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CONTENTS

PRACTICAL SECTION FOR GROWERS

This project explored the extent to which the performance of HNS in containers can be restricted by various stresses associated with the 'unnatural' environment to which the roots are exposed. In a container, the root system is uniquely accessible to visual inspection. This greatly increases our ability to recognise situations in which shoot growth has suffered from adverse environmental conditions around the roots. This project generated information to help growers and advisors interpret what they see when a plant is 'knocked out' of its container.

The project required systematic scientific study of the relationships between conditions in the container, the appearance of the root systems, and the growth and quality of the above ground parts of the plants. Plants were grown under conditions designed to impose stresses of various sorts. In some experiments very severe stresses were applied to elicit clear visual symptoms. In others, the treatments were designed to mimic less extreme conditions such as are likely to be encountered on a commercial container unit. As soon as the root system was sufficiently well developed to be able to remove the root-ball from the pot, systematic observations were made of the development of visible roots. At the end of the growing season, the effects on plant performance as a whole were measured.

Mainly we used *Cotinus coggygria* 'Royal Purple', *Garrya elliptica* 'James Roof', *Acer palmatum* 'Aureum', *Daphne* x *burkwoodii* 'Somerset', *Elaeagnus pungens* 'Maculata', and *Viburnum tinus* 'Eve Price'. Nine other species and varieties were also included in observational trials.

To improve the objectivity of the measurements of the amount and distribution of roots, a system was devised for dividing the surface of the root-ball into 13 zones, within each of which the percentage of the growing medium covered by roots was estimated by eye. The zones were small enough to take in at a glance so that estimates could be made quickly and reliably.

Under-watering

Uneven distribution of overhead irrigation, combined with large differences in water requirement between crops within a single irrigation area, makes it difficult to avoid some plants being under-watered while others are over-watered. We examined the effects of a short-term severe water shortage and also of chronic under-watering for a period of ten weeks.

In all subjects, water shortage tended to inhibit shoot extension strongly and also reduced the total amount of root produced. When the under-watering was chronic, roots tended to become concentrated in the lower third of the container so that root cover over the base was often extensive, sometimes more than that in the well-watered control plants. Roots tended to grow downwards more or less vertically and with minimal branching, whereas roots on well-watered plants tended to grow round the circumference and to be more branched. Water shortage also suppressed roots more on the south than on the north side of the pot. This suggests that higher temperatures on the side of the pot facing the sun were inhibiting root development but results of controlled temperature experiments suggest that it was more likely to be an indirect effect on the distribution of water within the container.

Over-watering

For containers on gravel, or any other non-capillary drainage surface, over-watering creates a saturated zone at the base of the pot which carries a risk of damage to the plants due to shortage of oxygen. In practice, this is often exacerbated by localised puddling on some parts of the bed. Similar problems can occur on a capillary irrigated bed when the level of water in the supply tanks is kept too high.

In relation to acute stress, we examined the response to standing the containers in water to a depth of 10cm for 72 hours on three separate occasions. This caused extensive death of roots within the saturated zone and 50% of *Garrya elliptica* 'James Roof' plants died. In contrast, there were no losses amongst *Cotinus coggygria* 'Royal Purple' plants and there was no detectable reduction of shoot growth despite the amount of root being halved. Most roots in the waterlogged zone became grey or blackened and died, and similar symptoms were seen in observational trials with other plants. There was also a tendency for the roots to be flattened against the walls of the containers and to develop a distinctive striated appearance or 'tram lines' symptom.

Repeated waterlogging to a depth of 2 cm for part of each day produced similar symptoms on the few roots present in the waterlogged layer but the main effect was to reduce the amount of root in the lower third of the pot, and particularly across the base. However, in all subjects except *Berberis stenophylla*, there was a corresponding increase in the amount of root in the well-aerated upper layers. A severe effect on the growth of shoots could arise if such waterlogging were to occur late in the season, particularly if existing roots were concentrated near the base of the pot. This highlights the need to avoid wild fluctuations in irrigation and the drainage regime.

'Salt' stress

Roots exposed to three times the recommend amount of controlled release fertiliser (CRF), or higher, developed symptoms that were strikingly similar to those associated with waterlogging but less consistent. Roots also tended to be flattened against the container wall and to have the 'tram-lines' symptom referred to above.

At lower amounts of CRF, convincing evidence of salt damage to roots was not observed in any subject, even though experiments were conducted on capillary beds so as to minimise leaching. Damage to roots was not observed when they grew through a localised high nutrient band in glass-sided root-observation boxes, despite concentrations of CRF up to 36 kg m^{-3}. Even where roots had grown into a cluster of CRF granules within this band, no visible symptoms of any damage were not observed.

Measurements of electrical conductivity (EC) in the various CRF treatments showed that many factors were operating to alter the degree of salt stress in unexpected ways. For example, in one experiment, large differences which were related to leaf area were observed between species. This strongly suggested that nutrients from the capillary bed were concentrated in the containers of the plants that were taking up the most water. The salts could have come from a previous crop or from adjacent plants or may even have been in the water supply itself. As water is always evaporating from a capillary bed, any impurities in the supply will tend to become concentrated. With the increased emphasis on the need to recirculate water, this sort of problem is likely to increase. Thus, while no grower would intentionally use CRF at three times the recommended amount, an equivalent salt concentration could readily accumulate if there were substantial amounts of salts in the irrigation water.

These results demonstrate the complexity of the processes controlling the accumulation of salts in container media. The topic deserves much more detailed investigation, especially as salt-stress is probably involved in some way in the development of the leaf-tip necrosis that is a common feature of poor plant performance in containers. The availability of a new sensor (from Delta-T Devices) which allows EC to be measured rapidly *in situ* should open up opportunities for growers to monitor this factor more effectively but the usefulness of the sensor was not examined in this project.

High root temperature

Measurements in containers at East Malling, in summer 1996, showed that the medium could reach close to 50 °C briefly in the middle of the day and could exceed 40 °C for more than about 5 hours. To determine whether such temperatures, alone, are sufficient to cause significant injury to plants, apparatus was developed to simulate the natural diurnal heating cycle in containers, using radiant electric heaters to mimic the effect of solar radiation.

Roots given a single exposure to peak temperatures more than 38 °C showed marked discoloration, especially in *Garrya*, but there was no evidence that this damage had any adverse effect on the growth or quality of the above ground parts of the plants. In the final year of the project, an improved version of the apparatus was used to impose repeated exposure to the most extreme conditions likely to be encountered in the UK. Almost half of the visible roots of *Berberis stenophylla* and *Elaeagnus pungens* 'Maculata' turned dark brown but again there was no significant adverse effect on shoot growth or leaf condition.

Leakage of electrolytes confirmed that the blackened roots were dead, so the absence of substantial effects on the shoots indicates that the plants in this experiment had more root than was necessary to sustain shoot growth. This is probably often the case for plants in containers with high levels of nutrients and generous watering. Damaging effects from high root temperature are only likely to occur when the root system is relatively small, perhaps because of damage from other stresses, and/or the medium being deficient in nutrients or water.

Other factors

No evidence was obtained to support the idea that the unnaturally high proportion of organic matter in modern media could underlie the problems with difficult crops such as Japanese maples and *Daphne*. Instead, nutrient and pH levels were shown to be critical for *Acer palmatum* and *Daphne* x *burkwoodii* respectively.

Conclusions and Action Points for Growers

- The appearance of roots seen when a plant is 'knocked out' of its container is useful in identifying the type of stress the plant has suffered. Adaptive changes in the distribution of roots over the surface of the root-ball can contribute to the diagnosis.

- A diagnostic key has been produced (Appendix 1) based on the root observations made in this project.

- A simple method for defining a series of 13 zones on the surface of the root-ball was developed to make visual assessment of the amount and distribution of roots more objective and reproducible. This could provide the basis for more effective exchange of information between researchers and growers.

- Consistent under-watering is indicated by a preponderance of roots near the base of pots and main roots tending to grow vertically.

- Intermittent waterlogging causes localised blackening of the roots and stimulates root growth in the upper layers. Despite this adaptive response, shoot growth can be reduced seriously in some subjects (e.g. *Berberis stenophylla*).

- If temperatures at the edge of the container facing the sun exceed about 40 °C, even if only briefly in the middle of the day, roots are likely to be visibly damaged. However, on its own, this is unlikely to substantially reduce shoot growth or induce symptoms such as leaf tip necrosis.

- If plants are under-watered, they may also show fewer roots on the southern sides of containers, especially at the edge of container beds. Whether this is because the medium dries out more rapidly on the warmer side of the container or is a direct effect of temperature on the roots, the application of additional irrigation around the edges of beds will help to counteract it.

- Electrolyte leakage can be a useful way to confirm that blackened or otherwise unhealthy looking roots are seriously dysfunctional.

- Capillary irrigation increases the risk of salt damage because, in addition to the absence of leaching, salts can accumulate in the bed itself.

- Further work is required to better understand the factors controlling salt concentrations in containers. This will be facilitated by the advent of Delta-T Devices' Sigma probe which allows EC to be measured *in situ*.

- High amounts of fertiliser (e.g. 6 g L^{-1} Ficote 180) should be avoided for *Acer palmatum*. Growth will benefit but plants are liable to suffer shoot damage during the following winter/spring.

- *Daphne* x *burkwoodii* 'Somerset' has a narrow pH tolerance range. The pH should be maintained between 6.0 and 7.0.

SCIENCE SECTION A. Relative sensitivity of roots to different components of the container environment

Introduction

The aim of this project was to explore the extent to which the performance of HNS in containers can be restricted by various stresses associated with the 'unnatural' environment to which the roots are exposed. This section of the report focuses on experiments in which the effects of many potential stress factors were compared on a representative range of commercially relevant HNS.

In a container, the root system is uniquely accessible to visual inspection. This greatly increases our ability to recognise situations in which shoot growth has suffered from adverse environmental conditions around the roots. However, interpretation of what is seen when a plant is 'knocked out' of its pot relies heavily on intuition because there is a dearth of information on which to base a more scientific approach. One of the aims of this part of the project was to start to accumulate this sort of information.

To achieve this aim, plants were grown under conditions designed to impose stresses of various sorts, in ways that were reproducible enough for successful experimentation but were also representative of the sorts of conditions which can occur on a commercial container unit. Detailed inspection of the roots, combined with measurements of the amount of root and shoot growth, was used to assess the relative effects of the various stress treatments. In the first year, the majority of treatments were short term but severe, representative of the sorts of conditions likely to occur only when there has been some failure of equipment or management. In the second year, repeated exposure to milder conditions was intended to simulate the sorts of suboptimal conditions that could be experienced by a small proportion of plants even on a well-managed nursery, for instance as a result of the uneven distribution of irrigation from sprinklers or impeded drainage in low-lying spots.

In the second year, the range of plants examined was extended, with greater emphasis being placed on visual assessment of responses in combination with photographic records of the appearance of typical root systems to provide a visual 'key' to the range of symptoms associated with each kind of stress.

The results from the first two years indicated that roots could be damaged by concentrations of controlled release fertiliser (CRF) 3 or 4 times above recommendations, but not by doubling the recommended amounts. In practice, insufficient mixing can result in localised accumulations of CRF granules and this might cause root damage. In the third year, this possibility was examined in transparent-sided 'root boxes'. This was complemented by other experiments that extended further the range of plants screened in relation to a range of acute stresses, including high salt concentration throughout the medium.

Materials and Methods

Year 1 (1996): responses to severe stress

Plant material

Subjects with contrasting root morphology were selected:

- *Cotinus coggygria* 'Royal Purple' - fine, dark brown roots
- *Garrya elliptica* 'James Roof', with thicker, white fleshy roots, often conspicuous root hairs

Plants were grown from cuttings, directly-stuck in August 1994 (*Cotinus*) and May 1995 (*Garrya*). In late February 1996 they were pruned to improve uniformity.

The treatments (with abbreviated title in brackets)

- Control ('Control') – plants maintained with minimal stress on a capillary sand bed in a medium of 70:30 peat:bark with 3 g l^{-1} (*Garrya*) or 4 g l^{-1} (*Cotinus*) Ficote 180, 16:10:10 + fritted trace elements (FTE) added.
- Single episode of drought stress ('Drought') - As Control until 6 August when the plants were moved from the capillary bed to gravel and thereafter received no water until the plants wilted severely (10 to 14 days), after which they were thoroughly rewetted and returned to the capillary bed. A small well-ventilated polythene structure acted as a rain shelter.
- Periodic severe waterlogging ('Waterlogged') - As Control until 6 August when the plants were placed in a water bath (10 cm deep) for 72 hours. This was repeated on 17 September and 19 October.
- Salt stress ('Salt') - As Control except that the concentration of CRF was increased three-fold. This included occasional overhead watering to supplement the capillary irrigation during hot weather; this would have limited the tendency for salts to accumulate near the surface of the medium.
- Low organic matter ('Low O.M.') - Based on a preliminary experiment to identify a suitable medium, the mixture consisted of 7% peat, 3% Cambark 100 and 90% fine-grade vermiculite (by volume). To this was added Ficote 180, 16:10:10 + FTE at the same concentrations as in the 'Control'. The purpose of this treatment was to investigate whether the unnaturally high organic matter content of modern growing media causes any problems, such as favouring saprophytic micro-organisms that might then increase the rate of turnover of fine roots.

Replication

There were 15 replicate plants per treatment.

Root inspection and other assessments

In addition to the 15 plants in normal containers, three plants per treatment were placed in special containers designed to facilitate root inspection early in the season. This consisted of a transparent plastic container placed in a conventional 2 litre pot, the gap between them being filled with additional medium to exclude light. The transparent container was removed periodically to inspect the roots that had reached the surface, without disturbance to the root system. Later in the season, when the root systems were sufficiently well developed to bind the medium, the roots in the conventional containers were inspected by knocking out the root-ball in the usual way. Root activity was quantified as the numbers of root tips present on the surface of the medium, which appeared to be growing (i.e. pale coloured, with a fresh and clean appearance).

Shoot length per plant was measured at the start and end of the experiment. Also, at the end of the experiment, half the plants were harvested to determine root dry weight and total root length (using a root scanner, Commonwealth Aircraft Corporation Ltd, Melbourne, Australia).

Physical measurements

The pH, electrical conductivity (EC) and air-filled porosity of the media were measured periodically. For pH and EC measurements, 3 x 7 ml sub-samples were bulked; any CRF granules were excluded and each subsample was diluted 1:6 w/v with purified water. Measurements were made after one hour. Samples were taken from three replicate plants per treatment on each occasion.

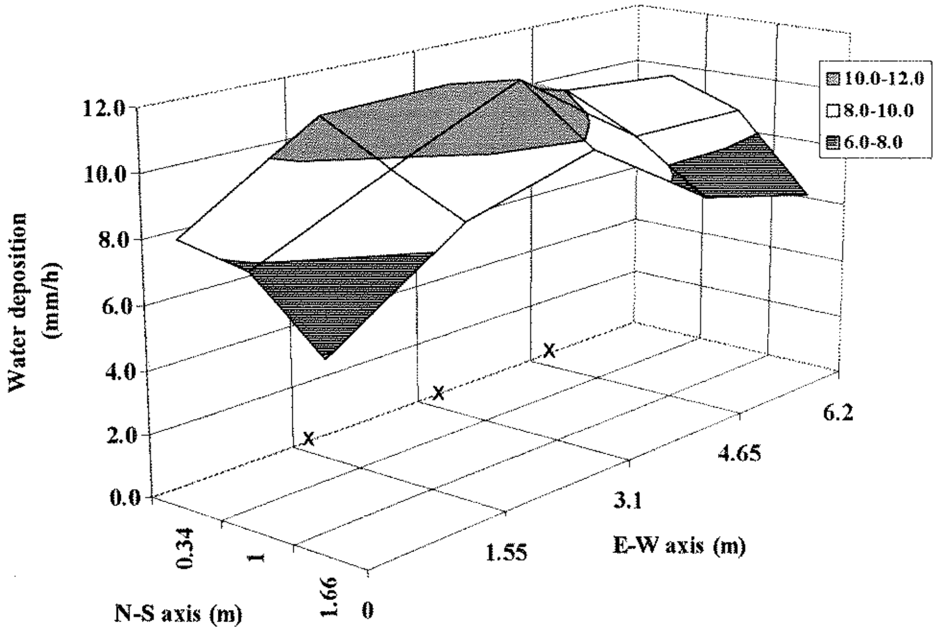
An *in situ* estimate of the percentage air-filled porosity of the media was made using containers with plants growing in them; there were three replicate plants per treatment. The medium was brought close to saturation by immersion in a water bath for one hour, after which it was drained into a measuring cylinder via a funnel. PVC tape placed over all but one of the drainage holes facilitated the transfer from the water bath to the funnel. The volume of water collected provided a measure of the volume of the pores, which would normally have been air-filled. This air-filled volume was expressed as a percentage of the total volume of the medium.

Year 2 (1997) – responses to prolonged moderate stress

Preliminary experiment on water deposition under overhead sprinklers

In a preliminary experiment, the variability in water deposition across the bed to be used for the main experiment was monitored by collecting in an array of beakers laid out over the bed. As is typical of this type of irrigation, Figure 1 shows that there was substantial variability in water deposition, despite data being collected on a quite still day and the bed being surrounded by a windbreak. Overall, there was a two-fold variation in water deposition, from 7.8 to 15.5 ml cm^{-2}. Under such situations, even if the *average* amount of irrigation applied exactly matches the plants' needs, containers in the middle will tend to be over-watered while those at the edge will be under-watered. Without positive drainage from a sand-bed, the over-watered containers are liable to suffer transient poor aeration towards the base. Therefore, even on well-managed nurseries, overhead irrigation may cause many plants to suffer repeated exposure to either short term waterlogging or mild water shortage. This provided the basis for the design of the stress treatments investigated in year 2.

Figure 1. Variation in water deposition over a bed irrigated by overhead sprinklers, under relatively wind free conditions (typical wind speed was 0.4 m s^{-1}). X marks the location of a sprinkler.



Plant material

In conjunction with the project co-ordinator, eight subjects were selected to provide a wide range of morphology and preferred habitat (Table 1). With the exception of *Cornus alba* 'Elegantissima' and *Viburnum tinus* 'Eve Price', all were propagated during August 1996 as directly-stuck softwood cuttings. The rooted cuttings were grown on in a 50:50 (v/v) peat / bark mix (with Ficote 180, 16:10:10 and fritted trace elements), over-wintered in 1½ litre containers under polythene, and potted up to 2 L in June using the amount of fertiliser shown in Table 1.

The *Viburnum* and *Cornus* were obtained from a commercial source in June 1997. Their shoots and roots were pruned lightly to improve uniformity and potted-on as above.

Table 1. A list of species and cultivars screened for root dysfunction in response to environmental stress. Amounts of Ficote 180 used in the water and salt treatments are shown in the two right-hand columns.

Plant subject	Amount of CRF (Ficote 180), g L^{-1}	
	'High Salt'	All other treatments
Cotinus coggygria 'Royal Purple'	12	6
Elaeagnus pungens 'Maculata'	12	6
Berberis stenophylla	12	6
Cornus alba 'Elegantissima'	12	6
Cornus alba 'Sibirica'	12	6
Viburnum tinus 'Eve Price'	12	6
Garrya elliptica 'James Roof'	6	3
Acer palmatum 'Aureum'	6	3

The treatments

- 'Overhead-control' - After potting up into 2 L containers in mid-June, plants were moved to a sprinkler irrigated bed, spaced at 22 x 22 cm, on a free draining substrate covered with Mypex, where they were irrigated twice daily for 40 minutes. The water distribution data shown in Figure 1 were used to minimise variation in irrigation applied to the experimental plants. Supplementary hand watering was applied as necessary.
- 'Overhead-wet' - This treatment simulated situations where frequent puddling occurs on container beds. From 25 July, the containers were placed in saucers (19 cm diameter x 3.5 cm deep) for a period of ten weeks. Two holes were drilled in the sides of each saucer, 2 cm above the base, to limit the depth of the 'puddle' to 2 cm. Excess water drained rapidly from the holes, so that variation in irrigation did not cause variation in the depth to which containers were waterlogged. After each automatic irrigation period (at 08:30 hrs and 16:30 hrs), supplementary hand watering was used to bring the water level up to the drainage holes where necessary (generally only on hot days and for the larger plants). At 12:30 p.m., any water not absorbed from the saucers by the plant was emptied, giving the roots a short period of time to recover before the next irrigation (15:50 hrs). Thus, the base of containers was waterlogged for a maximum of 21 hours per day.
- 'Overhead-dry' - Plants were maintained on a separate raised bed, identical to the one described above. The irrigation regime was as described for the controls until 25 July when the overhead irrigation was turned off. When plants began to show signs of water stress (i.e. wilting), they were watered manually with a hose. Additionally, they received any natural rainfall. This regime simulated the situation that can easily exist for containers in the relatively dry spots of the much larger overhead-irrigated container beds used in the industry.
- 'Capillary-Control' - Plants were maintained on an Efford-style capillary sand bed with occasional overhead watering by hand on extremely warm days.
- 'High-Salt' - Plants in this treatment were potted into medium containing twice the optimal amount of fertiliser (see Table 1) and were also placed on a capillary bed so that the nutrients would tend to accumulate in the containers rather than be leached out.

To allow the roots to establish and proliferate around the edges of their containers, the water treatments were not imposed until 25 July, after which plants were exposed to treatments for 10 weeks. Salt treatments commenced at the time of potting-on, when the fertiliser was added to the medium.

Measurements

Destructive measurement of root growth and distribution

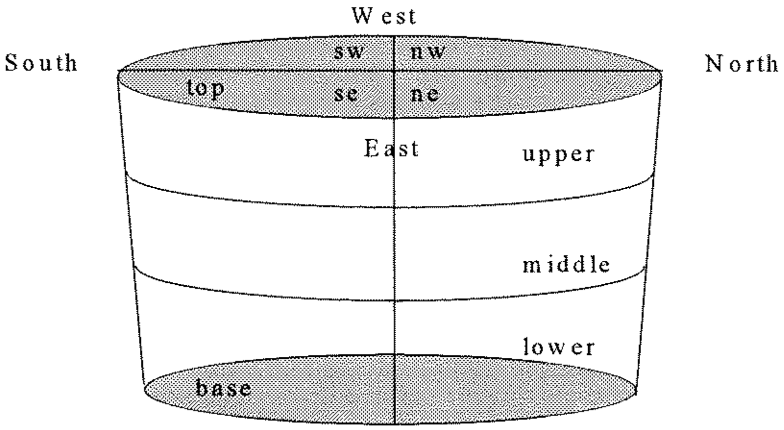
Detailed measurements of root weight and length, as well as of shoot growth, were made on two subjects (*Cotinus* and *Elaeagnus*) whilst all subjects were assessed visually for differences in root distribution and morphology (see below). Root dry weights were determined at the end of the experiment by destructively harvesting three plants from each treatment. For *Elaeagnus*, each plant was removed from the container and the root-ball was cut into three sections corresponding to the upper, middle and lower segments of the distribution system described below. With the *Cotinus* plants, the root systems were extracted intact because too much of the fine roots would have been lost in the root washing procedure if the root-ball had been segmented first. Root lengths were measured on a root scanner (Commonwealth Aircraft Corporation Ltd, Melbourne, Australia) before the roots were dried (three days at 80°C) and weighed. By dividing root weight by root length, a value relating to the average thickness of the roots was obtained.

