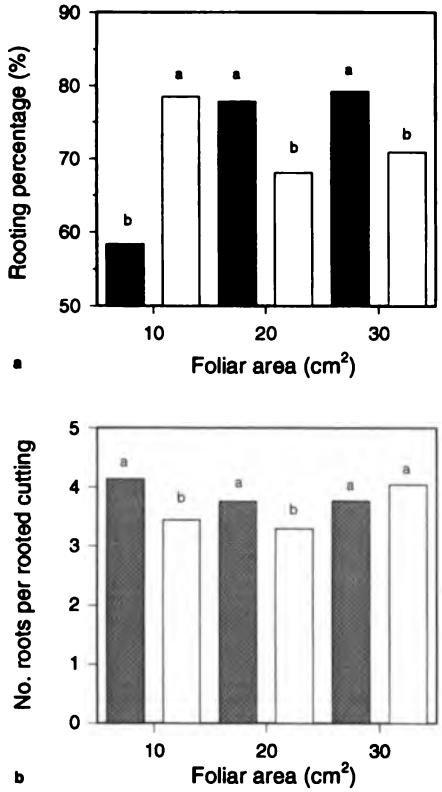

Fig. 3 The influence of propagation environment and cutting leaf area on (**a**) rooting percentage and (**b**) number of roots per rooted cutting of single-node, leafy stem cuttings of *C. alliodora* after 7 weeks in non-mist propagators with shade (*shaded bars*) or without shade (*open bars*); experiment 2 (see text). Data pooled from three clones and across all node positions; n = 144, means grouped by the same letter are not significantly different ($t_{0.05}$)

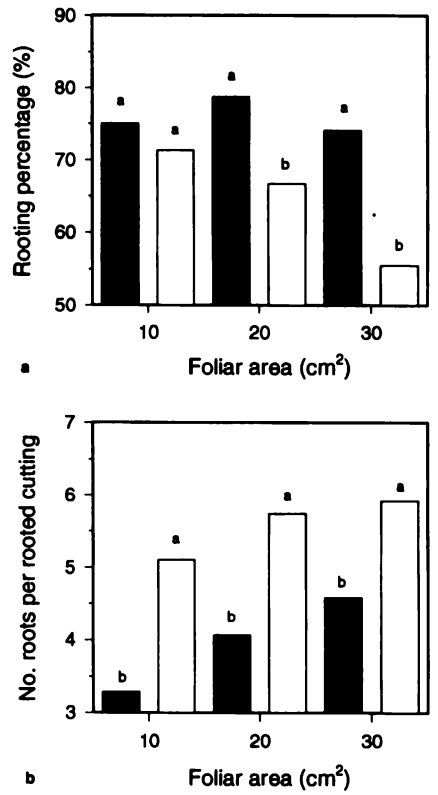

Fig. 4 The influence of propagation environment and cutting leaf area on (**a**) rooting percentage and (**b**) number of roots per rooted cutting of single-node, leafy stem cuttings of *C. alliodora* after 7 weeks in non-mist propagators with shade (*shaded bars*) or without shade (*open bars*); experiment 3 (see text). n = 108; means grouped by the same letter are not significantly different ($t_{0.05}$)

Experiment 3

As in the first and second experiments, pronounced differences were recorded between propagators for irradiance, leaf, air and foliar temperatures, relative humidity and VPD (Table 1c). However, higher irradiance and temperatures were recorded in both propagators compared to the second experiment. When values of PAR were correlated with VPD, strong positive correlations were found in both propagators (r^2 = 0.84 and r^2 = 0.93 for the shaded and unshaded propagator respectively).

Rooting percentage after 7 weeks was significantly higher ($P < 0.001$) in the shaded propagator, with no differences between the leaf area treatments. In the unshaded propagator, rooting percentage decreased with an increase in leaf area (Fig. 4a). In both propagation environments, the number of roots per rooted cutting increased with an increase in leaf area (Fig. 4b). Significantly fewer roots per rooted cutting were produced in the shaded propagator averaging over all leaf area treatments ($P < 0.001$).