Non-destructive assessment of root distribution

In addition to the detailed studies on *Cotinus* and *Elaeagnus* roots, the distribution of roots was quantified on all plants in the experiment using an alternative, non-destructive method designed to be relatively quick and easy and suitable for use by growers and their advisors. This method involved a systematic visual inspection of roots at the surface of the root-ball as seen after 'knocking out' from the container. It was applied to all plants in the experiment (48 plants per treatment; i.e. 8 subjects x 6 replicate plants) and yet took much less time than the destructive method described above.

Plants were removed from their containers and the surface of the root-ball was partitioned into three layers, each four centimetres deep, identified as the upper, middle and lower segments of the containers (Figure 2). These layers were further segmented at right angles to take into account the aspect of the container. Each of the resulting 12 zones was visually isolated using cardboard strips, and then the percentage of the surface covered by roots was estimated by eye in steps of 5%. The whole of the downward facing basal surface was treated as a single additional zone, making a total of 13 zones.

Figure 2. Diagrammatic representation of a two litre pot, partitioned to create segments for root zonation assessments.



Shoot growth

Shoot growth was recorded as the extension of the longest shoot per plant, over a ten-week period, measured at the start (29 July) and at the end (7th October) of the treatment period.

Stomatal conductance

Stomatal conductance was measured to quantify the stomatal closure of a sample of plants in each treatment. The overhead-irrigated plants were measured on 11 September and the capillary-irrigated plants on 22 September.

Medium pH and EC

Samples collected between 11 - 12 September were analysed as described for Year 1 above.

Replication and statistics

There were six replicate plants of each subject per treatment. The significance of treatment effects was established by analysis of variance (ANOVA).

Year 3 (1998)

A. Root box study on effects of localised high CRF concentrations

Plant material

Plants of *Viburnum tinus* and *Elaeagnus pungens*, in 0.5 and 1 L containers respectively, were transplanted into glass-sided root boxes in July.

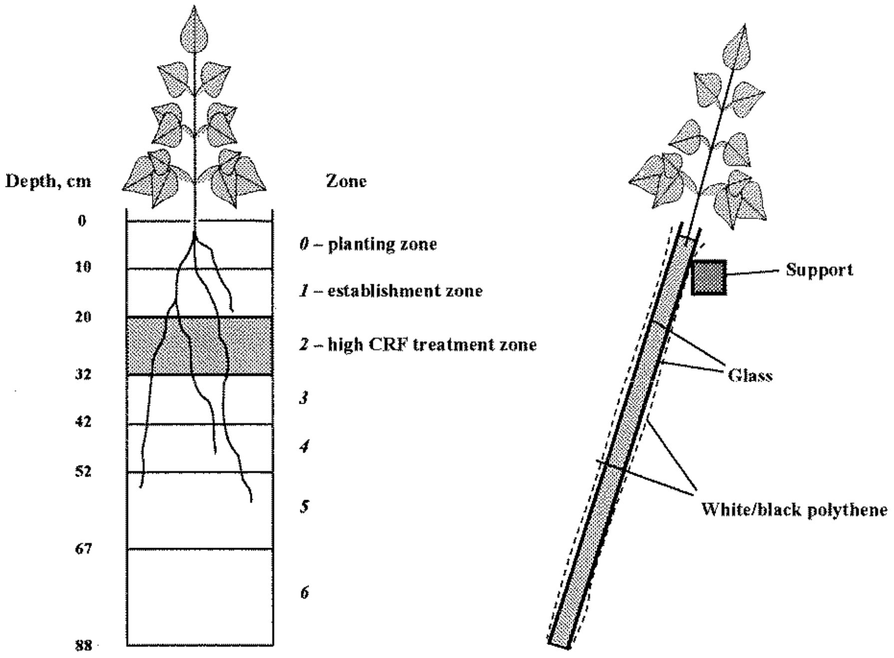
Root boxes

The root boxes were 27 cm wide x 88 cm tall but the depth between the two glass faces was only 2.5 cm so that a substantial proportion of the main roots would be visible through the glass. Some cutting back of the root-balls was required to fit them into these unusual 'containers'. The sides and bases of the boxes were wooden and the glass plates were held in place by additional wooden framework secured with bolts through the side pieces. There were holes for drainage at the base of each box.

Treatments

The medium was a 70:30 (v/v) mixture of peat : bark (Shamrock medium peat and Cambark 100). Through most of the profile this contained 3g L^{-1} Ficote 140 (16:10:10) but in a band from 12 to 20 cm below the surface the concentration of Ficote was either increased to 12, 24 or 36 g L^{-1} or held at the same concentration as the rest of the profile (Figure 3).

Figure 3. Diagram of the root boxes used to observe the effects of localised high concentrations of CRF on the appearance and development of roots. Apart from the treatment zone, in which CRF concentration ranged from 3 to 36 g L^{-1}*, the zones shown are arbitrary subdivisions of the profile used to describe root distribution. A layer of white/black polythene with its reflective white surface facing outwards served to exclude light and minimise radiant warming of the medium.*



Cultural conditions

The boxes were placed in a glasshouse with supplementary light maintaining a 16 hour photoperiod. The temperature was set to 20 °C. Sufficient water was added at the top of the boxes to keep the profile moist whilst minimising leaching of nutrients from the treatment zone.

Root growth and other assessments

The panels were inspected approximately twice per week to observe root development and condition. From 7 September, when the first roots approached the treatment zone, the extension of roots was quantified by marking the path of visible roots using a felt-tipped pen on a layer of polythene fixed to the glass. Comparison of the positions of root tips with marks made on the previous occasion, allowed root growth increments to be measured. Photographic records were unsatisfactory because of reflections from the glass.

The last regular root inspection was on 3 December, with an additional final inspection on 26 January. On 11 March, the boxes were opened and the medium was washed off the roots to determine the distribution of all roots, including those not in contact with the glass. To hold the roots in place as the medium was washed and teased away, rows of 7 cm nails, mounted in wooden battens at a spacing of about 7 cm, were inserted through the medium at the boundaries of the zones shown in Figure 3. The extracted roots were then transferred to a sheet of expanded polystyrene and photographed before the roots from each zone were excised, oven dried at 80 °C for 72 hours, and then weighed.

Physical measurements

On 8 February, four 30 cm^3 samples of the medium were extracted from each zone in one box of each treatment. The numbers of CRF granules in each sample were recorded before they were diluted 6-fold with purified water. The electrical conductivity (EC) and pH were determined one hour later.

B. Observational trials with additional plant subjects

This experiment extended the range of plants whose root behaviour in response to a variety of stresses had been described. It also included some that had already been examined because it was of interest to test the consistency of the observed symptoms which would determine the practical value of the diagnostic chart developed in Year 2.

Plant material

Most of the plants were grown from liners propagated in 1997; others were one year older. Through May and June, the liners were potted into one or two litre pots, depending on their size, and older plants were potted-on into three litre pots as detailed in Table 2 below:

Table 2. Details of the plants included in the observational trial in Year 3.

Plant subject	Container size, L	CRF in the salt stress treatment, g L^{-1}
Berberis stenophylla	1	30
Ceanothus 'Autumnal Blue'	1	30
Cornus alba 'Sibirica'	3	30
Cornus florida	3	30
Cotinus coggygria 'Royal Purple'	2	30
Elaeagnus pungens 'Maculata'	2	30
Forsythia x *intermedia* 'Lynwood'	1	30
Garrya elliptica 'James Roof'	2	15
Magnolia x *soulangiana*	1	30
Rhododendron 'Dopey'	1	15
Viburnum tinus 'Eve Price'	3	30

The treatments (with abbreviated title in brackets)

- Minimal stress ('Control') – potted into a 70:30 mixture of peat : bark with the standard amount of Ficote 180 (16:10:10) and fritted trace elements. Placed on a capillary sandbed and given additional watering overhead by hand when necessary.
- Short-term acute drought ('Drought') – treated the same as the Control plants until early August when the plants were transferred to a concrete path in a well-ventilated polythene tunnel where they received no irrigation until clear signs of stress had developed (up to three weeks). They were then thoroughly rewatered and returned to the capillary bed.
- Short-term acute waterlogging ('Waterlogged') - treated the same as the Control plants until early August when the plants were placed in a tank of water so that about half the depth of the container was submerged.

- Short-term high temperature stress ('Heat') – treated the same as the Control plants until 9 October when they were subjected to a single controlled heat treatment cycle similar to that shown in Figure 26 but with a maximum temperature of 47 °C in the final phase of the cycle. They were then returned to the capillary bed.
- Long-term exposure to high nutrient concentration ('Salt') – potted up into the same basic mix as Control plants but with 5 times the standard concentration of CRF (concentrations for each species shown in Table 2). The plants were then maintained on the capillary bed in the same way as the Control plants.

Root behaviour and other assessments

In late October, the plants were carefully knocked out of the containers and detailed observations were made of the appearance of the roots at the surface of the root-ball. At the same time a complete set of close-up photographs was taken of representative groups of roots of each combination of subject and treatment.

Systematic records of shoot response were not made but in general there was little or no visible effect on the shoots at the time that the plants were assessed. One exception, in relation to the Waterlogged treatment, is noted in the results section.

Results

Year 1

Survival

There were no losses of *Cotinus* in any of the treatments whereas *Garrya* was much more sensitive with many losses in the stress treatments (Table 3). *Garrya* was particularly sensitive to waterlogging which caused 60% of plants to die. Losses in the 'Low organic medium' were probably due to difficulty in maintaining an adequate water content in the medium on a capillary bed due to the very open texture of the vermiculite. The losses of *Garrya* plants may have been exacerbated by the general lack of vigour in this particular batch of *Garrya* plants, which was evident from the limited growth made by plants in the control treatment.

Table 3. Numbers of plants surviving the stress treatments (out of fifteen).

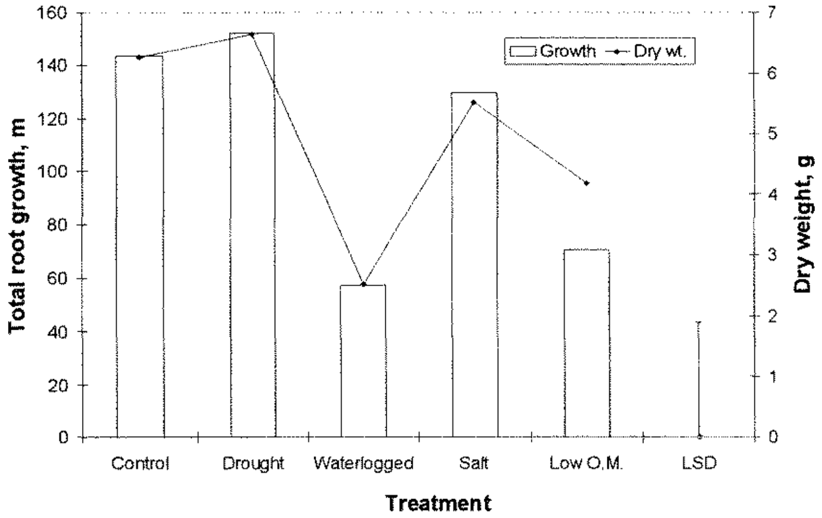
	Species	
Treatment	*Cotinus coggygria*	*Garrya elliptica*
Control	15	15
Drought	15	10
Waterlogged	15	6
Salt	15	12
Low O.M.	15	9

Appearance and growth of roots and shoots

Cotinus

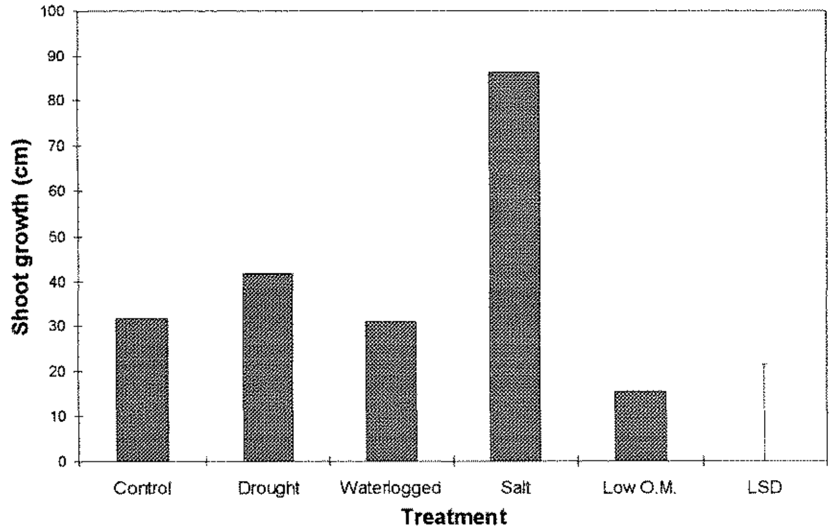
Waterlogging caused premature blackening of root tips compared to the pale pink or white root tips in the Control plants. This stress, and also the long term water shortage suffered by plants in the 'Low organic' medium, significantly reduced root growth (Figure 4).

Figure 4. Length and dry weight of roots in Cotinus coggygria *'Royal Purple' after exposure to environmental stress treatments in 1996.*



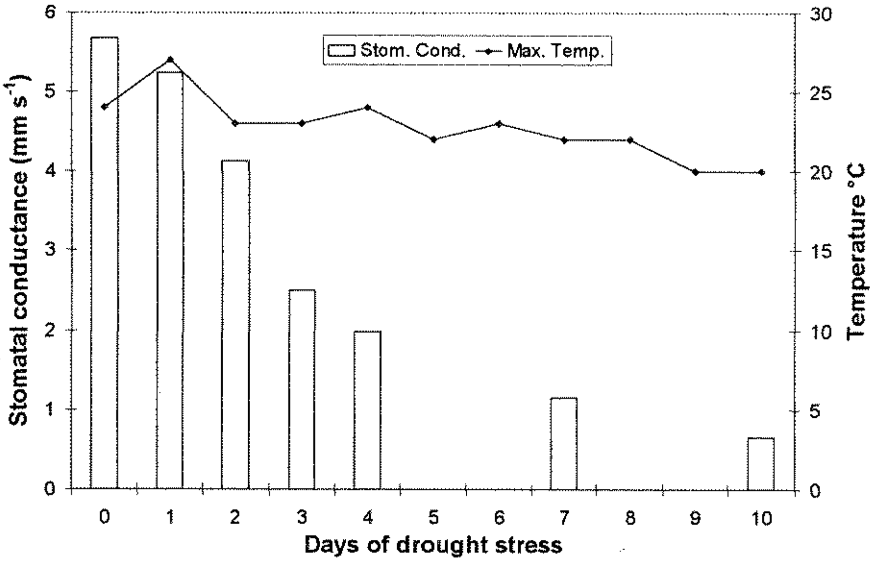
Shoot growth was not significantly reduced by any of the stresses; indeed the 'Salt' treatment increased growth significantly (Figure 5). This result indicates that 4 g L^{-1} of Ficote 180, the amount chosen as standard, was suboptimal for *Cotinus*, at least under the conditions of this experiment. This may result from the presence in the medium of 30% bark, which tends to lock up N. Furthermore, there was no evidence that three times this amount, in the 'Salt' treatment, of salt damage, such as leaf 'scorch'.

Figure 5. Increase in the total length of shoots per plant in Cotinus coggygria *'Royal Purple' during summer 1996.*



Exposure to a single episode of water deficit, over the course of about 10 days, in the Drought treatment had no significant long-term effect on the growth of roots or shoots, despite obvious wilting at the time. Measurements of stomatal conductance with a porometer showed that stomata closed rapidly to reduce water loss as the medium dried out (Figure 6).

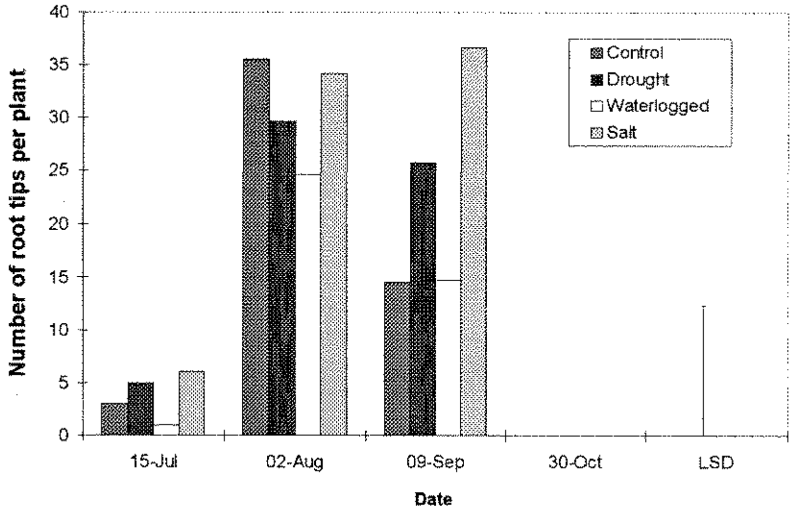
Figure 6. Stomatal conductance in Cotinus coggygria *'Royal Purple' over the course of the drought stress treatment together with daily maximum air temperature as an indicator of weather variations.*



Stomatal closure was also associated with the waterlogging treatment, suggesting that the shortage of oxygen around part of the root system either impeded the ability of roots to take up water or triggered a physiological signal, such as the release of plant hormones, which induced partial stomatal closure (Figure 6). However, 24 hours after returning the containers to the capillary bed where normal drainage could occur, stomatal apertures had already largely recovered.

The numbers of healthy root tips visible on the surface of the root-ball provided a simple and non-destructive means of monitoring the intensity of root growth activity over the course of the season. The results in Figure 7 showed that root activity decreased after August, except in the Salt treatment in which it reached a maximum in September, perhaps reflecting the shortage of nutrients in the other treatments that was mentioned earlier. At the end of October, when plants were destructively harvested, all evidence of new root extension had ceased.

Figure 7. Effects of stress treatment on the root activity of Cotinus coggygria *'Royal Purple', as measured by the numbers of growing root tips visible on the surface of the root-ball on three dates in 1996.*



Garrya

Shoot growth was slow even in the Control plants and, perhaps for this reason, there were no significant treatment effects (data not shown). In contrast, root growth was reduced in all the stress treatments, the effect being significant for the 'Salt' treatment (Figure 8). Roots in this treatment were characterised by a light grey, lustreless appearance, often with a dark brown colour in the centre (i.e. associated with the stele). The edge of the root (i.e. the cortex) often appeared almost translucent and was sometimes shrouded in fungal mycelium (this was probably a secondary infection by a saprophytic or weakly pathogenic fungus and was not the cause of the change in root appearance). In many plants there was limited root branching and few root hairs were observed. These symptoms, together with the relatively low dry matter content per metre of root, suggest that root membranes had been damaged by salt, causing cell lysis and a resultant loss of assimilates.

In contrast, waterlogged plants were characterised by localised areas of root growth. There was often some root activity in the top region of the medium, but little root development in the bottom third where shortage of oxygen would have been most severe. Most of the roots that had penetrated near the bottom of the pot prior to waterlogging became dark brown and necrotic during the waterlogging. Indeed, within a single root, tissues became progressively darker as they approached the base of the container. In contrast to salt-damaged roots, no fungal mycelium was seen on roots in the waterlogged plants, even after re-draining.

The amount of root recovered from the low organic medium was also less than from the Controls, but the visual assessments of roots *in situ* generally showed a well branched, proliferating root system over the surface of the medium. There were few very fine roots however and in areas of the medium it was apparent that roots were desiccated or had died, due to lack of water.

Figure 8. Length and dry weight of roots in Garrya elliptica *'James Roof' after exposure to environmental stress treatments in 1996.*

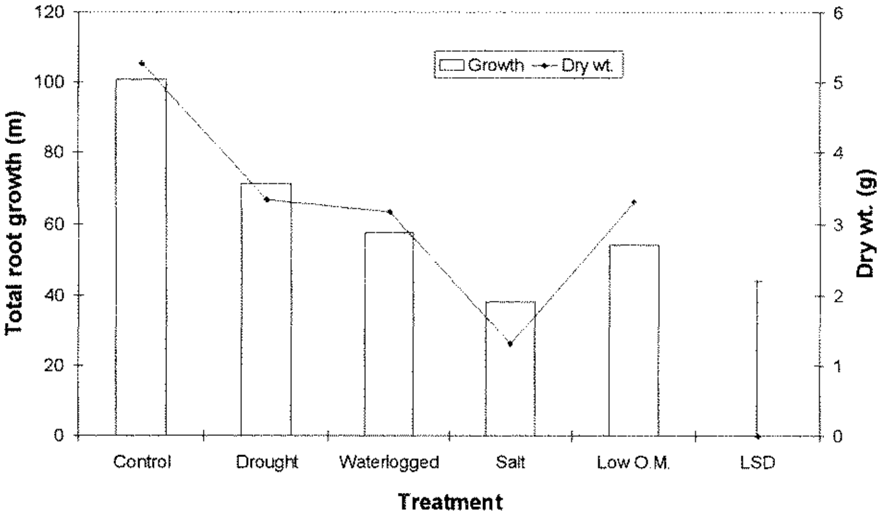
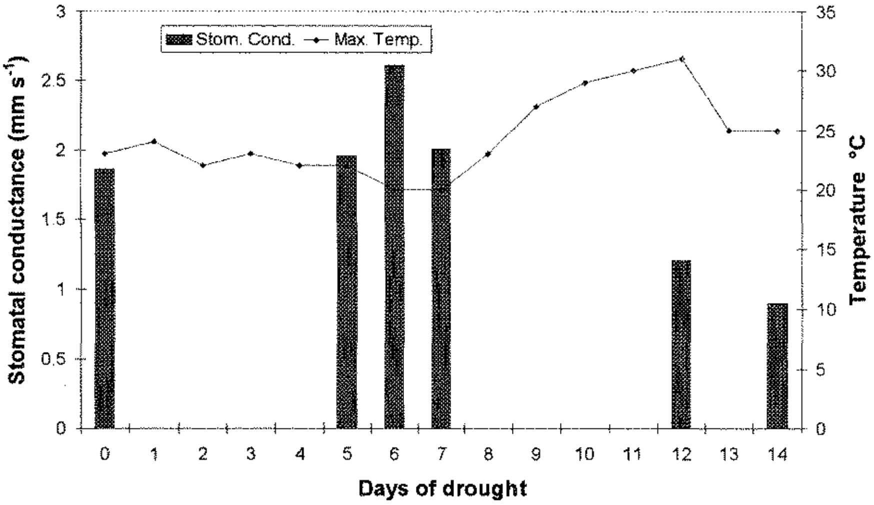


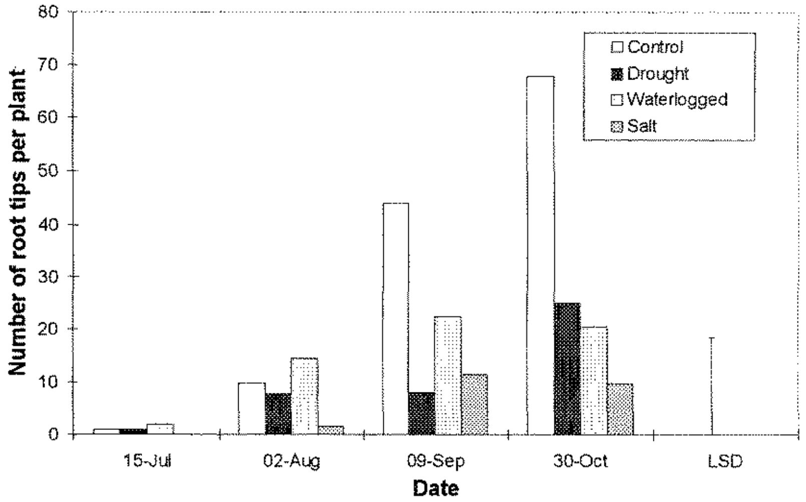
Figure 9. Stomatal conductance data for Garrya elliptica *'James Roof' showing a slow and inconsistent decrease over the course of the drought stress treatment. Daily maximum air temperature is included as an indicator of weather variations.*



Compared to *Cotinus*, stomata in plants of *Garrya elliptica* 'James Roof' were slow to respond to drought (Figure 9) and some plants failed to recover when eventually rewatered. Root activity was also significantly reduced by this stress (Figure 10) and the roots on the surviving plants had a distinctive appearance. They were mainly thick, unbranched and slightly contorted, perhaps because of the increase in resistance to root extension that occurs as a medium becomes drier.

In marked contrast to *Cotinus*, the numbers of active root tips on *Garrya* plants continued to increase late in the season (Figure 10). Even at the end of October, root activity in the Control plants was significantly greater than the other treatments. In the case of the Drought treatment, the brief stress exposure had ceased two months earlier. This result demonstrates, therefore, how long-lasting the influence of stress can be on subsequent growth and establishment.

Figure 10. Effects of stress treatment on root activity of Garrya elliptica *'James Roof', as measured by the numbers of growing root tips visible on the surface of the root-ball on three dates in 1996.*



Treatment effects on media pH, electrical conductivity and air-filled porosity.

The pH of the peat:bark medium started at 4.4 and tended to decrease slightly over the course of the experiment despite irrigation with water at pH 7.8. There was no evidence of any change in the pH of the vermiculite-based medium ('Low organic' treatment) which averaged about pH 5.9 (data not shown).

EC rose from low initial values to peak at about 850 μS cm^{-1} in August. In the high salt treatment it rose faster and reached a higher peak, about 1300 μS cm^{-1}. This represents an increase in peak EC of about 50%, which is much less than the 300% difference in fertiliser amount, suggesting that other factors, such as salt accumulation from the capillary irrigation water, may have contributed to the rise in EC.

The air-filled porosity of the standard medium (70:30, v/v peat : bark mix) was approximately 15 % when first measured 2 weeks after potting, whereas that of the Low organic medium (7:3:90, v/v/v peat, bark, vermiculite) was much higher (27 %). Air-filled porosity generally decreased with time in both media as they weathered and the roots proliferated within them. This was particularly marked with *Cotinus* and in the relatively open structure of the Low organic matter medium, where percentage air-filled porosity almost halved during the experiment (Table 4). The data for *Garrya* were less consistent, partly because slumping of the medium was more marked. This made it necessary to incorporate an allowance for this on the last measurement date because the surface of the medium had dropped to below the reference line on the container.

Table 4. Percentage air filled porosity determined by in situ *measurements.*

Date	Treatment					
	Control	Drought	Waterlogged	Salt	Low O.M.	LSD
***Cotinus* containers**						
25 June	17.6	15.5	15.5	14.3	29.9	
27 August	10.7	15.6	14.2	13.4	22.1	
9 October	12.5	12.0	9.9	10.3	17.7	3.3
***Garrya* containers**						
25 June	17.0	12.5	14.1	13.9	24.1	
27 August	12.7	18.1	11.1	15.0	22.9	
9 October	13.8	10.6	14.2	11.1	15.2	2.7

Year 2 responses to repeated exposure to mild stress

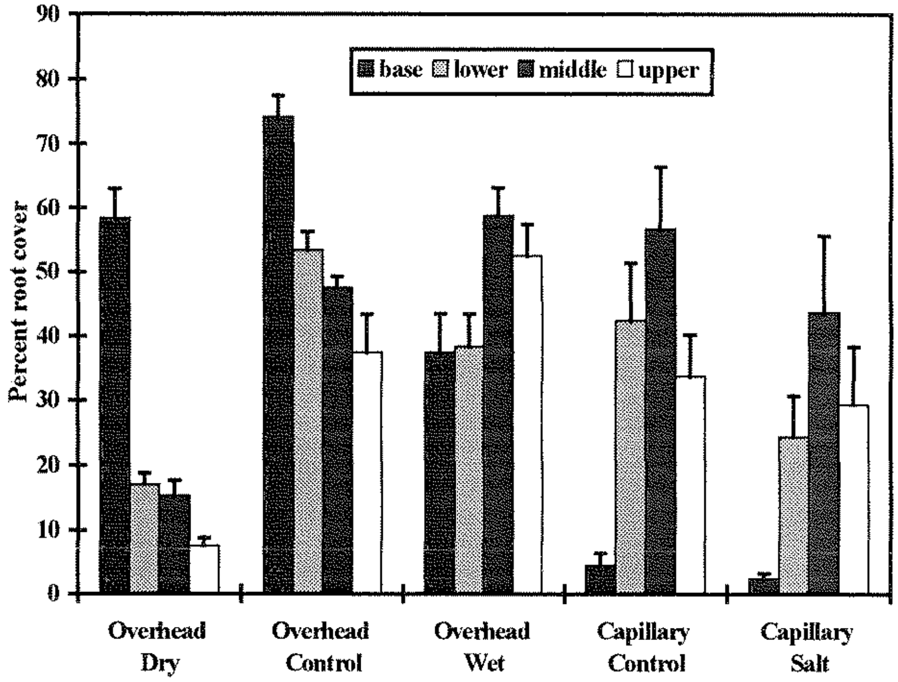
1. Cotinus coggygria 'Royal Purple'

Table 5 summarises the effects of all the treatments on root and shoot growth, as determined in the destructive harvest at the end of the experiment, together with the percentage of the root-ball surface covered with roots, as estimated in the non-destructive method. The additional data on the distribution of roots from the non-destructive method are shown in Figure 11.

Table 5. Root and shoot growth in Cotinus coggygria *'Royal Purple'. Dry weight per unit root length was calculated as an indicator of average root diameter ('root thickness index').*

	Treatment					
	Overhead			Capillary		
	Dry	Control	Wet	Control	Salt	LSD
root length (m)	110	284	197	165	110	115
root fresh weight (g)	17.0	47.9	32.8	26.6	19.6	15.3
root dry weight (g)	4.69	9.86	6.98	5.97	4.86	3.48
'root thickness' (g m^{-1})	0.043	0.035	0.037	0.037	0.045	0.010
total root cover (%)	21.2	51.1	53.4	41.8	30.5	13.9
shoot growth (cm)	4.5	28.6	26.3	27.5	27.0	14.1

Figure 11. Percentage root cover in different zones of pots containing Cotinus coggygria *'Royal Purple' grown under five treatments.*



Water Stress

Repeated drying out of the medium sufficient to cause slight wilting of plants in the 'overhead-dry' treatment significantly reduced the length, fresh weight and dry weight of roots, and shoot extension (Table 5). Root length was reduced by 60%, as was the percentage of the root-ball assessed visually to be covered with roots, and this was associated with a 50% decline in root dry weight. The reduction in visible roots was greatest in the upper layers and least across the base of the container (Figure 11). Presumably, as the plants began to suffer from water deficit, roots developed more in the lower layers where some water was still available. This tends to occur when containers are under-watered because the upper layer, which dries out rapidly due to additional water loss direct from the exposed surface, becomes hydrophobic. As a result, water passes through the upper layer rapidly without being absorbed into the smaller pores.

Visible roots on the sides of the root-ball were much thinner than in 'overhead-control' plants and many appeared severely desiccated where the medium was dry. However, this was not reflected in the root thickness index (i.e. dry weight per unit root length, Table 5), probably because the development of fine lateral roots was suppressed. The colour of the visible roots was a darker purple/brown than that of the deep red roots seen on the control plants.

Waterlogging

Plants grown under the 'overhead-wet' treatment had 30% less root than the control plants, in terms of length, fresh weight and dry weight, though none of these differences was quite large enough to be statistically significant. However this was associated with a significant change in the distribution of roots, with a reduction in cover over the base, from 74% to 37%, and the lower layer from 53% to 38%, together with a corresponding increase in root cover over the middle and upper zones

(Figure 11). As a result of this redistribution, the root system was able to largely avoid the zone that was subject to repeated waterlogging. This probably explains why there was no reduction in shoot growth. The few roots present in the lower layer were from deep purple to black in colour, compared to the bright red colour of healthy young roots in the upper layers and on the control plants. The roots in the upper layers were slightly thicker but less prolific than in control plants. No guttation was seen in this or any other treatment (i.e. drops of water exuding from the leaves, associated with generous water availability combined with low evaporative demand).

Salt stress

The high nutrient level in the 'capillary-salt' treatment reduced the length and weight of the roots, and the percentage of the root-ball covered with roots, but none of the differences was quite large enough to be significant (Table 5). There was no effect on root distribution (Figure 11), on the appearance of the roots, or on shoot growth (Table 5). No significant effect of the high salt treatment on EC or pH was detected. The reason for this surprising result may have been that the samples were taken towards the end of the experiment when much of nutrient would have been released from the CRF granules, or to an overriding effect of the accumulation of solutes taken up from the capillary bed.

Overhead versus capillary irrigation

The length and weight of roots harvested from control plants on the capillary beds were about 40% of those for the control plants under overhead irrigation (Table 5).

There was a smaller and non-significant difference in average root cover in the same direction. This was due mainly to an almost complete lack of roots over the base of the containers standing on the capillary bed. The appearance of roots in the lower zone was similar to those in the waterlogged plants, suggesting that the water table in the capillary bed was being maintained too high for these plants, leading to slightly anaerobic conditions at the base of the containers. However, this was not associated with any effect on shoot extension (Table 5).

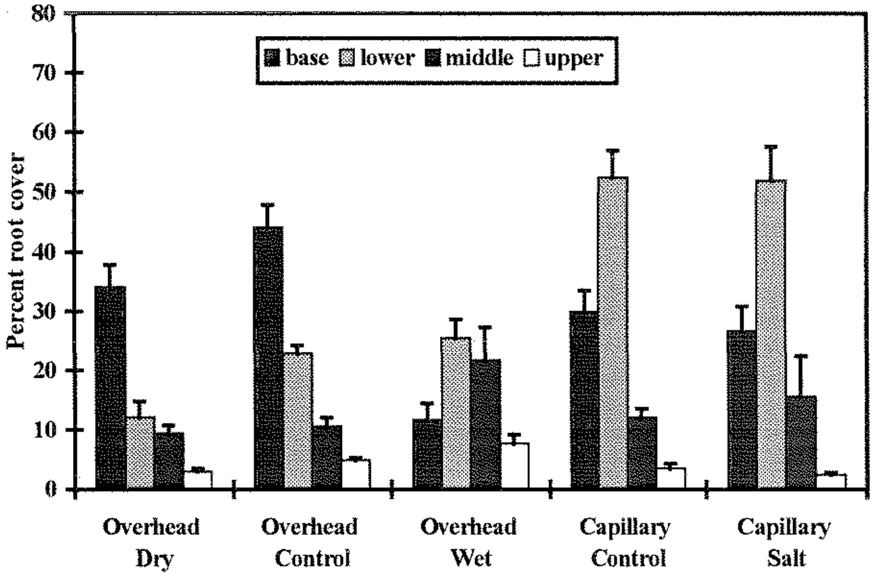
2. *Elaeagnus pungens* 'Maculata'

Table 6 summarises the effects of all the treatments on root and shoot growth, as determined in the destructive harvest at the end of the experiment, together with the percentage of the root-ball surface covered with roots, as estimated in the non-destructive method. The additional data on the distribution of roots from the non-destructive method are shown in
Figure 12.

Table 6. Root and shoot growth in Elaeagnus pungens *'Maculata'. Dry weight per unit root length was calculated as an indicator of average root diameter ('root thickness index').*

	Treatment					
	Overhead			Capillary		
	Dry	Control	Wet	Control	Salt	LSD
root length (m)	142	125	132	133	100	33
root fresh weight (g)	42.3	40.5	43.0	47.0	37.5	9.4
root dry weight (g)	8.05	6.9	6.8	7.3	6.0	1.7
'root thickness' ($g\ m^{-1}$)	0.06	0.06	0.05	0.06	0.06	0.01
total root cover (%)	12.7	18.3	17.1	24.0	23.9	4.4
shoot growth (cm)	7.0	15.3	15.5	10.7	12.5	4.8

Figure 12. Percentage root cover in different zones of the pot for Elaeagnus pungens *'Maculata' grown under five different treatments*



Water stress

Neither the length nor weight of roots was reduced by drought stress in Elaeagnus plants (Table *6*). Despite this, the percentage of the root-ball covered with roots was significantly less in the 'overhead-dry' treatment, 12.7% compared with 18.3% in control plants. This reduction is evident in the comparison of Plates 1 and 2 (in Appendix 2). Some of this reduction occurred over the basal layer of medium as well as near the surface (Figure *12*), but the proportion of roots visible over the base of the root-ball was increased by the drought treatment, as was observed in Cotinus. This indicated that a high proportion of visible roots on the base of the root-ball is a good indicator of chronic water shortage.

Another feature of the water-stressed plants was that roots tended to be distributed unevenly between opposite sides of the container (Plate 1). It appeared that this was usually associated with fewer roots on the south facing side of the container and to be caused by greater desiccation on this side. Those roots present were generally rather shrivelled and brown compared with roots on the side less exposed to the sun, which were actually whiter and more fleshy than those on the control plants. Furthermore, root branching was less frequent in the droughted plants. Water stress in the 'overhead-dry' treatment was associated with a significant suppression of shoot elongation (Table *6*).

Waterlogging

Waterlogging in the 'overhead-wet' treatment altered root distribution but had no effect on any other measured parameter of root behaviour. Plants showed a significant decrease in base cover with a concomitant increase in cover in the middle zone (Figure 12). Plate 3 shows this decrease in root cover over the base compared with 'overhead-control' (Plate 2). Unlike *Cotinus* roots, those roots of *Elaeagnus* plants that were present on the base of the container did not appear necrotic though a few grey and flaccid-looking roots were seen. Nonetheless, these results suggest that a low proportion of visible roots over the base of the container may be a useful indicator of intermittent waterlogging.

Salt stress

As in *Cotinus*, the 'capillary-salt' treatment reduced significantly the fresh weight and length of roots in Elaeagnus but not their dry weight or the percentage of the root-ball covered (Table *6*). In some plants, a greater proportion of vertically orientated roots was observed than in the 'capillary-control' treatment (Plates 4 and 5) and roots tended to be slightly thicker (Table *6* and Plate 5), with more of the brown colour which is associated with suberization. There was no effect on root distribution *(Figure 12*), nor was there any effect on leaf appearance or shoot growth (Table *6*).

In contrast to the puzzling results from the *Cotinus* containers, the EC of the medium was increased by 510 μS cm^{-1} in the 'capillary-salt' treatment.

Overhead versus capillary irrigation

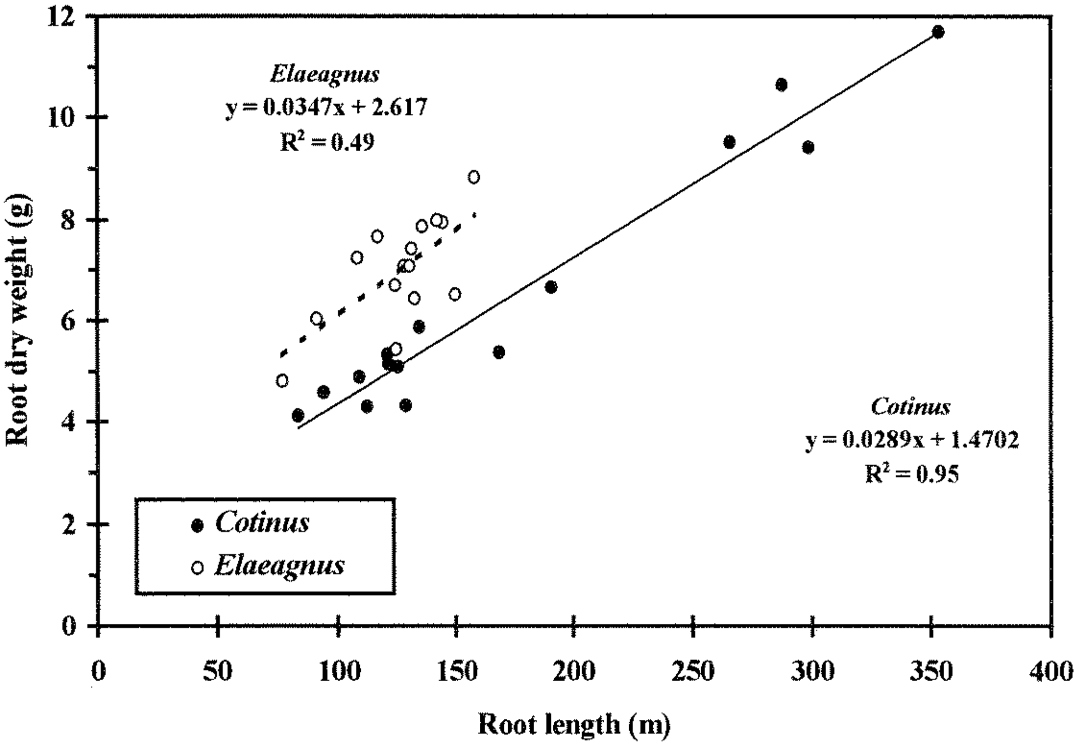
The type of irrigation had much less effect on *Elaeagnus* than it did on *Cotinus*. This is consistent with the suggestion that the adverse effect of capillary irrigation on *Cotinus* was due to marginally over-wet conditions near the base of the pot as *Elaeagnus* was relatively tolerant of the waterlogging treatment. Nonetheless, there was a reduction in the percentage root cover over the base, from 44% under overhead irrigation to 30% on the capillary bed, with a compensating increase in root cover at the sides of the lower zone (Figure 12). These differences are illustrated by comparison of Plates 3 and 4.

3. Validity of alternative methods of estimating the quantity of roots present

Figure 13 shows the relationship between the length and dry weight of the roots harvested from individual plants. A clear relationship is evident for both species, though there is substantial scatter of points around the regression line. The R^2 statistic, which provides an objective measure of the closeness of the relationship, was 0.95 for *Cotinus* (i.e. 95 % of the variation in root dry weight can be predicted from variation in root length) but only 0.49 for *Elaeagnus*. However, the graph suggests that the lower value for *Elaeagnus* is probably due mainly to the small range of root quantities observed (less than half the range observed in *Cotinus*) rather than an inherent difference in the precision of the relationship. Therefore, rarely would much be gained from the lengthy procedure of scanning the entire root system to estimate its root length. Instead, either root dry weight alone, or root dry weight combined with an estimate of dry weight per unit length from a small subsample, would usually provide almost as thorough a measure of the quantity of roots present.

The validity of visual assessments, often considered too subjective, was assessed by comparison with the root dry weight data for *Cotinus*. The R^2 value was 0.50, indicating that percentage cover of the root-ball, when done in a systematic way over 13 separate parts of the root-ball, can give a useful estimate of the relative amounts of root in different treatments or batches of plants. Considering how much less time-consuming it is, and that it is non-destructive, this result shows that the visual assessment of root quantity can be a valuable technique, at least when it is done systematically. Furthermore, the results of this experiment demonstrate clearly the additional value of the information on the distribution of roots that is provided by the zonal assessment system used here.

Figure 13. The relationship between root length and dry weight in Cotinus coggygria *'Royal Purple' and* Elaeagnus pungens *'Maculata'.*



4. Results for a wider range of subjects

Similar experiments on a wider range of subjects were based entirely on non-destructive measurements, particularly the quantification of root activity using the zonal visual assessment method. This was complemented by careful examination of the exposed roots to identify visual characteristics that might serve as diagnostic tools to help growers identify the type of stress that could be reducing the performance of their plants. Photographs were taken to try to document this aspect of the assessment but, as always, it was hard to capture truly typical symptoms in a photograph of a single plant. The photographs (in Appendix 2) concentrate on *Elaeagnus* and *Berberis* because these subjects provided the most visible contrasts between treatments.

Water stress

Repeated exposure to mild water stress over a ten-week period, in the 'overhead-dry' treatment, substantially decreased the root growth of all subjects compared to controls. Most extreme were the responses of *Cotinus* and *Viburnum* (60% reduction in root cover); *Elaeagnus* and *Garrya* (30% reduction in root cover) were least affected. A reduction in root cover under 'overhead-dry' was not surprising. If the water supply is consistently less than the potential evapotranspiration, plants will eventually suffer some form of desiccation stress. Growth will be slowed down either as a direct result of the water deficit or physiological processes such as cell expansion, or as the indirect effect of stomatal closure (discussed further below).

Table 7. The effect of drought (Dry) and waterlogging (Wet) treatments on percentage total root cover.

	Overhead Treatment			
Plant subject	Dry (%)	Control (%)	Wet (%)	LSD
Cotinus coggygria	21.2	51.1	53.4	13.9
Elaeagnus pungens	12.7	18.3	17.1	4.4
Garrya elliptica	17.0	24.5	22.7	9.7
Berberis stenophylla	18.3	34.9	14.9	5.3
Acer palmatum	2.8	5.0	4.1	3.2
Viburnum tinus	7.4	18.4	18.4	6.9
C. alba 'Sibirica'	20.4	34.2	25.2	8.1
C. alba 'Elegantissima'	19.7	42.1	37.5	9.4

A decrease in the growth of the shoot under drought conditions may not, initially, be reflected by a decrease in root growth. Under drought conditions, many plants reallocate resources away from shoot growth and invest in root growth in an adaptive response which, under natural conditions, will often increase water uptake. However, if the plant water deficit persists, as was the case in this experiment, then eventually root growth is limited by the smaller quantity of assimilates being produced by the above-ground part of the plant.