Clones also showed highly significant ($P < 0.001$) differences both in rooting percentage and mean number of roots per rooted cutting, with a similar ranking (clone 6 > clone 2 > clone 37) for both variables. At the end of 7 weeks rooting percentage was 83.8%, 71.3% and 55.6%, and number of roots per rooted cutting 5.5, 5.2 and 3.0 for clones 6, 2 and 37 respectively.

The most apical node showed the lowest rooting percentage (57.4%), with no significant differences between the other node positions, with values between 70% for node 2 to 76% for node 6. In terms of number of roots per rooted cutting, there was an increase from the apical to the basal node (from 3.9 for node 1 to 5.5 for node 6).

When the results were analysed by stepwise regression, rooting percentage was found to be highly dependent on propagation environment, clone, cutting leaf area and the node position within the stem, but was not affected by the mean cutting diameter. The number of roots per rooted cutting, on the other hand, was strongly affected by cutting diameter. Cutting diameter showed a progressive increase from the apical to the basal nodes (from 3.8 mm for node 1 to 5.7 mm for node 6), and when these values were correlated with mean number of roots per rooted cutting, a strong positive correlation was found (r^2 = 0.99).

Mean photosynthetic rate (P_n) per unit leaf area was significantly higher ($P < 0.001$) in the unshaded propagator. Within each environment, P_n decreased with an in-

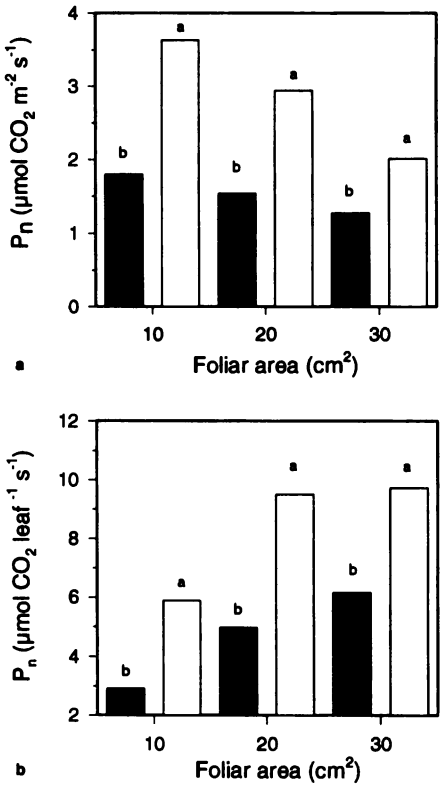

Fig. 5 The effect of propagator environment and leaf area treatment on (**a**) the net photosynthetic rate per unit leaf area and (**b**) the net photosynthetic rate leaf^{-1} of single-node, leafy stem cuttings of *C. alliodora*, set to root in non-mist propagators with shade (*shaded bars*) or without shade (*open bars*), experiment 3 (see text). n = 24–26. Values grouped by the same letter are not significantly different

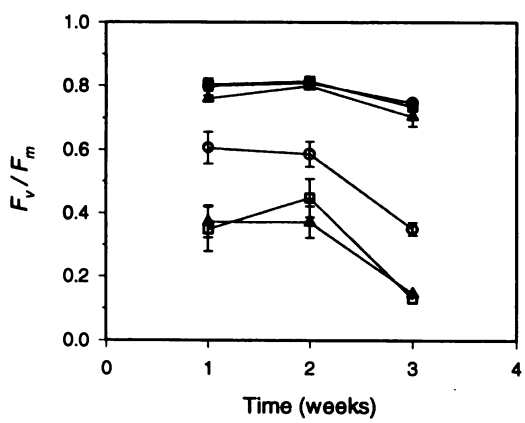

Fig. 6 The influence of two propagation environments and cutting leaf area (10 cm^2 = *circles*; 20 cm^2 = *squares*; 30 cm^2 = *triangles*) on the chlorophyll fluorescence ratio (*Fv/Fm*) in single-node, leafy stem cuttings of *C. alliodora* set to root in non-mist propagators with shade (*closed symbols*) or without shade (*open symbols*); experiment 3 (see text). n = 6, *vertical* bars, SE