The data for the percentage root cover over the base of the container (Figure 14) were particularly interesting. Although on average total root cover was reduced by 45% in the 'overhead-dry' treatment (Table 7), base cover actually increased by 5%. This increase was significant in *Berberis* and *Garrya*.

Plate 6 shows the extent of base cover in a droughted *Berberis* plant, and can be compared with a plant in 'overhead-control' (Plate 7). The photograph also indicates the influence of aspect on root distribution. As described earlier for *Elaeagnus*, roots on the south facing side of the container were very infrequent and severely desiccated, being much finer and more brown coloured. A few roots were shrivelled, brittle and presumably dead. Most other subjects responded in a similar manner but the size of the response was variable. With the simple protocol used to simulate repeated under-watering on a container bed that was used in this study, some of this variation was probably due to differences in the rate at which different species used water, and thus how quickly water stress developed, rather than to inherent differences in plant responses to water stress.

Figure 14. Percent base root cover in pots containing all eight subjects grown under three water treatments with overhead irrigation. Error bars represent least significant differences within subjects.

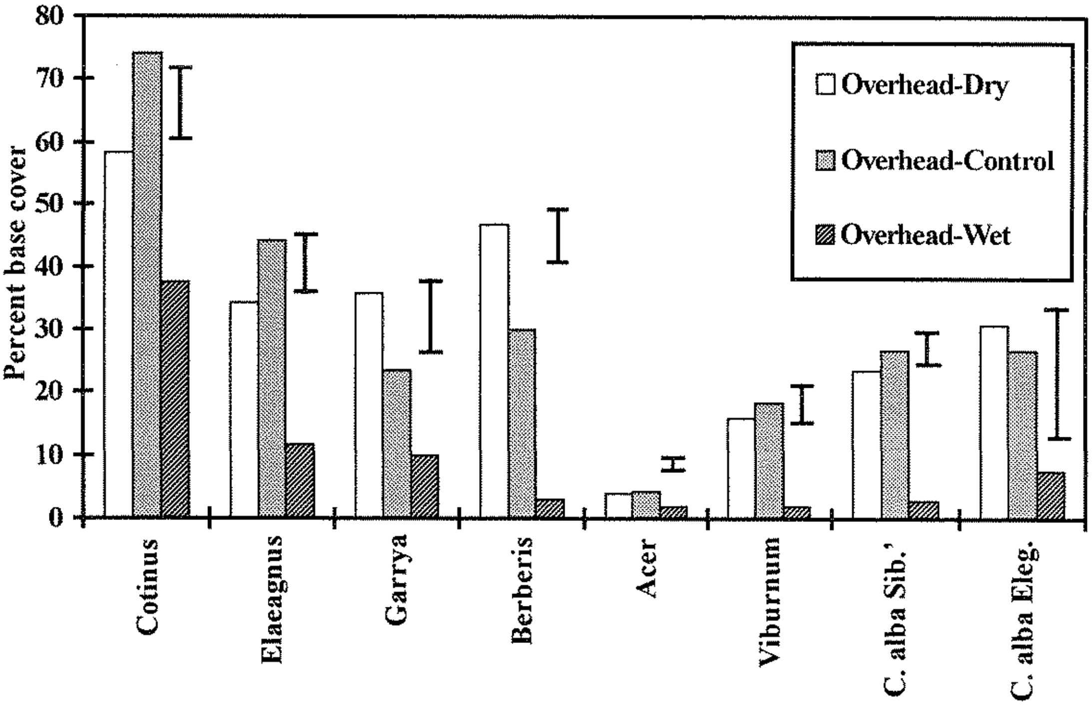


Table 8. Mean shoot growth increment over ten weeks in all subjects.

	Overhead Treatment			
	Dry (cm)	Control (cm)	Wet (cm)	LSD
Cotinus coggygria	4.5	28.6	26.3	14.1
Elaeagnus pungens	7.0	15.3	15.5	4.8
Garrya elliptica	9.4	27.4	24.0	6.8
Berberis stenophylla	8.9	46.5	19.6	19.3
Acer palmatum	20.1	27.1	18.8	13.5
Viburnum tinus	0.7	5.9	2.9	3.3
C. alba 'Sibirica'	0.6	0.0	0.0	1.9
C. alba 'Elegantissima'	1.9	19.0	23.0	6.4

Shoot extension was reduced significantly by water stress in all subjects except *Acer* and *C. alba* 'Sibirica' (Table 8). *Cornus alba* 'Sibirica' showed little or no shoot extension under any treatment because its shoots had terminated by the time the treatments were imposed. The lack of a significant decrease in shoot extension in *Acer* under the 'overhead-dry' treatment probably reflects the small leaf area of the plants relative to the size of their containers.

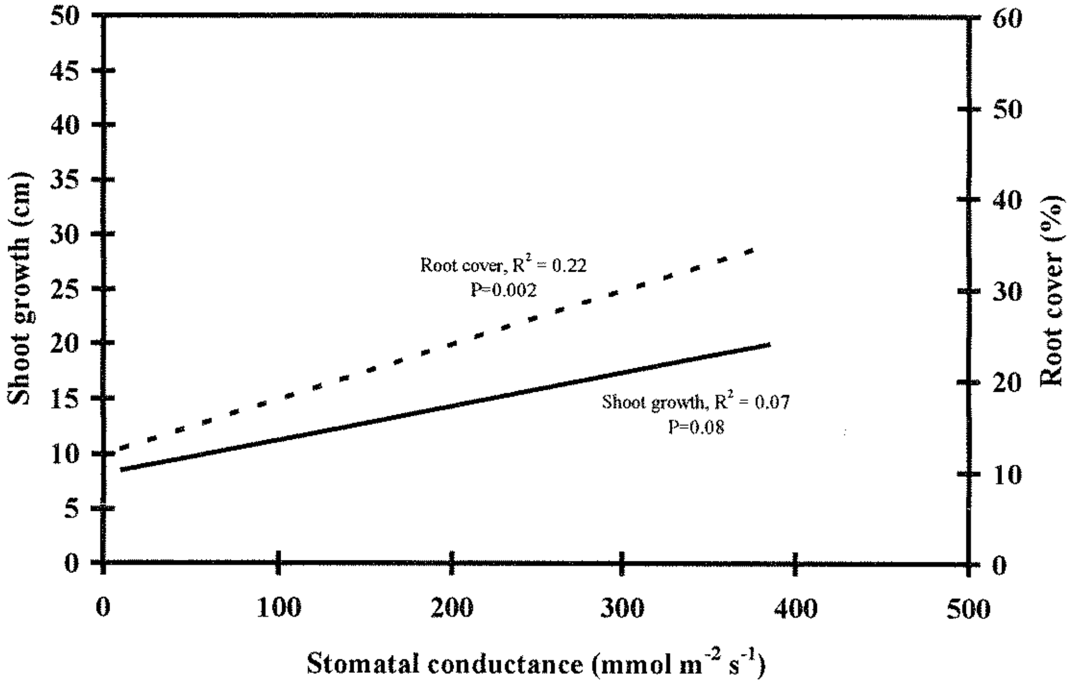
Stomatal conductance (g_s), which is related to the degree of opening of the stomatal pores, was measured as a useful indicator of the water status of plants. Stomata are pores in the leaves, which are necessary to allow carbon dioxide into the leaves where it can be assimilated to provide carbohydrates for growth and metabolism. While they are open, it is inevitable that water vapour will escape into the atmosphere, making the plant subject to potential water stress. If the plant becomes short of water, stomata will eventually close, to a greater or lesser extent, to reduce the rate of water loss and thus help to prevent damaging water stress. However, any strategy that the plant adopts to avoid water loss through stomatal closure will tend to cause a decrease in carbon dioxide assimilation and, hence, growth.

Table 9 shows the mean g_s for all subjects except *Berberis*, which was impossible to fit into the leaf chamber of a standard porometer. All subjects, with the exception of *Acer*, showed significant decreases in g_s in response to the 'overhead-dry' treatment. The greatest decrease was 90%, in *Viburnum* (Table 9) which also showed the greatest decline in total root cover (60%) and shoot growth (88%). Plants with the least root cover tended to have the lowest g_s ($P > 0.01$, Figure 15), whereas the correlation with shoot growth was not statistically significant.

Table 9. Stomatal conductance (g_s).

	Overhead Treatment			
	Dry Mmol m^{-2} s^{-1}	Control mmol m^{-2} s^{-1}	Wet mmol m^{-2} s^{-1}	LSD
Cotinus coggygria	44	130	173	71
Elaeagnus pungens	47	307	384	144
Garrya elliptica	76	306	301	89
Acer palmatum	71	105	151	55
Viburnum tinus	13	143	145	30
C. alba 'Sibirica'	128	276	181	121
C. alba 'Elegantissima'	87	320	324	123

Figure 15. Relationship between stomatal conductance, shoot growth (solid line) and root cover (broken line), based on data from plants in the Capillary-Dry and Capillary-Control treatments.



Waterlogging

There was a tendency for the percentage root cover over the root-ball as a whole to be reduced by the repeated waterlogging at the base of the containers in the 'overhead-wet' treatment (Table 7). However, the effect was statistically significant only in *Berberis* and *Cornus alba* 'Sibirica'. This was associated with a much more consistent reduction in root cover over the base of the root-ball (Figure 14), as illustrated for *Berberis* in Plate 8. Although the roots of *Berberis* were typically bright yellow in the control plants (Plate 7), any roots near the base in the 'overhead-wet' treatment were pale grey in colour and flaccid in appearance. Similar effects were observed in most subjects, especially with the two *Cornus* species and with *Garrya*. In the more vigorous subjects such as *Cornus*, roots avoiding the anaerobic environment were forced higher up the container and eventually through the top of the medium onto the surface (Plate 9). The redistribution of roots was particularly impressive in *C. alba* 'Elegantissima', in which the percentage of the upper zone covered with roots increased more than 5-fold, from 11 to 60% in the 'overhead-wet' treatment (data not shown).

These observations are almost certainly attributable to the development of anaerobic conditions in the waterlogged medium near the base of the containers. Long-term oxygen deprivation may lead to root dysfunction, reducing the capacity for uptake, reducing hormone production and sometimes causing roots to die, all of which can influence shoot growth. There was some evidence for reduced shoot growth when all subjects are considered together (Table 8). The most susceptible subject was *Berberis stenophylla*, which showed a significant decrease in shoot extension from 46.5 cm ['overhead-control'] to 19.6 cm ['overhead-wet']. Severe waterlogging can cause stomatal closure, as observed in *Cotinus* in Year 1, but there was no significant effect on *Cotinus*, or any other species, of the milder treatment used in this experiment (Table 9).

Salt stress

The 'capillary-salt' treatment did not significantly reduce root cover for any of the subjects (Table 7). This would suggest that, at these concentrations, salt toxicity was not a problem. Indeed the significant increase in cover in both *Cornus alba* varieties suggests the standard amount of CRF 6 g L^{-1}) was below the optimum for this species. However, some of the roots exposed to the higher CRF concentration (12 g L^{-1}) developed a purple/red colour suggestive of some degree of stress at this concentration (Plate 10), while over the base they blackened. None of the other subjects showed any visible difference in the appearance of their roots due to doubling the standard concentration of CRF (e.g. see Plates 11 and12 for *Berberis*).

Table 10. The effects of CRF concentration on percentage total root cover.

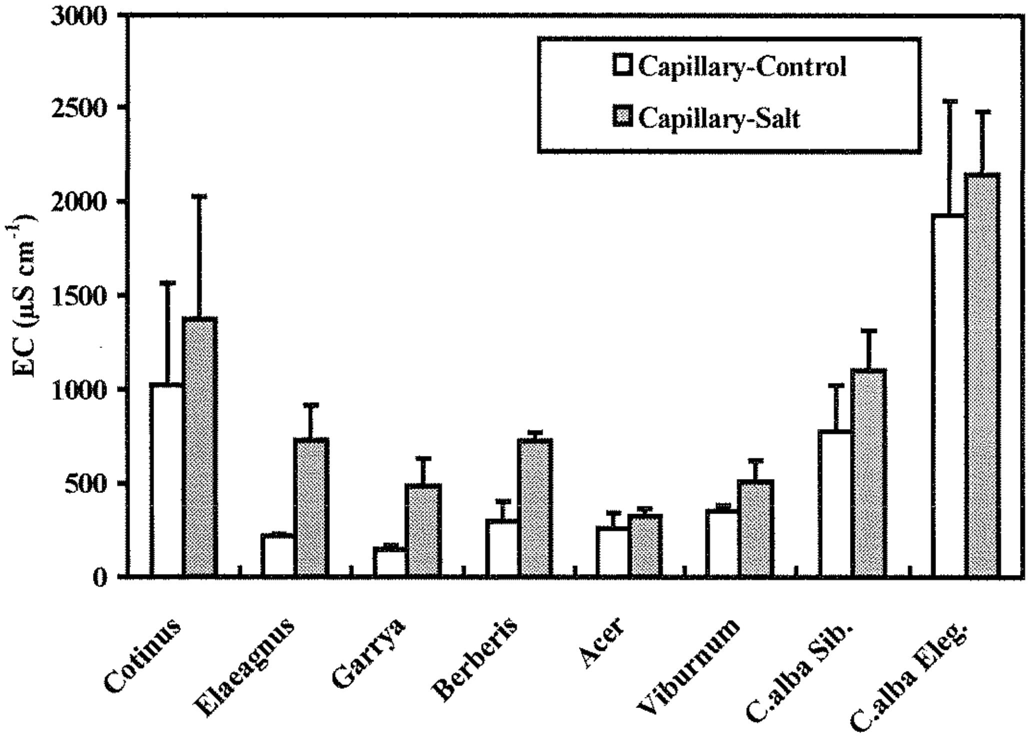
	Capillary-control (standard CRF amount)	Capillary-salt (double CRF amount)	LSD
Cotinus coggygria	41.8	30.5	13.9
Elaeagnus pungens	24.0	23.9	4.4
Berberis stenophylla	23.7	22.3	5.3
Acer palmatum	7.5	7.0	3.2
Viburnum tinus	18.3	18.3	6.9
C. alba 'Sibirica'	30.9	43.2	8.1
C. alba 'Elegantissima'	37.1	50.3	9.4

The root growth of *Garrya* was very limited on the capillary bed irrespective of the level of salts in the medium so that it was impossible to extract the root-ball intact for visual assessment. The reason for this was unclear but the poor growth is thought to have been partly attributable to the bed being run too wet (i.e. water table too high) for this species, whose roots seems to be particularly sensitive to waterlogging. Comparison of Plates 13 and 14 shows the similarity of roots from the capillary bed and the waterlogging treatment ('overhead wet').

Doubling the concentration of salts had no adverse effects on shoot growth or leaf appearance in any subject (data not shown).

Data for the pH and EC of the medium showed a consistent slight decrease in pH associated with the increase in EC at the higher CRF amount (Figure 16). The actual value of EC varied substantially between species, with the highest values (around 2000 µS cm^{-1}) in *Cornus alba* 'Elegantissima'. There apparent correlation between EC and the size of the plants could be attributable to reduced leaching during rainfall, but is more likely to indicate that a significant quantity of salts was being absorbed with the irrigation water from the sand bed. Partly these would be salts present in the water supply but there would also be nutrients that have diffused or been leached out of other containers.

Figure 16. Mean values of electrical conductivity (EC) for all eight subjects grown at two salt concentrations (6 g L^{-1} 'capillary-control' and 12 g L^{-1} 'capillary-salt' for all subjects except for Acer *and* Garrya *which were at 3 g L^{-1} and 6 g L^{-1}).*



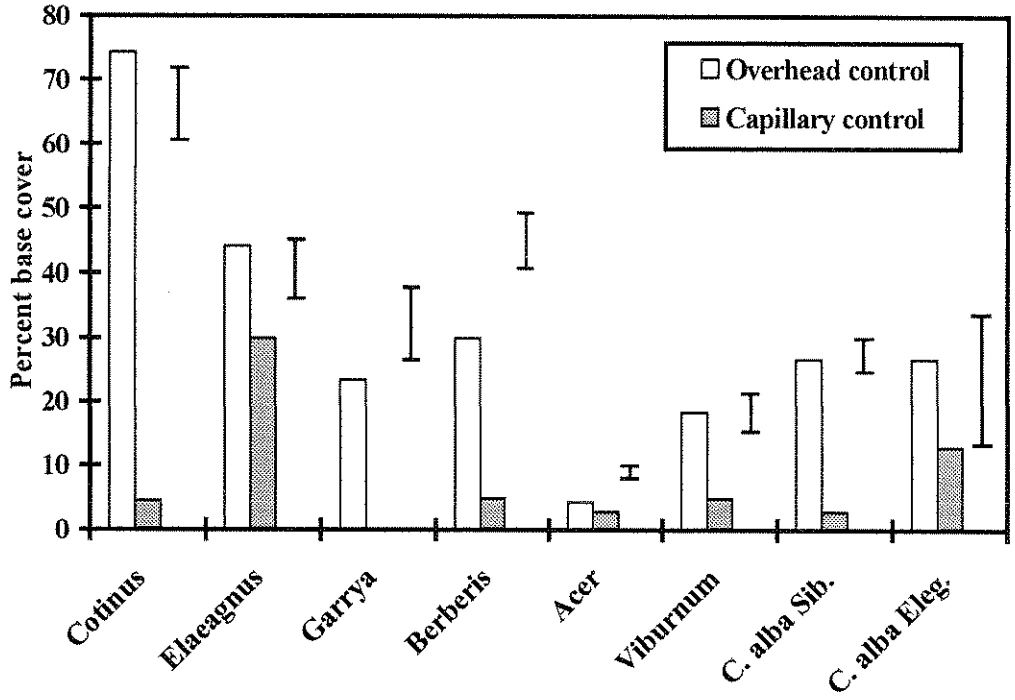
Overhead versus capillary irrigation

Apart from *Garrya, Berberis* was the only subject in which the control plants had significantly less visible root under capillary irrigation than under overhead irrigation (Table 8). However, almost all subjects had much less root visible across the base of the container when grown on the capillary bed (Figure 17).

Table 11. The effect of method of irrigation on the percentage of the entire root-ball covered with roots (control treatment in each case).

	Irrigation system		
	Overhead	Capillary	LSD
Cotinus coggygria	51.1	41.8	13.9
Elaeagnus pungens	18.3	24.0	4.4
Garrya elliptica	24.5	n/a	9.7
Berberis stenophylla	34.9	23.7	5.3
Acer palmatum	5.0	7.5	3.2
Viburnum tinus	18.4	18.3	6.9
C. alba 'Sibirica'	34.2	30.9	8.1
C. alba 'Elegantissima'	42.1	37.1	9.4

Figure 17. Comparison of root cover over the base of the root-ball in plants grown with overhead and capillary irrigation. Error bars represent least significant differences within each subject.



Year 3 (1998)

The results of the experiments conducted in the first two years identified the types of effects that are liable to occur where container plants repeatedly suffer either insufficient irrigation or over-irrigation. The picture with respect to excessive nutrient availability or 'salt stress' was much less clear. Doubling the recommended amounts of CRF had no effect on the appearance of shoots or roots of any of the subjects tested whereas the more extreme treatment used in Year 1 (3 times recommended amount) induced darkening of the roots of *Garrya* in a manner very similar to that caused by waterlogging. That result was reinforced by an additional experiment done overwinter under glass in which similar symptoms were observed in plants fed with a high concentration of a complete nutrient solution. Such extreme concentrations are unlikely to occur on a well-managed nursery so those symptoms are unlikely to be encountered. On the other hand, some non-uniformity of mixing of CRF granules can occur easily, and the granules may sometimes clump together, leading to localised high concentrations. The main experiment in the final year used transparent-sided root boxes to see how individual roots behaved as they extended into a band of medium in which the local CRF concentration was up to six times that recommended.

A. Root box study on effects of localised high CRF concentrations

Technically the root box approach worked well with many roots growing alongside the glass, most of which remained visible for many weeks. However, there was absolutely no change in the appearance of the roots as they encountered the high salt band, even in the treatment with 36 g L^{-1} of the fertiliser, and where they passed close to groups of CRF granules (Plate 20).

Great care was taken in extracting the root systems at the end of the experiment to look for evidence of more subtle effects of the high nutrient band. Plates 21 and 22 show the extracted root systems of the two subjects which indicate that the length of root in the lower layers decreased at the higher CRF concentrations in both species. With *Viburnum tinus* 'Eve Price' this was accompanied by a slight increase in the amount of fine branch roots produced in the layers of medium above the CRF treatment layer. Figure 18 and Figure 19 show the distribution of root dry weight; they confirm the impression gained from the photographs and show additionally that stimulation of rooting with high amounts of CRF occurred also in *Elaeagnus pungens* 'Maculata'.

Figure 18. The effect of the concentration of CRF in a layer, 12-20 cm below surface of a root box (marked FERT), on the distribution of roots of Viburnum tinus 'Eve Price'.

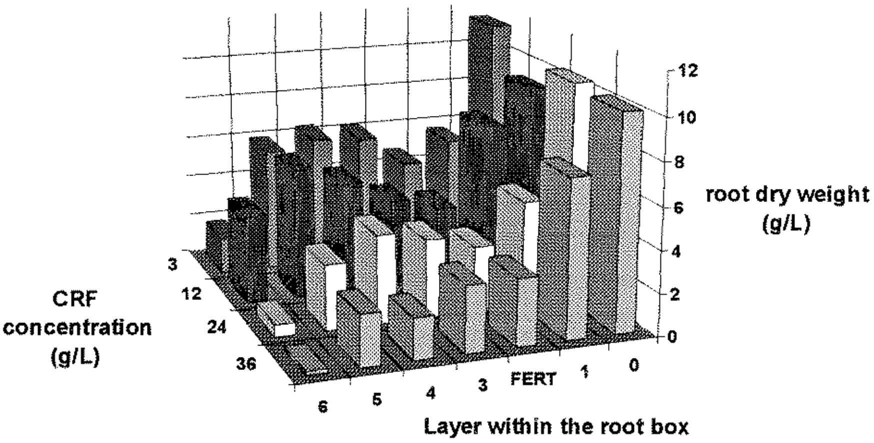


Figure 19. The effect of the concentration of CRF in a layer, 12-20 cm below surface of a root box (marked FERT*), on the distribution of roots of* Elaeagnus pungens *'Maculata'.*

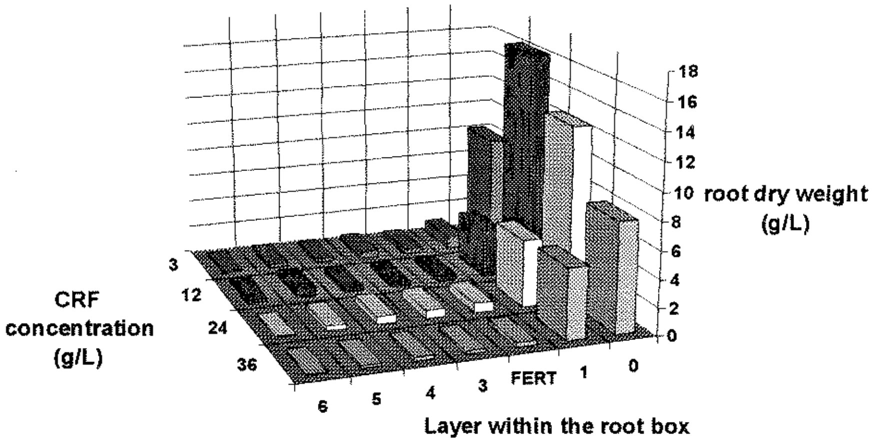
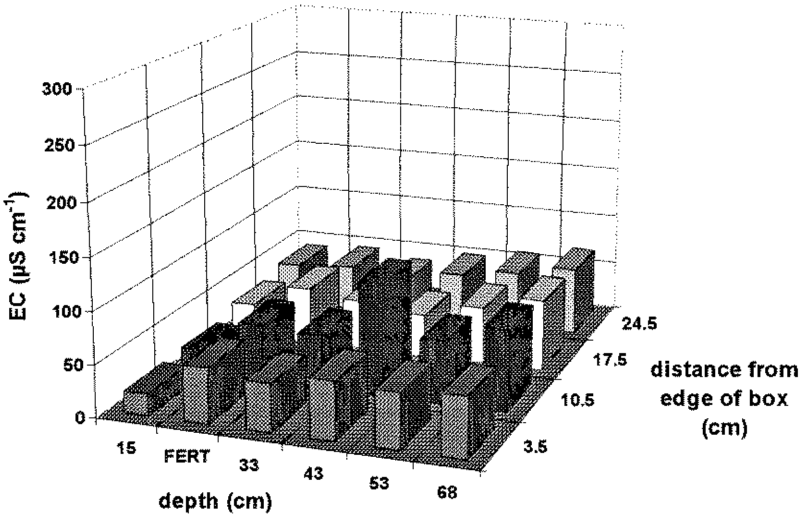


Figure 20. Vertical and horizontal variation in electrical conductivity (EC) of the medium within a root box 36 g L^{-1} of CRF in the treated layer (marked FERT*) measured at the end of the experiment.*



Measurements of EC made at the end of the experiment showed that salts were spread more or less uniformly throughout the profile (Figure 20). As this seemed surprising, a sample of CRF granules was removed from the medium, their contents were extracted into pure water, and the EC was determined. This showed that there were still nutrients in the granules though comparison with a sample of unused granules indicated that they were only about 20% of those originally present. Therefore, the uniformity of EC shown in Figure 20 implies that, once released from the CRF granule, the nutrient ions must have diffused away from the treated zone so rapidly that the high concentration of granules was not reflected in a high salt concentration in the medium around them. The practical conclusion is that 'salt stress' around local accumulations of CRF granules is unlikely to be a problem in practice, even without leaching by excess rainfall or irrigation.

B. Observational trial with additional plant subjects

Effects on the roots

Water stress

In all subjects, roots in the Drought treatment were distinctly thin and brown compared with the more fleshy and pale coloured roots of the controls. In the case of the naturally dark red coloured roots of *Cotinus coggygria* 'Royal Purple', the difference in colour was more difficult to see but nevertheless present. As the Drought treatment was a short term but acute treatment, the redistribution of roots noted in the Year 2 experiments was not observed.

Waterlogging

Most roots in the lower half of the container were grey or blackened, many with a flattened shape. In *Garrya, Ceanothus* and *Elaeagnus*, the stele of many roots was darkened more than the surrounding cortex. Combined with the flattening against the wall of container, this gave the roots a distinctive striated appearance for which we coined the term 'tram lines' symptom (see Plate 25).

Salt stress
Roots on all subjects looked less 'healthy' than the controls but it was difficult to identify precise symptoms that characterised their appearance. In general they were a darker colour, often grey to black (*Ceanothus*, *Elaeagnus*, *Garrya*, and both *Cornus* spp.) but sometimes brown (*Viburnum*, *Forsythia*, and *Magnolia*). Many also showed the same flattened appearance that was seen in response to waterlogging and the 'tram lines' symptom, also characteristic of waterlogging, was observed in *Cornus florida*.

Heat stress
In some subjects damage from the localised high temperature stress was evident as darkening of the roots on the side of the container which was heat-treated. In *Berberis* the roots were grey compared to their normal yellow colour, in *Viburnum* orange compared to their normal creamy yellow colour. In *Forsythia*, *Garrya* and *Cornus florida* they were brown, the most marked reaction being in the *Cornus*.

There was no visible damage in *Ceanothus*, *Rhododendron*, *Magnolia*, *Elaeagnus*, *Cotinus*, and *Cornus alba* 'Elegantissima'.

Effects on the shoots
By the time that the plants were examined, some weeks after the end of the short term stress periods, there were no obvious visible effects of any of the stress treatments. One exception was with some of the plants, which had been waterlogged, which showed clear evidence of advanced autumn colour, particularly both *Cornus* species which developed strikingly pink foliage.

Conclusions

This part of the project had two aims. The first was to compare the many adverse conditions to which roots can be subjected in a container, in terms of the severity of their effects on the growth and the appearance of roots and shoots. The second was to identify visible characteristics of roots affected by particular stresses to help nurserymen and advisors diagnose problems of poor growth or loss of quality on commercial nurseries.

The conditions examined can be ranked broadly in the following order of increasingly severe reduction in growth:

1. Unnaturally high organic matter content of the medium (no evidence was obtained in support of the hypothesis that this can cause high rates of root breakdown).
2. Locally high concentrations of CRF granules (which can occur due to clumping or poor mixing).
3. Short term exposure to very high temperatures (which can occur on the south side of containers under certain conditions: roots may turn brown but no evidence that shoot growth is influenced – see Section B for more detailed consideration of this type of stress).
4. Short-term but severe drought.
5. Amounts of CRF up to twice the recommended amounts (even when used on a capillary bed with minimal leaching).
6. Short-term but severe waterlogging.

7. Amounts of CRF more than twice the recommended amounts (when used with capillary irrigation and there is minimal rainfall or overhead irrigation to leach out accumulating salts).
8. Repeated temporary waterlogging (which can occur where uneven and slowly draining beds, combined with excessive overhead irrigation, result in plants standing in 1 to 2 cm of water for a few hours every day).
9. Repeated occurrence of water shortage sufficient to cause slight wilting (which can occur where insufficient attention is paid to supplementary hand watering around the edges of overhead-irrigated beds).

This ranking is intended to reflect the probability of damage occurring on a reasonably well-managed nursery. It is clear that, for any type of stress, the reaction of the plants depends on the severity of the stress, so that the relative damage seen in these experiments depended on the intensity and duration of each type of stress. Considerable attention was paid to matching the severity of the stress treatments to conditions thought likely to occur from time to time on nurseries and in garden centres.

The more severe effect of repeated slight water stress compared to a single severe event is interesting. It seems likely that the explanation has to do with the way plants adapt to a shortage of water by reduced opening of the stomatal pores in their leaves. With repeated occurrence of stress, it is likely that stomata would not have opened fully when rewatered. This would have restricted the uptake of carbon dioxide which is essential for photosynthesis and therefore for growth. With a single severe drought there is much less opportunity for the plant to adapt and therefore the adverse effect is also short-lived as long as the water deficit does not reach the point where some tissues are damaged irreversibly, or severe defoliation induced.

Other adaptive processes were evident in the effects of repeated water shortage on the distribution of growth between roots and shoots and also on the distribution of roots within the container. The weight of roots, and the percentage of the root-ball covered with roots, was reduced much less than shoot growth. This type of adaptation, in which root elongation occurs at the expense of shoot growth to maximise the potential for water uptake, has been observed in many plants (e.g. Krizek *et al.* 1985). In the containers this was accompanied by a trend towards more of the visible roots being over the base of the root-ball, as was seen particularly clearly in *Berberis* and *Garrya*.

Similar adaptation was seen in relation to repeated waterlogging of the base of the container, with root growth in the upper layers being favoured relative to the base. There is no fundamental reason to expect waterlogging stress to impinge on the shoots, as is the case with water shortage. However, severe waterlogging can disrupt plant hormones and reduce the capacity for water and nutrient uptake so that shoots are affected. Clear symptoms of such disruption were seen rarely, the best example being the advanced autumn coloration seen in *Cornus* in response to very severe waterlogging over several days. Only in *Berberis* was there a large and significant reduction in shoot growth.

Since adaptation to the stresses related to under- and over-watering involved opposite effects on the distribution of roots, it is reasonable to presume that conditions which fluctuate between these two problems would have a particularly adverse effect on plant performance. A good root system at the base of the pot that had developed while the plant was under-watered could be rendered useless to the plant if damaged by a few days of serious waterlogging. However, this possibility was not examined experimentally.

The effects of high concentrations of mineral salts ('salt' stress) were the least clearly definable. In these experiments there was little evidence of the expected symptoms such as marginal leaf necrosis ('scorching'), despite the inclusion of an ericaceous plant (*Rhododendron*). Only *Acer palmatum* suffered this sort of symptom at all and then only in the season following the high salt treatment (see Section C for more detail). Instead, there was usually a distinct change in the appearance of the roots, suggestive of poor condition, but no adverse effect on the shoots. It is possible that the change in appearance was actually of no physiological significance, and may even have been the result of reduced allocation of carbohydrates to the root system when water and nutrients are in plentiful supply, leading to a change in the physical structure of the roots. In five out of seven subjects, the mean root cover decreased in response to increased nutrient availability. These findings are consistent with reports for many other species (McConnaughay and Bazzaz, 1991).

The similarity in appearance between salt damage and waterlogging injury was striking and may reflect a causal link that would make it likely that the two factors would interact strongly. Signs of possible salt damage to roots were seen only with capillary irrigation which, in addition to creating the conditions where salts can accumulate, also tends to keep the medium over-wet, unless the height of the water table is adjusted frequently in relation to varying evaporative demand. Such an interaction would help to explain the absence of any signs of damage on roots growing through a band of very high CRF concentration (up to 36 g L^{-1}) in the root boxes, as they were intentionally kept relatively dry to avoid leaching.

The above attempt to draw conclusions about the importance of 'salt stress' serves to highlight the complexities and uncertainties that characterise this area. Much more work, particularly at the strategic level, will be required to unravel the full significance of the term 'salt damage'.

With respect to the second objective, reference has been made already to differences in the appearance and distribution of roots associated with particular stress factors but none of these was sufficiently distinctive and reproducible to provide a firm diagnosis of the stress factor involved. This is not surprising perhaps as the same could be said about attempting to diagnose 'physiological disorders' (which are in essence responses to abiotic stress) from the appearance of shoots. Nonetheless, a useful body of knowledge was gained that forms the foundation for the accumulation of knowledge from practitioners in the field, that has been incorporated into a diagnostic key (Appendix 1). The systematic method of visual assessment of root cover, which was developed for this study and shown to correlate quite well with the weight and length of the entire root system, will help to reduce discrepancies in assessments between observers.

The diagnostic key is supplemented by photographs in Appendix 2 of some typical symptoms. Appendix 2 does not include the very large number of photographs from the observational trials in year 3. The possible value of making these available in a more convenient handbook format is being considered.

SCIENCE SECTION B. High temperature stress on roots

Introduction

In the natural environment, plant root systems are protected from extremes of temperature by the buffering effect of the surrounding soil. This is not the case for plants in containers and, during fine sunny weather, exposure of the container to intense solar radiation may cause rapid increases in the temperature of the growing medium. Inhibition of root development on the south or west sides of containers has been reported in the USA (Ruter and van de Werken, 1988) and many growers in the UK believe that this is a major cause of uneven growth in containers. They describe poorly growing plants in which no roots were visible on the southern side of the container, that is on the side with the greatest exposure to solar radiation. However, such evidence does not prove that the absence of roots on the southern side was the cause of the poor shoot growth because the shoots themselves are exposed to more radiation and warmer, drier air at the edge of a bed. Particularly in a plant that is slightly short of water, this could cause water stress sufficient to reduce growth, and/or visible injury such as leaf 'scorch', or yellowing of leaves from photooxidation. As the stress would be greatest on the southern side of the plant, the injury also would tend to be there.

The aim of this part of the project was to test the hypothesis that high root temperature is a significant stress on plants in containers in the UK and to identify threshold temperatures to allow growers to determine whether they need to take steps to reduce container temperatures.

Measurements made in 1996 showed that the temperature of the growing medium on the southern side of a 2-litre container could reach almost 50 °C. While this was a brief and localised maximum, the temperature was above 40 °C for 5 hours.

By reference to published studies, it was clear that these temperatures are sufficiently high for root damage to be a possibility but not a certainty. Temperatures of 42 °C reduced plant survival of elm (*Ulmus parvifolia* 'Drake') by 50 % (Martin *et al.*, 1989) and root and shoot dry weight were reduced significantly in *Magnolia grandiflora* 'St. Mary' by repeated exposure to similar temperatures (Martin and Ingram, 1991). During a five-day exposure period, root damage occurred at 40°C in *Buxus microphylla japonica* and at 35°C in *Berberis thunbergii* 'Atropurpurea' and *Pittosporum tobira* 'Wheeler' (Newman and Davis, 1988). Effects of such heat stress to roots included reduced growth, leaf wilting and chlorosis, altered branching habit and inhibition of flower initiation (Ingram *et al.*, 1989).

In attempting to separate the effects of high root temperature from other factors, many workers have immersed containers in a water bath. Unfortunately this does not simulate effectively the localised high temperature created by solar warming. This section reports experiments in which specially developed apparatus (using radiant electric heaters to simulate the effect of solar radiation) was used to simulate the most extreme heat stress likely to occur in the UK.

Compared with the use of natural conditions, this approach has two important advantages:

- High root temperatures can be imposed without any alteration of the environment of the shoots so that effects of root temperature can be separated from other effects of bright sunshine such as high leaf temperature and/or water stress.
- The treatment can be defined precisely and reproduced, allowing the effect of a 'worst possible case' to be tested.

A prototype system achieved temperatures up to 50 to 55 °C at the surface of the pot after two to three hours while the core temperature remained at about 20 °C. This was enough to cause browning of roots on the hot side but no effect on shoot growth was detected. In the final year, thermostatic control was introduced and used to expose roots to 50 °C for 4.5 hours, and pots were rotated to simulate the change in the position of the sun over the course of the day. This resulted in massive death of roots but again this was not reflected in shoot growth of *Elaeagnus pungens* 'Maculata' or *Berberis stenophylla*.

Materials and Methods

General methods

Temperature measurements

Temperature was measured with electrical sensors connected to a data logger (thermistors connected to a Delta-T Devices DL2 logger, or platinum resistance sensors connected to an in-house system). For measurements of temperatures in containers, probes were inserted into the growing medium vertically, taking care that the sensitive part of the probe was at the desired depth.

Year 1 (1996): Temperature measurements on outdoor container beds

Probes were inserted to a depth of 5 cm into 2 litre containers in which *Cotinus coggygria* plants were growing. The plants were part of the first environmental stress screening experiment. They were spaced at about 26 x 26 cm (i.e. about 10 cm between adjacent pots) on a capillary sand bed.

The locations of probes were:

- In a container of peat:bark mix placed near the centre of the bed -
    - one probe on the south side
    - one probe in the centre
    - one probe on the north side
- In a container of the 90% vermiculite mix placed near the centre of the bed
- On the south side of a container on the southern edge of the bed
- On the south side of a container placed on concrete hardstanding

Air temperature was also recorded by a nearby, screened air temperature probe. Measurements were made from June to October, 1996.

Year 2 (1997): Controlled temperature experiments

Prototype temperature control apparatus

The prototype apparatus used freestanding 1 kW domestic radiant heaters to achieve unilateral heating of containers. Aluminium foil, fixed to a weldmesh support, reflected radiation away from the shoots and concentrated it on the containers (Figure 21). With two heaters placed side by side, it was possible to treat up to six plants in 2-L pots at one time. The apparatus was operated in a constant temperature room so that the air temperature around the shoots was maintained at 20 ± 2 °C. Artificial light was provided by two metal halide lamps (400 W) 2.5 m above the plants.

The performance of the system was monitored using a container, filled with growing medium but without a plant, into which eight temperature probes were inserted at the locations shown in Figure 22. In preliminary tests, the temperature was monitored for four hours with the heaters on continuously. The data in Figure 23 and Figure 24 show that the temperature close to the surface facing the heaters tended to plateau at about 55 °C while the temperature in the centre reached only 25 °C. The variation in temperature around the circumference of the pot, evident in Figure 24, provided a means of relating visible root damage to the temperature to which individual roots were exposed. The temperature data are considered in more detail in the results section below.

Plants were watered generously and placed in the constant temperature room for 24 hours. They then received a single exposure to the high temperature treatment illustrated in Figure 23 and Figure 24.

Figure 21. Diagram of the prototype apparatus used in 1997for simulating solar heating of containers.

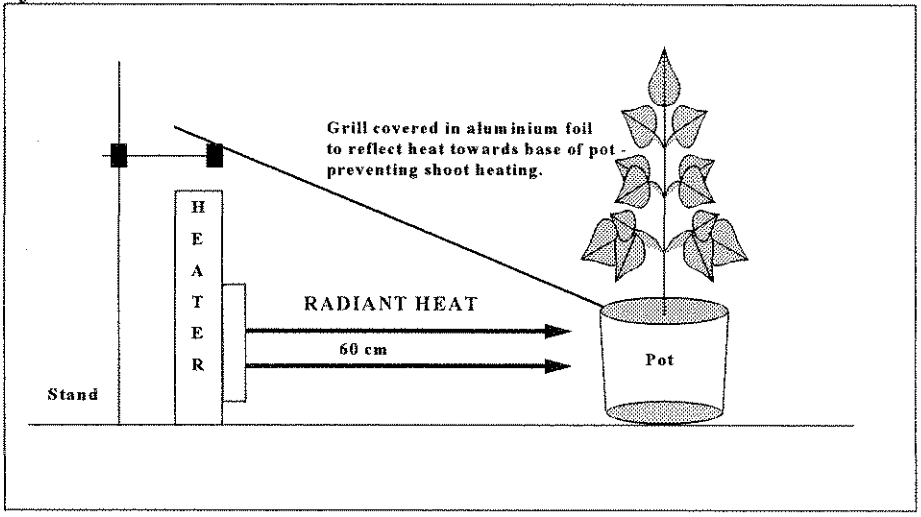


Figure 22. Plan view of pot (2 litre) to show positions of temperature probes in relation to the source of radiant heat, and the labels used to identify them.

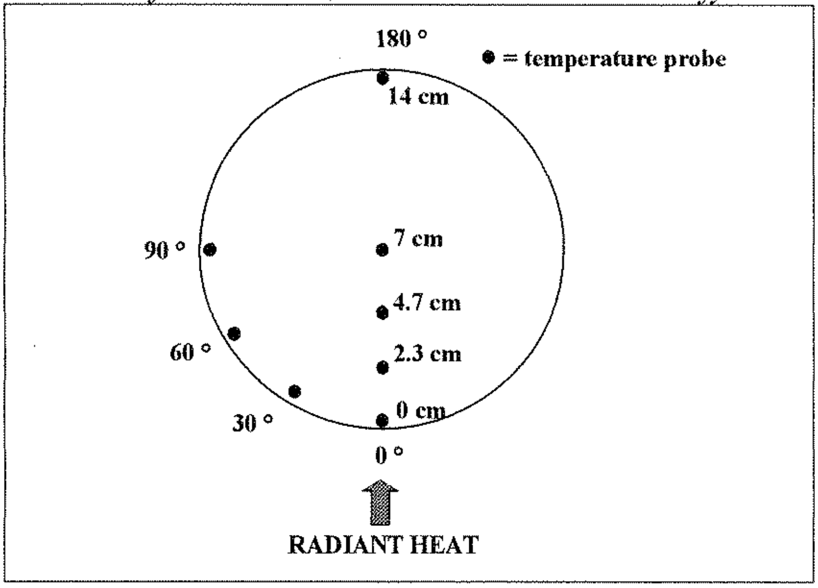


Figure 23. Heat transmission into the container as evident from changes in temperature at different distances from heated face (see Figure 22 for further explanation of probe positions).

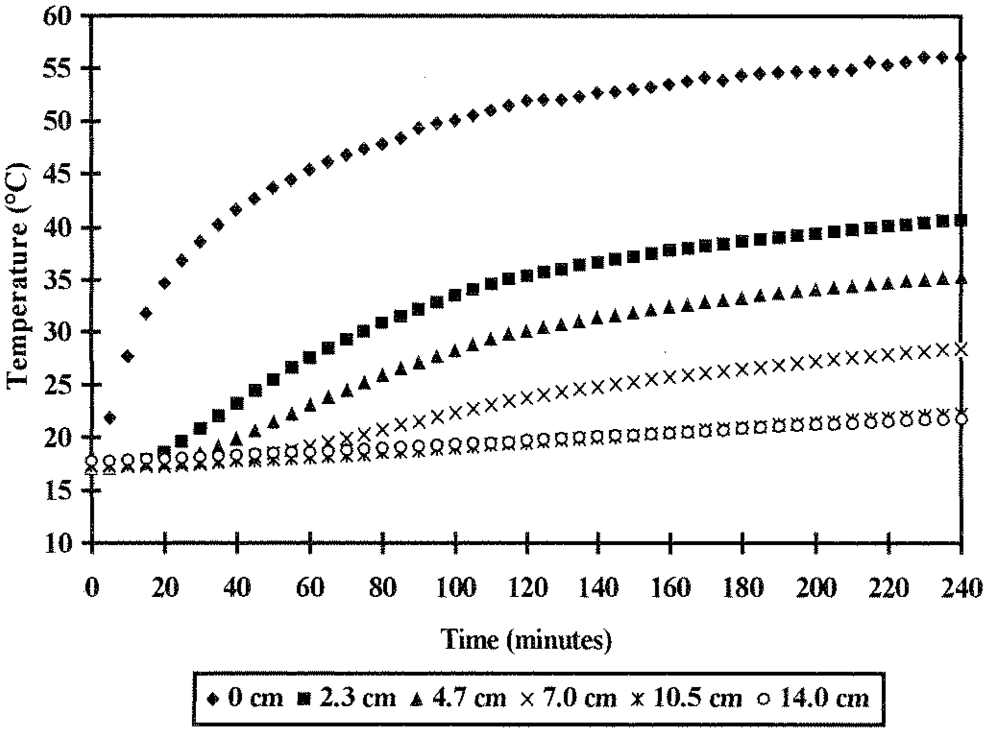
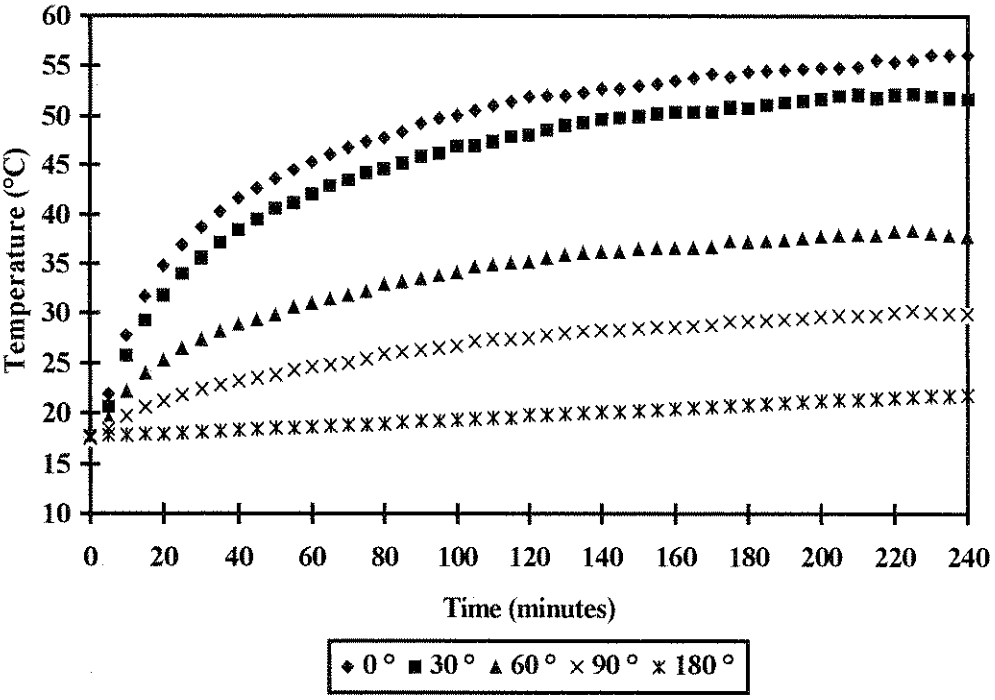


Figure 24. Time course of temperature at various positions around the circumference of a container exposed to radiant heat on one side , identified by degrees of rotation away from the heated face (see Figure 22 for further explanation of probe position).