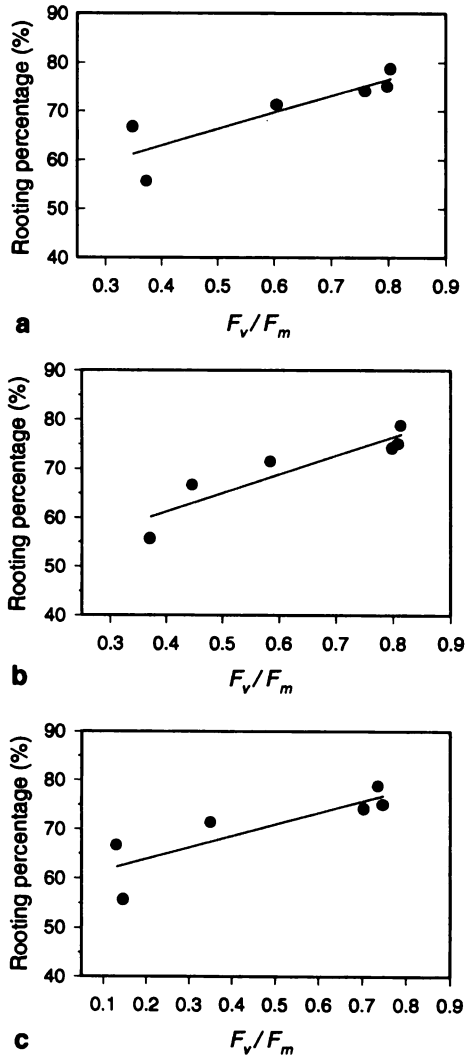

Fig. 7 The relationship between rooting of single-node, leafy stem cuttings of *C. alliodora* and the ratio of chlorophyll fluorescence (*Fv/Fm*) at (**a**) week 1 (y = 49.246 + 34.126 x; r^2 = 0.87), (**b**) week 2 (y = 46.080 + 37.888 x; r^2 = 0.92) and (**c**) week 3 (y = 59.197 + 23.549 x; r^2 = 0.85); experiment 3. Each value for rooting is the mean of 108 cuttings, pooling cuttings from the three clones and across all node positions

crease in leaf area (Fig. 5a). When the results were expressed as μmol CO_2 leaf^{-1}, mean P_n in both environments increased with successive increases in leaf area, although the difference between the 20 cm^2 and 30 cm^2 treatments was not significant (Fig. 5b). When values of PAR were correlated with P_n, a strong positive relationship was found between these variables in the shaded propagator (r^2 = 0.51), while a negative relationship was found in the unshaded propagator (r^2 = 0.49). No significant differences in g_s were found between treatments, with values between 130 mmol H_2O m^{-2} s^{-1} and 194 mmol H_2O m^{-2} s^{-1} for all treatments. When the results were analysed by stepwise regression, P_n was found to be highly dependent on both PE and leaf area.