Plant material and cultural conditions

Plants tested with the prototype apparatus were as follows:

- *Garrya elliptica* 'James Roof'
- *Daphne* x *burkwoodii* 'Somerset'
- *Rhododendron* 'Elsie Straver'

Rhododendron 'Elsie Straver' was selected because it is reputed to be particularly prone to leaf tip necrosis when growing in a container in a location conducive to high root temperature.

The *Rhododendron* plants were purchased in July 1997 and repotted immediately into 2 litre containers. The *Garrya* were derived from cuttings, direct-stuck in August 1996. The *Daphne* plants were micropropagated in May 1996, potted-on into 1 litre containers in September and grown-on under glass at 20 ± 5 °C until June 1997.

The growing medium was 70/30 (v/v) peat / bark with 3 g L^{-1} Ficote 180 (16%N: 10% P_2O_5: 10% K_2O; w/w/w) and FTE added.

Year 3 (1998): More severe controlled heat-stress treatments

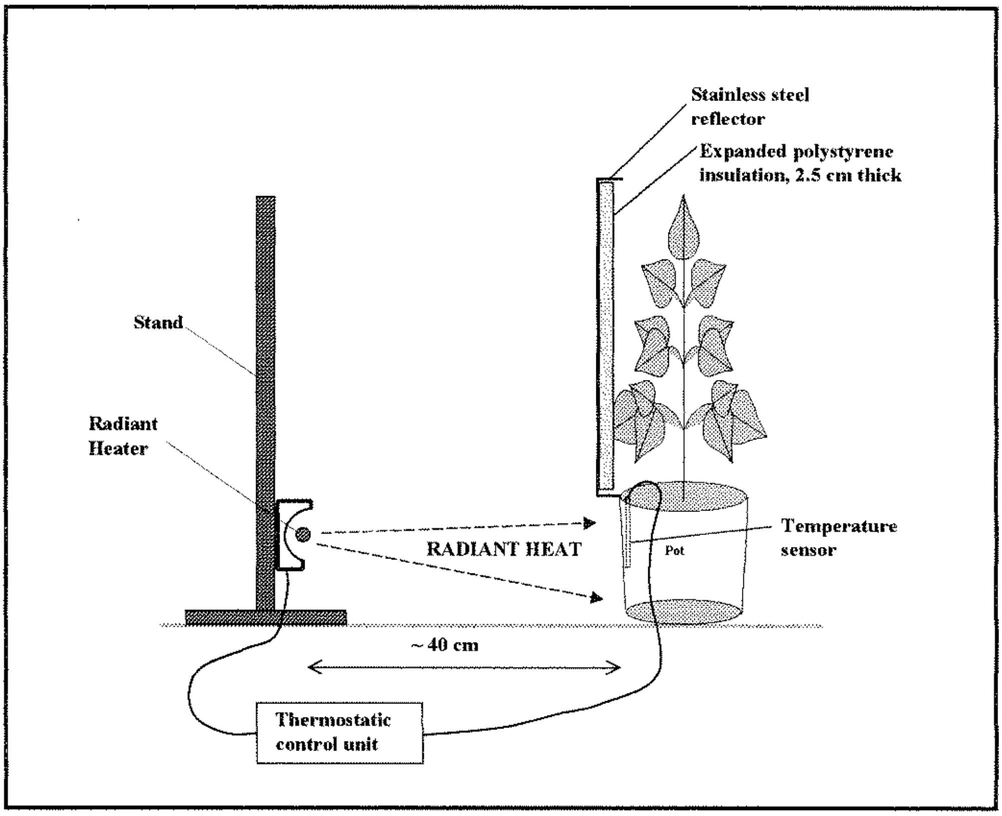
Improved controlled soil temperature apparatus

The prototype apparatus showed that it was feasible to use electrical radiant room heaters to simulate the localised heating of containers that occurs outdoors as a result of solar radiation. However, although the heat output of the heaters is fixed and reproducible, the temperatures reached in the containers was highly sensitive to the distance between the heater and the container, and any factor (such as the composition, degree of compaction, and water content of the medium) which has an influence on the thermal properties of the growing medium.

To avoid this difficulty, an electronic temperature controller was added (type CVR/H/P2, from Clare Instruments, Nobel Engineering Division, Goring by Sea, Worthing). The controlling sensor was a platinum resistance probe, 6 mm diameter, inserted into the medium as close as possible to the heated face of one of the containers. This could have been one of the treated plants but, to avoid physical damage to roots that later would be inspected for heat stress damage, a container of the same medium without a plant was used instead.

The free standing heaters used in the prototype were replaced by surface mounted units (Dimplex IRX, 1000 kW) attached to a substantial galvanised steel framework that allowed their height to be adjusted to achieve optimum heat transfer (Figure 25 and Plate 23). With these heaters 40 cm from the front face of the containers, temperatures greater than 60 °C would be reached if not limited by the temperature controller.

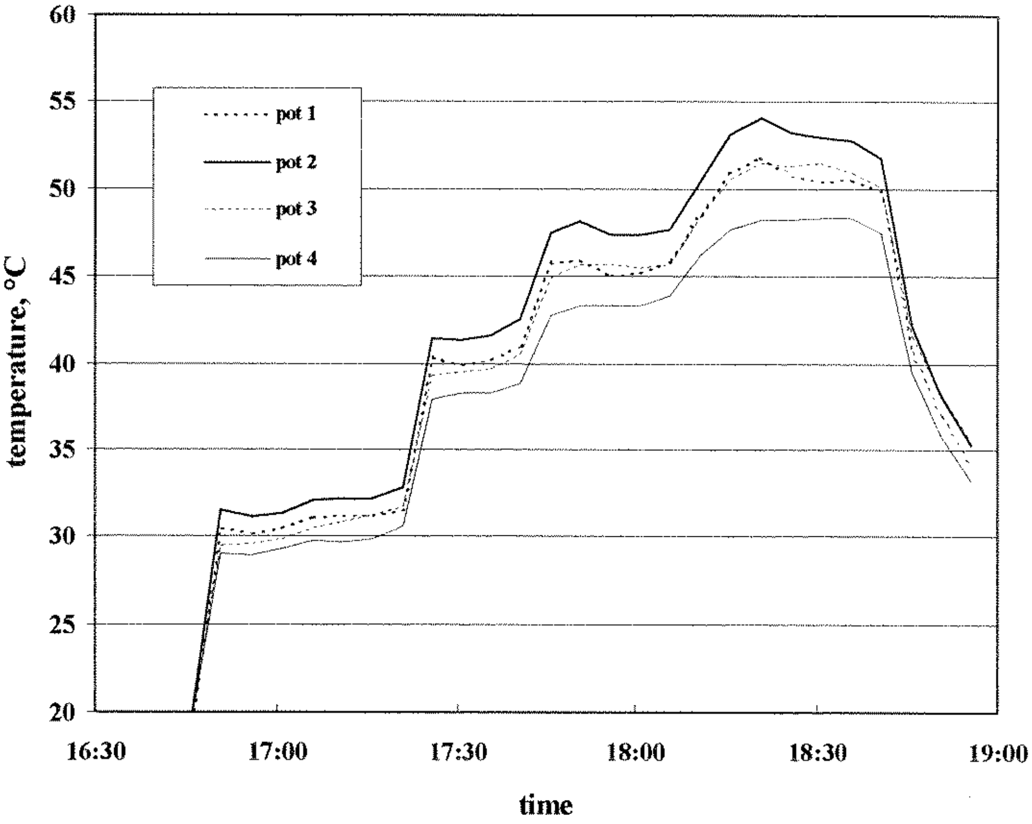
Figure 25. Schematic diagram of current version of the apparatus for simulating the heating of containers by solar radiation, as used in 1998.



The rate of temperature rise was controlled by increasing the set point of the temperature controller in a series of steps. Figure 26 shows a typical time course. To achieve a smooth increase in temperature, it would be necessary to use a relatively complex programmable temperature controller. Figure 26 shows that there was some variation in temperature between replicate containers, some of which was associated with position in the row. It might be possible to reduce the decline in temperature at each end of the row by adding reflective panels at each end but some variation in the thermal properties between replicates is unavoidable and is bound to be reflected in the temperatures reached. Clearly there is scope for further refinement of the apparatus but, even in the configuration used here, it provided the means to apply a severe high temperature stress in a predictable and reproducible way, something that would have been impossible under natural conditions.

More effective isolation of the shoots of treated plants from any heating effect was achieved by the combination of a reflective metal screen backed by a sheet of expanded polystyrene, 2.5 cm thick. Held vertically by stands at each end, it was placed as close to the plants as their branches would permit, as shown in Figure 25.

Figure 26. Time course of the stepped increase in temperature used in 1998. Temperature sensors were inserted 5 cm into the rooting medium and in contact with the side of the container facing the heat source. Pots 1 and 4 were at the two ends of the apparatus, 2 and 3 were central.



The heat treatments

The 1997 experiments showed that the superficial roots were damaged visibly by a single experience of temperatures in the range of 40 to 55 °C, yet there was no effect on shoot growth. Higher temperatures are most unlikely to be experienced in containers outdoors in the UK, but plants could be exposed repeatedly to lower but still damaging temperatures. Furthermore, as the direction of the radiation from the sun changes over the course of the day, the damage could extend to about one quarter of the circumference of the container. Therefore, the intention in 1998 was to create similar temperature maxima but to apply the treatment repeatedly and to rotate the pots over the course of the treatment to simulate the movement of the sun, as outlined below:

First face:	30 min 30°C
	20 min 40°C
	35 min 45°C
	90 min 50°C
Second face (45° to the first):	90 min 50°C
Third face (90° to the first):	90 min 50°C

Treatments were imposed on sets of four plants per day at two-week intervals (3-5/2/99, 17-19/2/99, 3-5/3/99, and 17-19/3/99).

Non-stress controls were moved from the growth chamber to an area adjacent to the heat stress apparatus so that any effect of the change in aerial environment would not be confounded with the heat stress.

Interaction with nutrient level

One of the ways in which damaged roots may influence the growth and development of shoots is through an effect on nutrient uptake. Even if a large proportion of the root system has been lost, growth of the shoots may not be affected if the availability of nutrients is such that the remaining roots can absorb all that is needed. On the other hand, the immediate effect of damage to cell membranes in the roots caused by the heat stress will be to allow nutrient ions to be drawn in with the transpiration stream *more easily* than through living roots. How long that situation lasts depends on how quickly the xylem in the injured roots becomes blocked; this could take several days. If the nutrient concentration in the medium is high, it is possible that nutrients could accumulate to damaging levels in the leaves, particularly in plants which are sensitive to 'salt damage'. To test for such interactions between temperature stress and nutrient availability, the amount of fertiliser in the mix was kept moderately low and half of the plants received additional liquid feed as detailed below:

Low nutrient: 3 g L^{-1} Ficote 140, irrigated with mains water
High nutrient: 3 g L^{-1} Ficote 140, fertigated with Peter's Professional at 500 mg nitrogen L^{-1}

Treatment combinations

Table 12 summarises the treatment combinations applied and the codes used to identify them.

Table 12. List of treatments and the codes used to identify them

Treatment code	Heat treatment	Irrigation / fertigation $^{+}$
+H-F	6 hour cycle x 4 occasions	Irrigation
+H+F	6 hour cycle x 4 occasions	Fertigation
-H-F	No heat stress	Irrigation
-H+F	No heat stress	Fertigation

$^{+}$ From the time of the first heat treatment, plants were irrigated with either mains water or a high concentration of complete nutrient solution (Peters Professional) containing 500 mg L^{-1} N.

Replication

For each species, there were three replicate plants of each treatment in a completely randomised design.

Plant material and cultural conditions

Plants of *Berberis stenophylla* and *Elaeagnus pungens* that had been propagated from cuttings in late 1997 were potted-up into 2 L pots of 70:30 peat:bark medium containing 3g L^{-1} Ficote 140 and overwintered under glass. In mid January, plants were placed in a growth chamber at 20 ± 2 °C, 130 μmol m^{-2} s^{-1} PPFD, where they remained until the end of the experiment. The *Berberis* plants were cut back to stimulate new growth.

Measurements of plant response

Roots at the edge of the pot were inspected and photographed, on 8/3/99, after the plants in the +H treatment had been heat-treated three times.

The total extension growth of all shoots was measured at weekly intervals. This was facilitated with the *Elaeagnus* plants by marking the point 5 cm below the tip of each growing shoot at the start of the experiment. This was not necessary with *Berberis* because there were no elongating shoots at the start of the experiment, the plants having been pruned recently.

Stomatal conductance

Stomatal conductance was measured using a steady state diffusion porometer (PP Systems) just before starting heat treatment and on five occasions over the following 30 days. Measurements were confined to *Elaeagnus pungens* 'Maculata', 6 leaves per plant were measured on 3 replicate plants of each treatment.

Chlorophyll fluorescence

Various stresses can disturb the function of the photosynthetic apparatus of the leaves in a way that is reflected in altered patterns of fluorescence from the chlorophyll molecules involved in capturing the light energy that drives photosynthesis. To determine whether heat stress on the roots had any short term effect on this aspect of plant productivity, chlorophyll fluorescence was measured on the samples of leaves used for stomatal conductance. Fluorescence was measured with a Hansatech FMS following a 20 minute period of dark adaptation.

Leaf nutrient analysis

Samples were taken for mineral analysis 27 days after the first exposure to the high temperature treatment. Samples were individual whole leaves which were weighed, oven-dried and then acid digested prior to the determination of Ca, Cu, Mn, Mg, N, P, K, Na, and Zn. For each treatment, one of the fully expanded leaves used for stomatal conductance measurements, and one expanding leaf, were taken from each plant.

Root electrolyte leakage

Root electrolyte leakage provides a measure of the integrity of cell membranes and was used to make an objective assessment of the severity of physiological disruption caused by the high temperature treatments. It was measured on three replicate plants of *Elaeagnus pungens* 'Maculata' in each treatment. With the heat treated plants, a sample was taken from the brown roots on the side which had faced directly towards the radiant heat source (the 'hot side') and a separate sample was taken from the opposite side where the roots remained white and healthy looking (the 'cool side').

Samples were taken by removing the container and cutting out from the exposed surface a sample of about 500 mg of main root segments, 40-80 mm long, and with a representative range of diameters. Any medium attached to the roots was removed before the roots were weighed, cut into 10 mm lengths and placed in universal bottles containing 20 ml purified water.

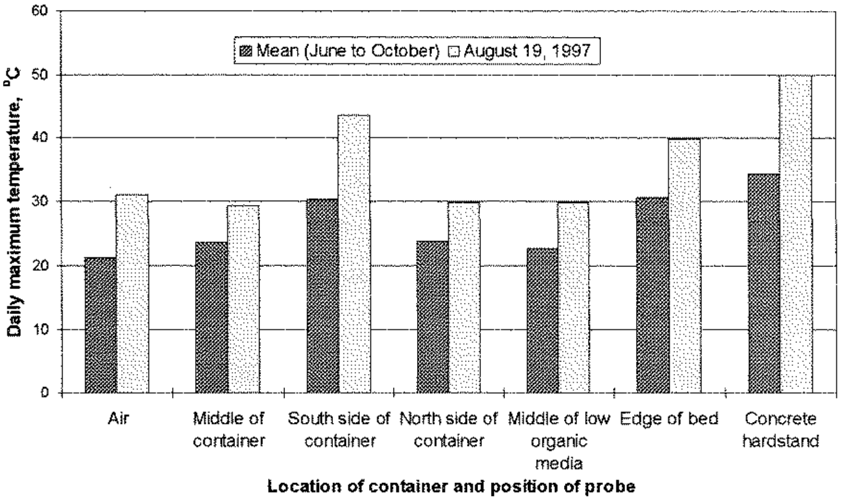
The samples were held at 22 °C for 17 hours before the bottles were transferred to an autoclave and brought to 115 °C to kill all cells and thus allow free exchange of electrolytes with the bathing solution. The electrical conductivity (EC) of the bathing solution was determined after one hour, 17 hours and after autoclaving, as soon as the vials had cooled to room temperature. Conductivity was measured with a Bibby SMC1 conductivity meter (J Bibby Science Products Ltd, Stone, ST15 0SA).

Results

Year 1: Temperatures recorded in containers outdoors (June – October, 1996)

Daily maximum temperatures recorded in each position averaged over the entire period, together with the absolute maxima recorded on 19 August, are shown in Figure 27.

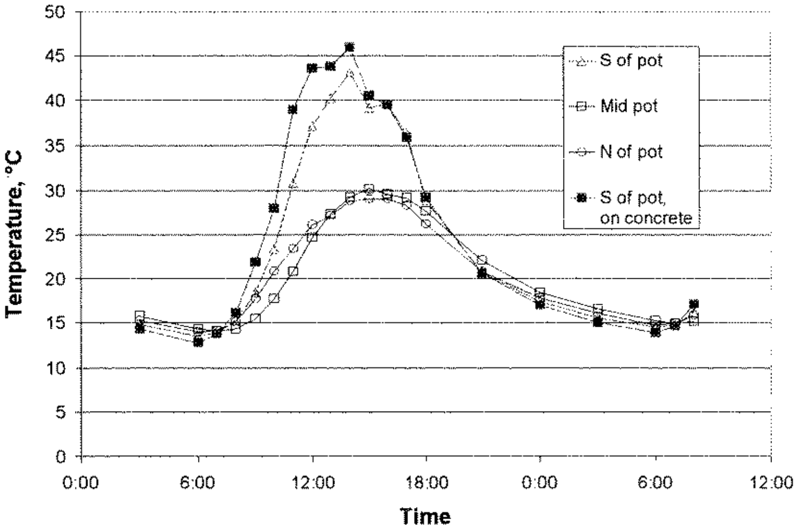
Figure 27. Comparison of maximum temperatures recorded in the growing medium of Cotinus coggygria 'Royal Purple' plants growing in 2 L black plastic containers on a capillary sand-bed or on an adjacent concrete surface.



Placing containers on the concrete hardstanding resulted in the highest temperatures, with a maximum of close to 50°C. The dry concrete surface heated up much more than the moist sand, because of the absence of evaporative cooling, and some of this heat would have been transferred to the pot by convection and radiation. Similar results would be expected from other dry standing surfaces such as gravel. Temperatures substantially above air temperature were confined to the south side of the container but, with the plants spaced 15 cm apart, there was no evidence that temperatures were higher in the containers at the southern edge of the bed, despite slightly greater exposure to solar radiation.

On the hot days such as 19 August, the temperature on the south side of the container was above 40°C for almost 5 hours (Figure 28). The temperature cycle observed on 19 August, in the container standing on concrete, was taken as a model for the controlled heat stress treatments.

Figure 28. Hourly temperature measurements at 5 cm depth in 2 L black plastic containers of peat:bark medium on the hottest day in 1996 (19 August).



Year 2: Effects of controlled high root temperatures using the prototype apparatus

A typical time course of temperature at various locations in the container over the course of four hours' exposure to radiant heating in the prototype apparatus is shown in Figure 23. After two hours, a maximum temperature of 52 °C was recorded at the front of the container (i.e. the side facing the heaters, labelled 0 cm). A further two hours of heat increased the temperature by only a further 4 °C. This was slightly greater than the maximum observed outside in 1996 and was therefore appropriate for a severe test of whether roots in containers are liable to be damaged by high temperatures, under extreme conditions, in the UK.

Comparisons between the lines in Figure 23 indicate the steepness of the temperature gradients that existed across the container. At 2.3 cm away from the surface, temperatures reached a maximum of just below 40 °C, already 12 °C lower than at the surface but still high enough for root damage to be expected.

Looking at the decline in temperature around the circumference shown in Figure 24, there was little change in temperature over the first 30° of rotation. This implies that about one sixth of the circumference (i.e. 2 x 30 / 360) would have experienced temperatures of or near 50 °C. Beyond an angle of 30°, the temperature declined rapidly as the radiant energy was spread over a rapidly increasing area, such that, at an angle of 60° away from the front face, the maximum temperature was 35 °C.

Plates 15 and 16 show the roots of *Daphne* x *burkwoodii* 'Somerset' from a control plant and a plant exposed to a single four hour heat treatment. Within 24 hours, severe root discoloration was observed at the front of the pot (Plate 17), extending to 45 ° away from the front face. From the temperature profiles, this is estimated to correspond to the zone where temperatures exceeded about 45 °C. These values are similar to the maximum recorded in containers outside (Figure 27). After one month, roots of *Daphne* were still discoloured but did not appear to be dead. The results with

Rhododendron 'Elsie Straver' were similar. The extent of dysfunction of these discoloured roots was not assessed in these experiments.

Garrya elliptica 'James Roof' suffered pronounced root discoloration where temperatures were estimated to have exceeded about 38°C (Plates 18 &19). Furthermore, roots 2 cm to either side of the front of the container exhibited severe blackening, suggesting that the severity of damage to cell membranes increases steeply over the range 50 to 55 °C.

The treated plants, together with non-treated controls, were grown on under glass with supplementary light for several months. Observation of their appearance showed no evidence that the root damage caused by the high temperature treatment had any effect on growth or on specific symptoms such as leaf tip necrosis.

Year 3: Effects of repeated exposure to high root temperatures

Appearance of roots

After a single exposure to the heat stress treatment roots on the heated side of the container were very clearly brown compared with those on the cool side of the container and those on control plants. The colour of the majority of these undamaged roots was white in *Elaeagnus* but pale yellow in *Berberis*. When next inspected, four weeks later, and after a further two exposures to the heat stress cycle, the heated roots had become a darker brown but there was no evidence that they were decomposing (Plate 24).

Shoot growth and development

In *Elaeagnus*, shoots continued to grow in all treatments and there was no significant difference in average extension rate between any of the treatments (Table 13). The large least significant difference (LSD) values reflect that the plants were rather variable and that, therefore, the experiment would not have been able to detect small effects. However, there were no visible signs of any adverse effects on the leaves or the shoot tips, such as marginal necrosis or yellowing.

Table 13. The effects of high root temperature stress, with and without a high concentration liquid feed (±'Fert.'), on shoot growth of Elaeagnus pungens *'Maculata' over a period of 46 days. All shoots >5 cm at the start of the experiment were measured. Neither the main effects nor the interactions were statistically significant.*

	Heat – stress treatment		Non-heated control		LSD
	+ Fert.	– Fert.	+ Fert.	– Fert.	
Mean extension per measured shoot, cm	2.72	2.94	2.94	2.93	1.71
Mean extension per growing shoot (i.e. > 0.5cm growth), cm	3.78	4.69	3.96	3.70	3.05
Number of growing shoots	10.0	10.7	9.0	9.0	5.6

Similarly, effects on growth were not detected in *Berberis stenophylla*. In this case growth was from lateral shoots which did not start to grow until after the heat treatments had been applied at least once. The numbers of shoots that developed were very variable but there was no evidence that they were affected by the high temperature stress or by the application of a high concentration of mineral nutrients (Table 14).

Table 14. The effects of high root temperature stress, with and without a high concentration liquid feed (±'Fert.'), on the development and growth of new lateral shoots in Berberis stenophylla. *All shoots developed after the first application of the heat stress treatment. Neither the main effects nor the interactions were statistically significant.*

	Heat – stress treatment		Non-heated control		LSD
	+ Fert.	– Fert.	+ Fert.	– Fert.	
Total shoot extension per plant, cm	273	297	329	288	173.5
Numbers of growing shoots	17.7	15.3	14.3	19.0	11.8

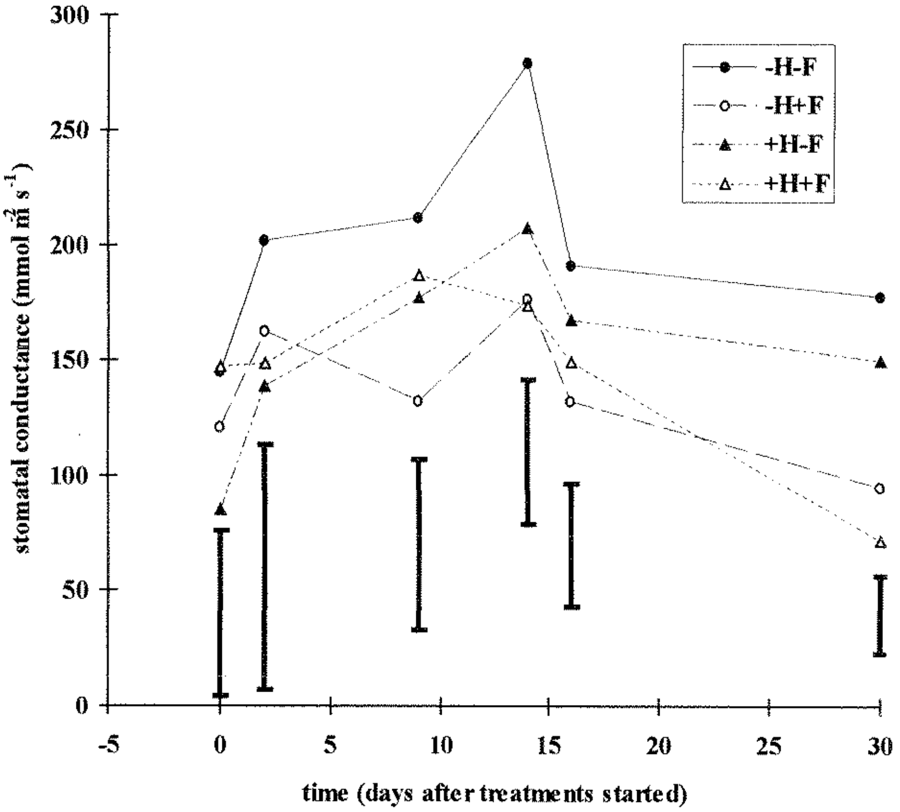
Stomatal conductance

Any damage to the root system that reduces its capacity for water uptake is likely to be reflected in the water status of the leaves and/or a reduction in the apertures of stomatal pores. Stomatal conductance was therefore measured before and after heat treatment to look for early indications of the effect of the treatment on the shoots.

The results in Figure 29 show that stomatal conductance increased between the measurement made prior to treatment and the first measurement made after heat treatments had been applied, but the size of the change was not related to treatment. The increase was smallest in the 'heat stress + fertigation' (+H+F) treatment, but the difference was not significant and had largely disappeared when measured one week later. Stomatal conductances decreased following the second heat stress cycle but again this was not related to the heat stress treatment. The most probable cause of the day-to-day fluctuations observed in all treatments is slight changes in the time of day when the measurements were made or in the carbon dioxide concentration in the growth chamber.

However, the results show that, over the longer term, fertigation caused progressive reduction in stomatal conductance, reaching significance after 30 days. There is a suggestion that heat stress may have exacerbated this effect but the difference was not quite large enough to be significant in this experiment.

Figure 29. Changes in the stomatal conductance in leaves of Elaeagnus pungens *'Maculata' before and after the start of heat stress (+H) and high fertigation (+F) treatments. Plants in the +H treatments were subjected to a 6-hour heat stress cycle on day 1 and day 15. Values are means and error bars represent the LSD for each measurement date.*

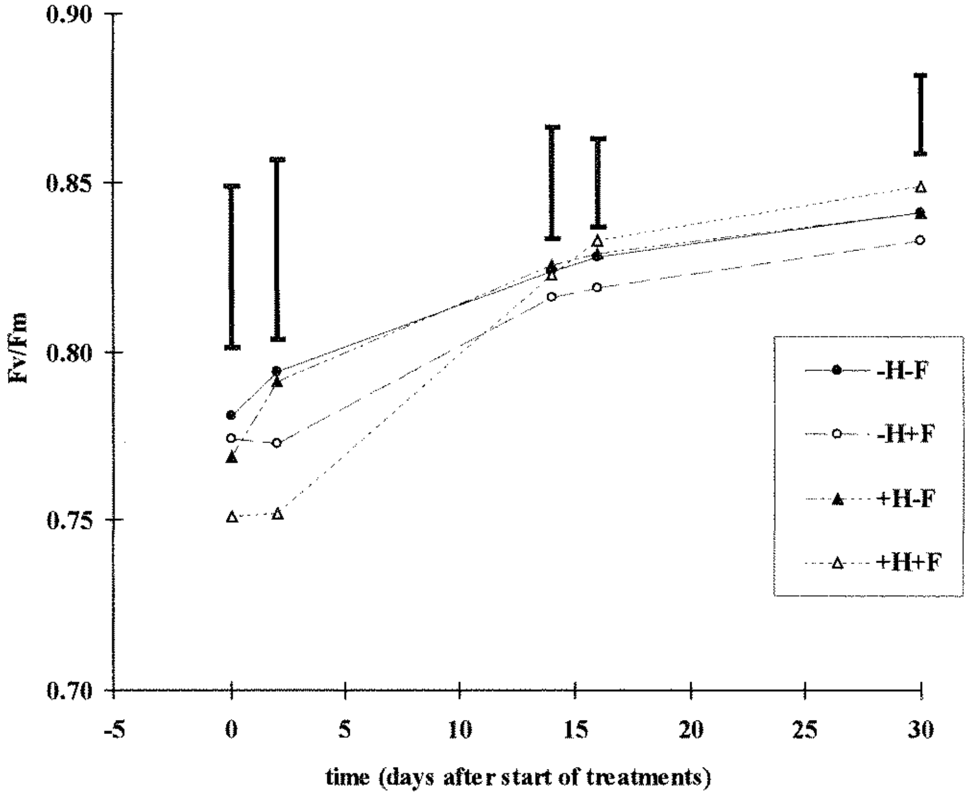


Chlorophyll fluorescence

Chlorophyll fluorescence provides a simple and non-destructive test of the condition of the part of the photosynthetic apparatus concerned with the capture and transfer of light energy. As such it can be useful for the detection of many types of stress, particularly cold stress. It was therefore of interest to see whether it would detect the effects of heat stress on the roots. Results are expressed as the ratio of two parts of a fluorescence time course, variable fluorescence / maximum fluorescence or F_v/F_m.

The results in Figure 30 show that there was no abrupt change associated with heat treatment nor any significant difference due to heat or fertigation treatment at any stage. There was a clear trend of increasing F_v/F_m over the course of 30 days, which is likely to have been due to adaptation to the light environment in the growth chamber.

Figure 30. Chlorophyll fluorescence data for leaves of Elaeagnus pungens *'Maculata' before and after the start of heat stress (+H) and high fertigation (+F) treatments. Plants in the +H treatments were subjected to a 6-hour heat stress cycle on day 1 and day 15. Values are means and error bars represent the LSD for each measurement date.*



Leaf nutrient analysis

Any damage to roots which affects either their ability to absorb nutrients or their ability to selectively exclude some ions, such as sodium, is likely to affect the concentration of nutrients in the shoots and leaves, particularly those parts formed after the damage occurred. Therefore, samples of newly expanded leaves, and of leaves that were present at the start of the experiment, were analysed for a range of mineral elements. The results are shown in full in Table 15.

Effects of leaf age

Young leaves had generally lower concentrations of mineral elements than the larger older leaves. Averaged over all elements, the concentrations in the young leaves were 55% lower than in the older leaves, the difference being significant for all elements except potassium, and particularly large for sodium (young leaves 17% of old leaves). However, there were no significant differences in the responses of the two leaf types to the heat stress or fertiliser treatments.

Effects of heat stress

On average, heat stress reduced mineral concentrations by 12%, the largest effect being on manganese (Figure 31). There was no evidence that heat stress resulted in unrestricted uptake of any element, including sodium, despite the relatively high concentration of sodium in mains water at East Malling.

Effect of fertigation

Addition of nutrients to the irrigation water significantly increased the levels of the trace elements manganese and zinc, as well as the macronutrients (N, P, and K). The largest effect was on phosphorus (Figure 32).

Table 15. The effects of heat stress of roots and fertigation on the concentrations of nutrient elements in expanding ('young') and fully expanded ('old') leaves of Elaeagnus pungens *'Maculata'. Samples were taken for analysis 27 days after the first exposure to high root temperatures.*

Element	YOUNG LEAVES				OLD LEAVES			
	0 mg fertiliser		500 mg fertiliser		0 mg fertiliser		500 mg fertiliser	
	no heat	heat	no heat	heat	no heat	heat	No heat	heat
calcium (%)	0.56	0.48	0.55	0.40	1.18	1.24	1.37	1.31
copper (ppm)	11.2	10.9	12.3	11.0	26.5	21.5	27.5	22.1
magnesium (%)	0.15	0.14	0.15	0.12	0.24	0.26	0.26	0.25
manganese (ppm)	264	147	389	157	476	402	732	486
nitrogen (%)	2.62	2.53	3.79	3.55	3.28	3.36	4.05	4.02
phosphorus (%)	0.18	0.17	0.35	0.29	0.22	0.20	0.44	0.38
potassium (%)	1.73	1.63	2.35	2.06	2.10	1.92	2.37	2.24
sodium (ppm)	83	130	138	76	670	576	694	563
zinc (ppm)	20.3	18.6	24.2	19.9	29.7	28.3	36.6	30.7

Significance levels from ANOVA: -

Element	Heat stress (H)	Age of leaf (L)	Fertigation (F)	HxL	HxF	LxF
calcium	NS	***	NS	NS	NS	NS
copper	*	***	NS	NS	NS	NS
magnesium	NS	***	NS	NS	NS	NS
manganese	**	***	*	NS	NS	NS
nitrogen	NS	**	***	NS	NS	NS
phosphorus	*	***	***	NS	NS	NS
potassium	NS	NS	**	NS	NS	NS
sodium	NS	***	NS	NS	NS	NS
zinc	NS	***	*	NS	NS	NS

Key to significance levels: NS = not significant (P>0.05), * P<0.05, ** P<0.01, *** P<0.001

Figure 31. The effect of heat stress and fertigation treatment on the concentration of manganese in leaves of Elaeagnus pungens *'Maculata'. 'Young' leaves were still expanding when removed for analysis; 'Old' leaves were fully expanded when the heat-stress treatment was started 27 days earlier.*

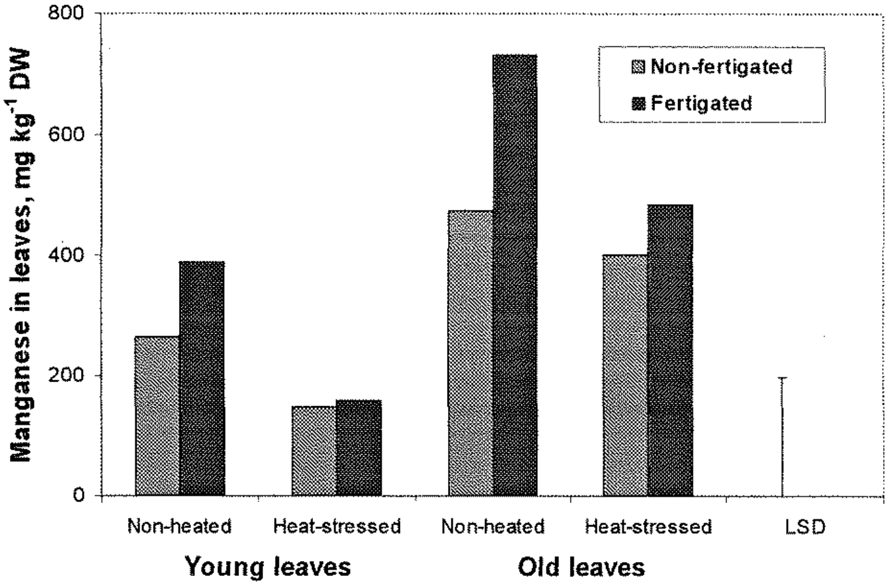
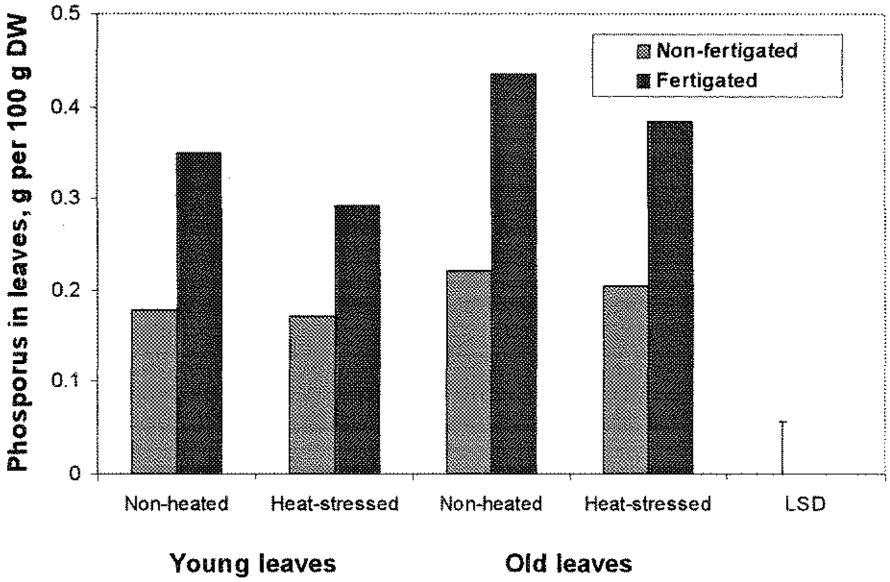


Figure 32. The effect of heat stress and fertigation treatment on the concentration of phosphorus in leaves of Elaeagnus pungens *'Maculata'. 'Young' leaves were still expanding when removed for analysis; 'Old' leaves were fully expanded when the heat-stress treatment was started 27 days earlier.*



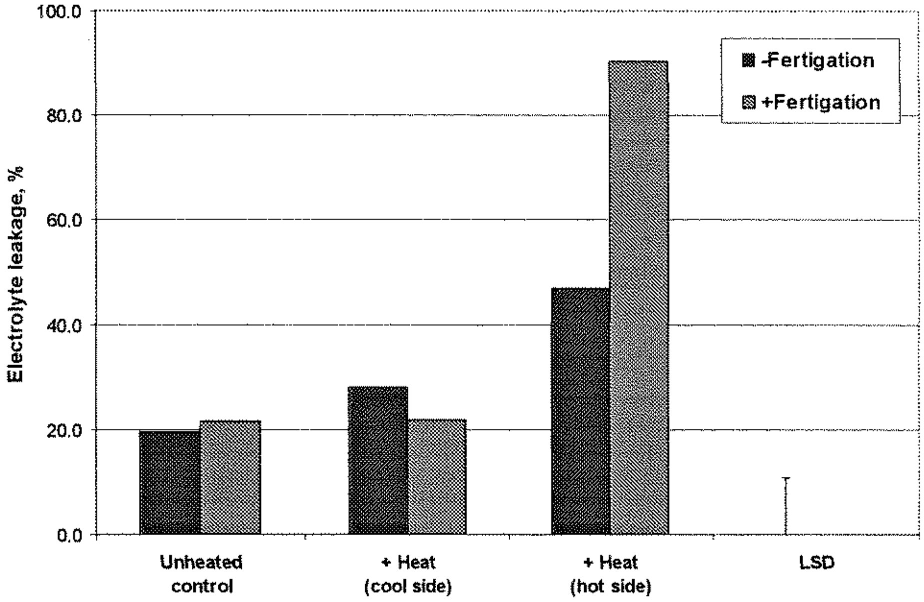
Root electrolyte leakage

In view of the extensive browning of roots on the heated side of the container, it was surprising that there were no obvious effects on shoot growth, the appearance of leaves, or stomatal behaviour. Therefore, to provide an objective test of the severity of physiological damage in the brown roots, electrolyte leakage was measured.

Leakage rate

The proportion of total electrolytes which leaked out of the root samples within the first hour was significantly greater from heat-damaged brown roots than from healthy looking white roots, either from the cool side of the same plant or from non-heated plants (Figure 33). The contrast was greater in the fertigated plants, from which 90% of electrolytes leaked from heat-damaged roots within one hour, compared with 22% from white roots. By comparison, only 47% of electrolytes leaked out in the first hour from the brown roots on non-fertigated plants compared with 20 to 28 % from white roots.

Figure 33. The effects of heat stress and fertigation treatment on electrolyte leakage from roots of Elaeagnus pungens *'Maculata', over the first hour after excision. Roots from the side facing the radiant heat source (the 'hot side') were compared with those from the 'cool side' of the same plants and from unheated control plants.*

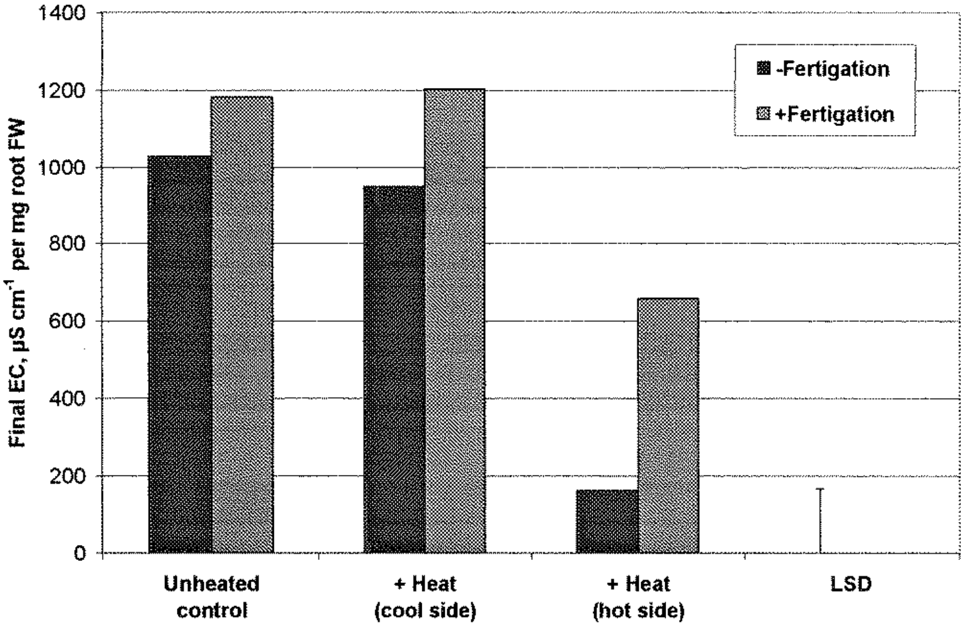


Electrolytes continued to leak from the root segments over the next 16 hours, but the rate was much reduced so that the response trends were similar when measured again 17 hours after excision (data not shown).

Total electrolytes

The total quantity of electrolytes present in the root segments was much reduced in the brown roots from the heated side of the containers ($P<0.001$, Figure 34). The effect was particularly marked in the non-fertigated plants which contained 92% less than white roots from the cool side of the same containers.