A highly significant (P < 0.001) difference in chlorophyll fluorescence ratio (*Fv/Fm*) was recorded between PE treatments, and a highly significant (P < 0.001) interaction was recorded for PE×leaf area. In the shaded

propagator, *Fv/Fm* remained roughly constant from week 1 to week 3, with no significant differences between the three leaf area treatments. In the unshaded propagator, the 10 cm^2 showed significantly higher values than the 20 cm^2 and 30 cm^2 treatments, but all of the three leaf areas showed lower values than those of the shaded propagator. After 3 weeks there was a marked decrease in *Fv/Fm* in the unshaded propagator for all three leaf area treatments (Fig. 6). Mean values over the 3 weeks for the three leaf areas (10 cm^2, 20 cm^2 and 30 cm^2) were 0.79, 0.79 and 0.76 for the shaded propagator and 0.51, 0.36 and 0.33 for the unshaded propagator, respectively. When the results of *Fv/Fm* were correlated with final rooting percentage (week 6), strong positive correlations were found between rooting percentage and mean *Fv/Fm* at weeks 1, 2 and 3 (Fig. 7a–c).

Discussion

The results from experiment 1 indicate clearly that cuttings undergo a physiological shock immediately after insertion, and that this shock is more severe under higher irradiance. The low values of P_n (< 1 μmol $m^{-2}s^{-1}$), recorded during the first 2 days after insertion in the unshaded propagator, may be largely attributable to the onset of water deficits in the cuttings, as illustrated by the low (< 100 mmol $m^{-2}s^{-1}$) values of g_s recorded concurrently. However, the relatively low values of *Fv/Fm* recorded at this time suggest that photosynthetic efficiency may also have been impaired (cf. Bolhar-Nordenkampf et al. 1989), which may reflect the onset of photoinhibition (Kamaluddin and Grace 1992). The suggestion that cuttings may be susceptible to photoinhibition following insertion (Eliasson and Brunes 1980) has apparently not been tested previously (Davis 1988). Interestingly, although g_s remained low throughout the period of measurement, values of P_n and *Fv/Fm* tended to increase with time, indicating that the cuttings were able to photosynthetically acclimate to the higher irradiance environment within the first eight days after insertion (cf. Kamaluddin and Grace 1992).

Few other studies have measured photosynthetic activity of cuttings following insertion. Under a PAR of 280 μmol $m^{-2}s^{-1}$, P_n of pea cuttings was found to decline continuously for the first 6 days after excision, thereafter increasing to values higher than those recorded initially, concurrent with root emergence (Davis and Potter 1981). Newton and Jones (1993b) noted the occurrence of pronounced water deficits in cuttings of four tropical tree species after insertion in a non-mist propagator similar to that used here. Although water potentials below –2.0 MPa were recorded together with g_s values as low as 40 mmol $m^{-2}s^{-1}$ (0.1 cms^{-1}) within the first week after insertion, values thereafter tended to increase, even prior to root emergence (Newton and Jones 1993b). The importance of this acclimatization for root development is unclear; although rooting may be influenced by cutting water status (Loach 1977), no clear relationship between the onset of water deficits and eventual rooting was recorded either by Newton and Jones (1993b) or Grange and Loach (1984).

The results of both experiments 1 and 3 indicate clearly that cuttings of *C. alliodora* are able to photosynthesize during propagation prior to root formation. In this respect these results support those obtained with *Terminalia spinosa*, where P_n rates of up to 6 μmol $m^{-2}s^{-1}$ were recorded in a similar propagation system (Newton et al. 1992). The relationship between photosynthetic rate and rooting performance is, however, less clear. In experiment 3, although higher photosynthetic rates were recorded in the high irradiance treatment, rooting percentage was either unaffected or reduced by irradiance, depending on leaf area. In contrast, the number of roots per rooted cutting was higher in the unshaded treatment; root number was closely related to P_n when the latter was calculated on a total leaf basis. The results from this experiment indicate that photosynthetic efficiency, as indicated by *Fv/Fm*, is more closely related to rooting percentage than P_n measured by gas exchange techniques. This may reflect the importance of high photosynthetic efficiency for light capture in a low irradiance environment; as noted by Davis (1988), even low rates of P_n can significantly influence the carbon balance of a cutting. We are unaware of previous measurements of *Fv/Fm* in cuttings; these results suggest that such measurements may provide a more sensitive indication of the photosynthetic activity of cuttings than more traditional gas exchange techniques.

One of the most striking features of these results was the interactive effect of leaf area and irradiance recorded in both experiments 2 and 3. Rooting percentage was reduced by high irradiance in the 20 and 30 cm^2 treatments, but increased by high irradiance in the 10 cm^2 treatment. These results are consistent with the suggestion that variation in leaf area influences rooting through a balance between the processes of transpiration and photosynthesis (Okoro and Grace 1976; Eliasson and Brunes 1980). The reduction in rooting in the 20 and 30 cm^2 treatments under high irradiance may be attributed to the occurrence of more pronounced water deficits, which limit carbon fixation. The smaller-leaved cuttings, which are less prone to water deficits, would be more dependent on high irradiance to achieve a positive carbon balance. The differences between the two experiments may be attributed at least in part to the higher irradiances recorded in experiment 3, which may also explain the reduced treatment effect on the 10 cm^2 cuttings. The reduction in P_n and *Fv/Fm* with increasing leaf area, recorded in experiment 3, is also consistent with the above hypothesis. The lack of a consistent relationship between either node position or cutting diameter and rooting percentage is also consistent with post-severance rather than pre-severance carbohydrate production being important for rooting of this species (cf. Leakey et al. 1994).

A number of other studies have varied irradiance during the rooting period, with contrasting results. For example, Eliasson (1978) and Davis and Potter (1981) both recorded increased rooting with increased irradiance, suggesting that rooting was enhanced by photosynthesis. In contrast, Loach and Gay (1979) found that cuttings of *Forsythia* and

Weigelia rooted less well at higher irradiances, which was attributed to carbohydrate accumulation at high irradiance having a negative effect on photosynthetic rate. However, as noted by Davis (1988), higher irradiance may have other physiological effects which may influence rooting, such as increasing the transport of auxin to the base of the cutting by elevating leaf temperature (Baadsmand and Andersen 1984). The higher leaf temperatures recorded here under high irradiance may also have been associated with higher tissue respiration rates, which would tend to reduce the rate of carbon fixation. Higher irradiance may also influence foliar water potentials, and thereby a range of physiological processes (Loach and Gay 1979).

In both experiments 1 and 3, positive correlations were recorded between PAR and P_n under shade, whereas without shade, the correlations were negative. This indicates that the highest values of P_n in cuttings of *C. alliodora* occur at relatively low irradiances of around 400 µmol $m^{-2}s^{-1}$, a value that compares closely with those obtained for *Pisum sativum* (Davis and Potter 1981) and a range of ornamental shrub species (Machida et al. 1977). The decline in P_n at higher irradiances, presumably reflecting the onset of water deficits (Davis 1988), emphasizes the importance of managing propagator microclimate if optimum rooting is to be achieved (Newton and Jones 1993a).

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References

Baadsmand S, Andersen AS (1984) Transport and accumulation of indole-3-acetic acid in pea cuttings under two levels of irradiance. Physiol Plant 61: 107–113

Bolhar-Nordenkampf HR, Long SP, Baker NR, Öquist G, Schreiber U, Lechner EG (1989) Chlorophyll fluorescence as a probe of the photosynthesis competence of leaves in the field: a review of current instrumentation. Funct Ecol 3: 497–514

Davis T (1988) Photosynthesis during adventitious rooting. In: Davis TD, Haissig BE, Sankhla N (eds) Adventitious root formation in cuttings. Dioscorides Press, Portland, Oregon, pp 79–87

Davis T, Potter JR (1981) Current photosynthesis as a limiting factor in adventitious root formation on leafy pea cuttings. J Am Soc Hort Sci 106: 278–282

Eliasson L (1978) Effects of nutrients and light on growth and root formation in *Pisum sativum* cuttings. Physiol Plant 43: 13–18

Eliasson L, Brunes L (1980) Light effects on root formation in aspen and willow cuttings. Physiol Plant 48: 261–265

Grange RI, Loach K (1984) Comparative rooting of eighty-one species of leafy cuttings in open and polyethylene-enclosed mist systems. J Hortic Sci 59: 15–22

Greaves A, McCarter PS (1990) *Cordia alliodora* – a promising tree for tropical agroforestry. Tropical Forestry Papers No. 22, Oxford Forestry Institute, Department of Plant Sciences, University of Oxford

Kamaluddin M, Grace J (1992) Photoinhibition and light acclimation in seedlings of *Bischofia javanica*, a tropical forest tree from Asia. Ann Bot 69: 47–52

Leakey RRB (1985) The capacity for vegetative propagation in trees. In: Cannell MGR, Jackson JE (eds) Attributes of trees as crop plants. Institute of Terrestrial Ecology, Abbots Ripton, Huntingdon, pp 110–133

Leakey RRB, Coutts MP (1989) The dynamics of rooting in *Triplochiton scleroxylon* K. Schum. cuttings: their relation to leaf areas, node position, dry weight accumulation, leaf water potential and carbohydrate composition. Tree Physiol 5: 135–146

Leakey RRB, Mesén JF, Tchoundjeu Z, Longman KA, Dick JMcP, Newton A, Matin A, Grace J, Munro RC, Muthoka PN (1990) Low-technology techniques for the vegetative propagation of tropical trees. Commonw For Rev 69: 247–257

Leakey RRB, Newton AC, Dick JMcP (1994) Capture of genetic variation by vegetative propagation: processes determining success. In: Leakey RRB, Newton AC (eds) Tropical trees: the potential for domestication and the rebuilding of forest resources. HMSO, London, pp 72–83

Loach K (1977) Leaf water potential and rooting of cuttings under mist and polythene. Physiol Plant 40: 191–197

Loach K, Gay AP (1979) The light requirement for propagating hardy ornamental species from leafy cuttings. Sci Hortic 10: 217–230

Machida H, Ooshi A, Hosoi T, Komatsu H, Kamota F (1977) Studies on photosynthesis in cuttings during propagation. I. Changes in the rate of apparent photosynthesis in the cuttings of several plants after planting. J Jpn Soc Hortic Sci 46: 274–282

Mesén JF (1993) Vegetative propagation of central American hardwoods. PhD Thesis, University of Edinburgh

Mesén JF, Boshier DH, Cornelius JP (1994) Genetic improvement of trees in Central America with particular reference to Costa Rica. In: Leakey RRB, Newton AC (eds) Tropical trees: the potential for domestication and the rebuilding of forest resources. HMSO, London, pp 249–255

Morison JIL, Gifford RM (1984) Plant growth and water use with limited water supply in high CO_2 concentrations. I. Leaf area, water use and transpiration. Aust J Plant Physiol 11: 361–374

Newton AC, Jones AC (1993a) Characterisation of microclimate of mist and non-mist propagation systems. J Hortic Sci 68: 421–430

Newton AC, Jones AC (1993b) The water status of leafy cuttings of four tropical tree species during propagation. J Hortic Sci 68: 653–663

Newton AC, Muthoka P, Dick JMcP (1992) The influence of leaf area on the rooting physiology of leafy stem cuttings of *Terminalia spinosa* Engl. Trees 6: 210–215

Okoro OO, Grace J (1976) The physiology of rooting *Populus* cuttings. I. Carbohydrates and photosynthesis. Physiol Plant 36: 133–138

Payne RW, Lane PW, Ainsley AE, Bicknell KE, Digby PGN, Harding SA, Leech PK, Simpson HR, Todd AD, Verrier PJ, White RB (1987) GENSTAT 5: reference manual. Clarendon Press, Oxford

SAS (1980) SAS User's guide: statistics, version 5 edition. SAS Institute, Cary, N. C., pp 113–137