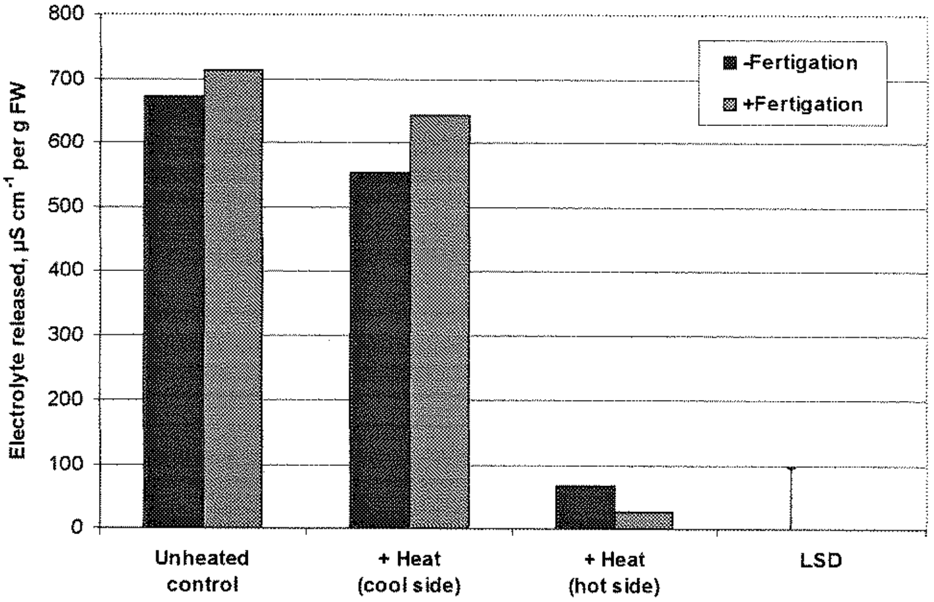
Figure 34. The effects of heat stress and fertigation treatment on the total quantity of electrolytes extracted from roots of Elaeagnus pungens *'Maculata', as indicated by the EC of the bathing solution after autoclaving. Roots from the side facing the radiant heat source (the 'hot side') were compared with those from the 'cool side' of the same plants and from unheated control plants.*



The effect of autoclaving

Autoclaving was used as a means of destroying all cell membranes so that the total quantity of electrolytes could be determined to provide a basis for calculating the percentage leaking out before this disruption. In damaged roots, the quantity of additional electrolytes released by autoclaving also provides a measure of the quantity of electrolytes held by any remaining viable cells, independent of the quantity of electrolytes held passively in the remains of damaged cells. Figure 35 shows that there was some release of electrolytes from brown roots during autoclaving but it was only 5 to 10% of the quantity released from white roots. In contrast to those released at room temperature, brown roots from the non-fertigated treatment actually released more electrolytes during autoclaving than those from the nutrient-rich medium of the fertigated plants. This suggests that there were marginally more viable cells surviving in the non-fertigated roots, and therefore that the slower release prior to autoclaving (Figure 33) was genuinely indicative of reduced damage in the non-fertigated plants, and not an artefact associated with the much smaller total quantity of electrolytes present (Figure 34).

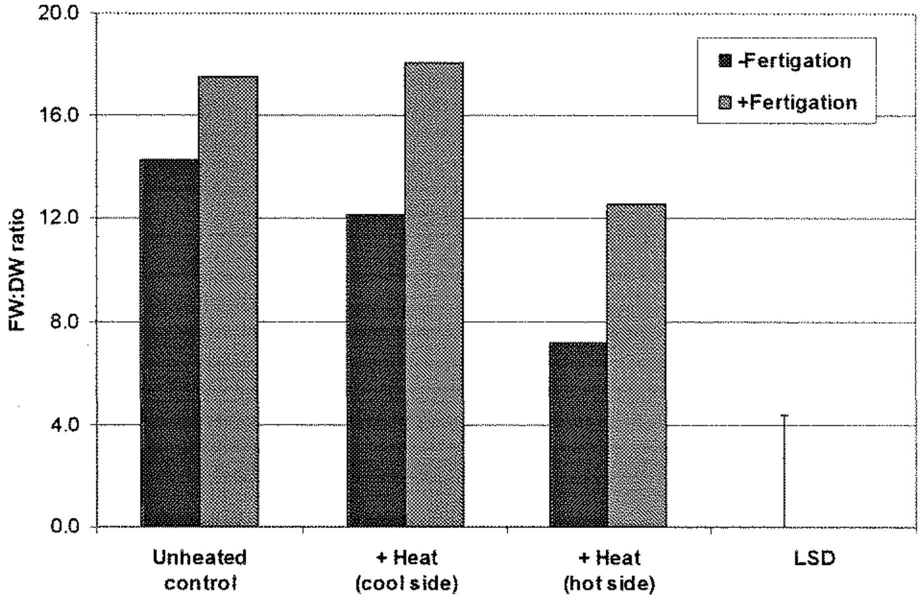
Figure 35. The effects of heat stress and fertigation treatment on the additional quantity of electrolytes released by autoclaving roots of Elaeagnus pungens *'Maculata', following 17 hours immersion in water at 22 °C. Roots from the side facing the radiant heat source (the 'hot side') were compared with those from the 'cool side' of the same plants and from unheated control plants.*



Degree of hydration of roots

The interaction between high temperature stress and nutrition noted in the previous subsection may be related to the greater hydration of the roots from the fertigated plants which was evident in the ratios of fresh to dry weights (Figure 36), and was also suggested by the thicker and more fleshy appearance of the roots on the fertigated plants.

Figure 36. The effects of heat stress and fertigation treatment on the degree of hydration of the roots of Elaeagnus pungens *'Maculata', as indicated by the ratio of fresh to dry weight. Roots from the side facing the radiant heat source (the 'hot side') were compared with those from the 'cool side' of the same plants and from unheated control plants.*



Conclusions from the electrolyte leakage data

The above results are consistent with drastic damage to the membranes of the majority of cells, if not of all cells, in the brown roots. The membranes are responsible for regulating the passage of electrolytes into and out of all living cells so that damage to the membranes will result in the electrolyte concentration in damaged or dead tissues being determined by the concentration in the surroundings. The higher concentrations in the roots of the fertigated plants would therefore be expected, not because the damage was less in those plants but because electrolytes in the nutrient solution would permeate the damaged roots by a simple diffusion process.

The observation that there was an additional release of electrolytes during autoclaving of the brown roots suggests that some living cells survived despite their unhealthy appearance. However, the possibility that autoclaving stimulated release by enhancing purely physical processes, such as interactions with ion-exchange surfaces or simply the rate of diffusion, cannot be ruled out. Furthermore, the amount released was so small that it seems unlikely to indicate survival of more than 10% of the cells of the root, not sufficient to contradict the visual impression that the root as a functional organ could reasonably be considered as dead.

The clear results obtained in this experiment from measurements of electrolyte leakage from small samples of roots suggest strongly that it is a valuable technique for objective assessment of root damage in containers. This is especially true as the method was successful despite the large differences in the nutritional status being tested, which might be expected to complicate comparisons of leakage rates. Further work is required to test electrolyte leakage under a wide range of conditions and in relation to a range of stresses, particularly desiccation and waterlogging. As the test is simple to perform (see page 52), there is no reason why it should not be used by advisors or by growers themselves. A simple conductivity meter would generally be adequate (e.g. Whatman CDM 600, at about £110) and, for small numbers of samples, a pressure cooker could be used in place of an autoclave.

Conclusions

Temperature measurements in containers placed near the edge of outdoor capillary beds, or on adjacent concrete paths, showed that temperatures could reach close to 50 C in the middle of the day. When averaged over six hours, the temperatures were much lower but could still exceed 35 °C.

In the experiment with the prototype apparatus in 1997,a single exposure to temperatures above about 38 °C was sufficient to cause pronounced browning of roots on *Garrya elliptica* 'James Roof', while temperatures above about 45 °C were needed to have a clearly visible effect on *Rhododendron* 'Elsie Straver' and *Daphne* x *burkwoodii* 'Somerset'. However, as there was no obvious effect on shoot growth or the appearance of the foliage, browning of roots at the surface of the root-ball cannot be taken as a reliable sign that roots have been damaged in a way that will reduce plant quality significantly. Indeed, the brown roots on heat-treated plants of *Daphne* and *Rhododendron* had not deteriorated further one month after the heat stress treatment, suggesting that roots may be able to recover from a single isolated exposure to high temperature.

In an experiment with the current, thermostatically controlled version of the apparatus, roots were exposed to high temperatures repeatedly, and a greater proportion of the roots was exposed to temperatures around 50 °C. The result was very marked browning of roots on the parts of the container that had been exposed to these temperatures but, again, effects on shoot growth or leaf appearance were not detected. Measurements of stomatal behaviour and chlorophyll fluorescence did not indicate any disturbance of leaf physiology following heat stress to the roots. However, the small decrease in mineral nutrients in the leaves on heat-treated plants, an average of 12% compared to unheated control plants, suggests that nutrient uptake was reduced slightly by the damage to the roots.

Electrolyte leakage provided convincing evidence that the dark brown roots on the heated side of the container were essentially dead, though the release of a small quantity of additional electrolytes when they were heated to 115 °C in an autoclave suggest that a few cells had survived. To what depth within the root-ball roots were killed was not investigated because of the need to grow the plants on to look for delayed effects. It probably extended a few centimetres, because the stress treatment involved rotating the pot through 90 ° over the course of the day, allowing time for the heat to penetrate the pot much more than in the earlier experiment. Nonetheless it is unlikely that more than one quarter of all roots were killed by the heat treatment.

The lack of obvious effects on shoot growth indicates that, under the conditions of this experiment, the normal root system is larger than necessary to meet the needs of the shoots. The slight reduction in leaf nutrient levels suggests that, if nutrients or water had been in shorter supply, or temperature and light levels had been adequate to support more rapid shoot growth, an adverse effect of heat stress on growth would have been observed.

The cell membranes of the living cells in the outer layers of the root exercise some control on the quantity of individual nutrient ions taken up into the plant. In contrast, the water-conducting xylem vessels are non-living, completely non-selective, and remain capable of transporting water and nutrients until they are physically blocked. These facts led us to test the hypothesis that heat damage sufficient to kill the living cells in the outer layers of the root might cause *increased* and *unregulated* uptake of nutrients until the xylem became blocked. Particularly if nutrient levels in the medium were high, this might be sufficient to cause 'salt stress' in sensitive plants. The results provided no evidence in support of this hypothesis. Once again, the growing conditions could have mitigated against an effect in this experiment because the rate at which ions could potentially accumulate in leaves would be a function of the evaporative demand, which was relatively low in the controlled environment chamber.

It is reasonable to conclude from the results obtained in these experiments that high temperatures may kill some roots in containers in the UK but it will usually be a small proportion of the root system and unlikely, on its own, to be a major cause of poor growth in containers. Further work is required to resolve some of these uncertainties, to extend the approach to a wider range of plants, and to explore interactions with other variables such as container size, water availability and the stage of development of the root system. It is possible, for example, that liners may be at much greater danger than plants in larger containers, despite being exposed to less lateral radiation by virtue of their closer spacing.

The results obtained here strongly suggest that electrolyte leakage is a valuable method for distinguishing seriously damaged roots from those that have become brown due to the natural processes of suberization and sloughing off of superficial cells. As such, it should prove useful in future studies of root behaviour in response to all types of stress. However, it will be important to recognise its limitations. As roots must be able to absorb nutrients, they are inherently more 'leaky' than shoot tissues. The observed rate of leakage will vary depending on the age and stage of anatomical development of the root as well as on the vitality of the tissues. Therefore, it will be important to regard it as a technique for identifying seriously damaged roots rather than for quantifying precisely the degree of injury present.

SCIENCE SECTION C. Medium requirements of *Acer palmatum* and *Daphne* x *burkwoodii*

Introduction

Certain species of HONS appear to perform particularly poorly in containers and are often cited as having root-related problems. Within this category are *Acer palmatum* cultivars and *Daphne* species or cultivars, both highly desirable garden plants where production cannot always meet demand. An initial experiment, involving factorial combinations of two types of medium and two fertiliser rates, showed that the root growth of *D. x burkwoodii* 'Somerset' was strongly inhibited in a typical highly organic medium (70:30 peat:bark) compared to one based on vermiculite. In contrast, the root performance of *Acer palmatum* 'Aureum' was best in the organic medium and benefited also from relatively high fertiliser rate. Further experiments into the requirements of these two subjects are described in more detail below.

Material and Methods (general)

Apart from Experiment 1, plants for all the experiments were produced as follows.

Acer palmatum cuttings were rooted during June 1996 and potted into 1 L containers on 24 August 1996. These were overwintered under polyethylene, graded and potted-on into 2 L containers on 10 July 1997.

Plants *of Daphne* x *burkwoodii* 'Somerset' were micropropagated on 5 May 1996, weaned by 12 September 1996, potted into 1 litre containers and grown under glass at 20 +/- 5 °C until June 1997. The plants were graded into comparable sizes on 10 July 1997, potted into 2 litre containers, and grown on capillary sandbeds under polyethylene.

Measurements were made of shoot and root development and of pH and EC in the media using methods described in Section 1. Air-filled porosity was measured as before but in the absence of a plant.

Results

Experiment 1: Effects of organic matter content, nutrition, irrigation on overwintering of *Acer palmatum* 'Aureum'

This extended the media x fertiliser rate experiment of the first growing season to examine how these and other factors affected overwintering. Plants were moved onto capillary sandbeds within a polyethylene tunnel on 30 October 1996 and divided between a 'Dry' regime (infrequently watered, i.e. only when the surface of the medium was evidently dry) and a 'Wet' regime (watered three times a week). The experiment then comprised all combinations of the following three factors:

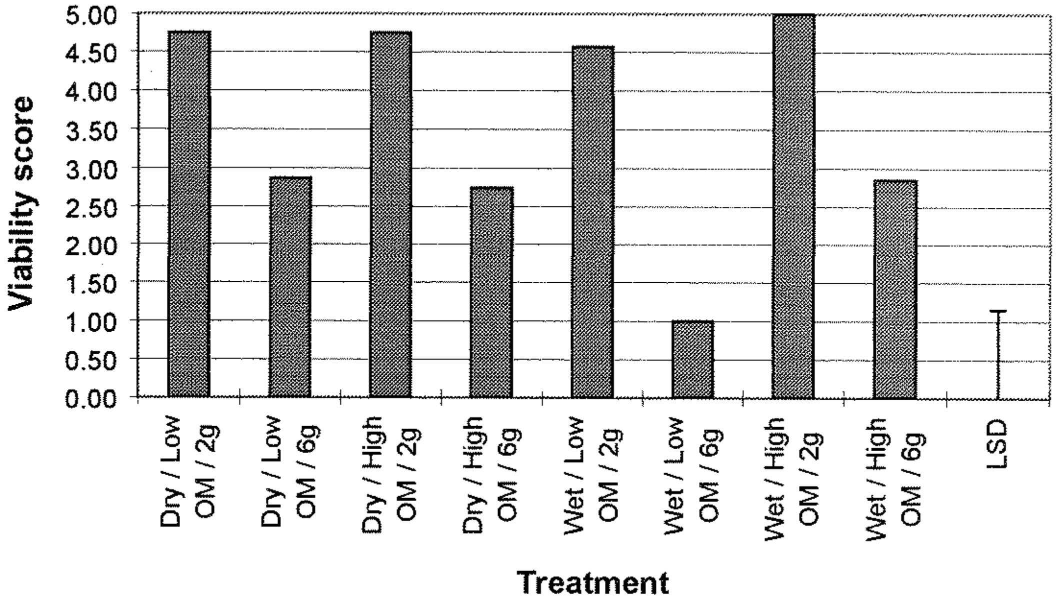
Irrigation Regime:	**Dry** v **Wet**
Organic Matter Content:	**High organic matter** v **Low organic matter** (High O.M. = 70% peat: 30% Cambark 100 v/v; Low O.M. = 90% fine-grade vermiculite: 7 % peat: 3 % Cambark 100 v/v/v)
Fertiliser Rate:	**Control (2 g L^{-1})** v **High (6 g L^{-1})** CRF (Ficote 180, (16%N: 10% P_2O_5: 10% K_2O; w/w/w) plus FTE

There were 8 replicate plants per treatment. Plants were assessed for viability and quality throughout the winter and a final assessment was carried out on 21 April 1997, after budbreak and leaf expansion.

The amount of fertiliser had a significant effect on plant survival and quality during the winter period ($P < 0.001$). In contrast to the beneficial effect of the higher amount of fertiliser during the previous summer, the higher amount of fertiliser was associated with lower survival rates and severe dieback of stem tissues (Figure 37). The greatest fatalities and dieback of growth were associated with plants in the Wet / Low OM / high fertiliser amount treatment. Dieback was characterised by wilting of the young shoots, followed by necrosis of the tips and the formation of dark brown lesions at the base of the main stem.

Roots generally appeared healthy until *after* severe shoot dieback was already apparent, except in some of the heavily watered treatments, which showed blackening of roots at the base of the root-ball typical of waterlogging stress. It was suspected that vascular damage at the neck of the plant might have been the cause of leaf wilting, as pale to dark brown discoloration on the base of the stem often coincided with wilting of the foliage. However, examination of a number of wilted specimens failed to identify any pathogen such as *Verticillium*.

Figure 37. Acer palmatum *'Aureum' - The effects of irrigation, organic matter content and amount of fertiliser on plant viability during winter / spring 1997.*
[Viability score: 0 = Plant dead, 1 = Severe dieback and necrosis, 2 = Widespread dieback, necrosis and leaf wilting, 3 = More than 50% of plant affected by stem dieback or leaf wilting, 4 = Less than 50% of stems with dieback or leaf wilting, 5 = No or limited evidence of dieback or leaf wilting.]



In the vermiculite-based medium, EC values measured at the end of the winter reflected the fertiliser treatments but this was not the case in the peat/bark medium (Table 16). This may reflect the nutrient binding effect of the bark, or may be due to greater nutrient uptake by the plants in the peat/bark. Therefore, it is possible that plant dieback was a consequence of high nutrient levels within the tissues *per se*. Alternatively, the 'soft' growth induced in the previous summer by high nutrition may have been more susceptible to abiotic or biotic stresses over the winter period.

Table 16. Acer palmatum *'Aureum' - The effects of irrigation, organic matter content and amount of fertiliser on pH and electrical conductivity (EC) values recorded in March 1998.*

Watering regime	Organic matter content	Fertiliser rate (g L^{-1})	pH	EC (µS cm^{-1})
Dry	Low	2	6.1	424
Dry	Low	6	5.8	1003
Dry	High	2	4.3	342
Dry	High	6	4.3	348
Wet	Low	2	6.3	464
Wet	Low	6	5.2	1231
Wet	High	2	4.6	360
Wet	High	6	4.5	302
LSD			0.21	253

In this experiment where plants were dormant during the winter period, the influence of the irrigation regime and the proportion of organic matter in the medium were relatively insignificant in determining the survival and quality of *Acer* plants, certainly in comparison to nutrient levels.

Experiment 2 : Organic matter content of the medium

Plants of *Acer palmatum* 'Aureum' and *Daphne x burkwoodii* 'Somerset' were divided between the following 6 treatments:

1.	'Peat'	100% Peat (medium grade)
2.	'Bark'	100% Bark (fine grade)
3.	'Peat/Bark'	50% Peat / 50% Bark (v/v)
4.	'Peat/Sand'	50% Peat / 50% Sand (fine grained) (v/v)
5.	'Ver/Sand'	50% Vermiculite 50% Sand (v/v)
6.	'Sand'	100% Sand

To each mix were added: 4 g L^{-1} Ficote 270 CRF (14%N: 8% P_2O_5: 8% K_2O; w/w/w plus trace elements) and 1.5 g L^{-1} magnesium carbonate. Each treatment was applied to 10 replicate plants of *Daphne* and 5 replicates of *Acer*.

Acer palmatum

Shoot growth was greatest in the media containing high levels of organic matter, and very limited in media containing sand (Figure 38). Root development measured in March 1998, whilst generally poor, was greatest in the peat/bark mix (not significant, Figure 39). At this time, buds were breaking and the severity of shoot tip dieback was scored. Dieback was significantly less in sand and the vermiculite/sand mix than in the more organic media (Table 17).

Figure 38. Acer palmatum *'Aureum'- The effects of organic matter content on total shoot length as measured in July and October 1997.*

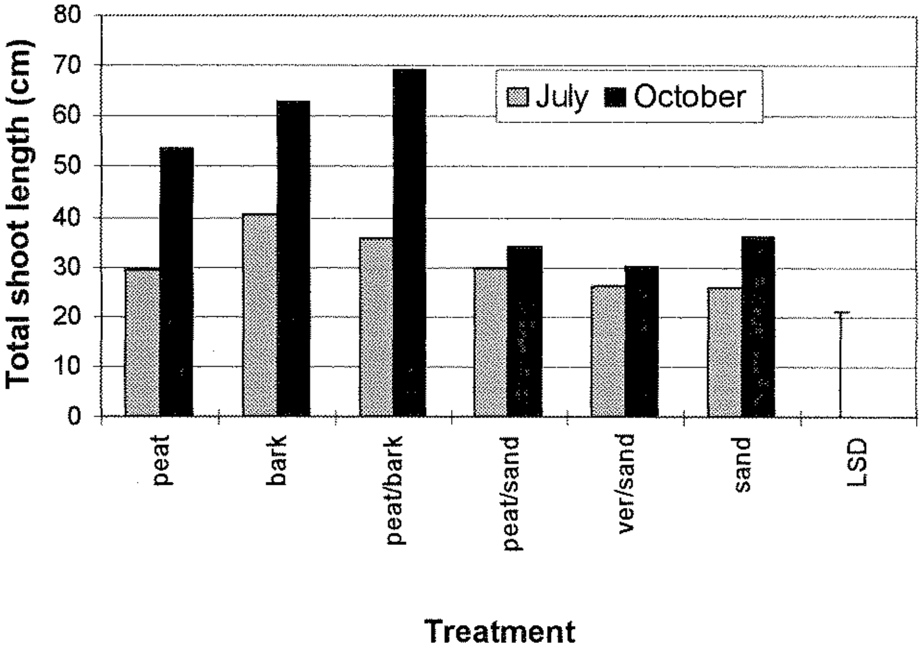
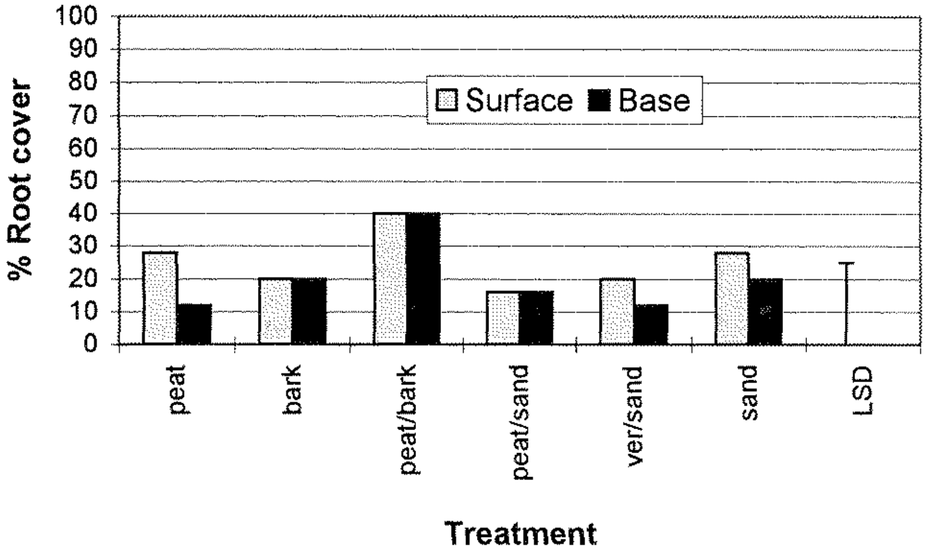


Figure 39. Acer palmatum *'Aureum'- The effects of organic matter content on percentage root cover on the surface and base of the medium.*



Interestingly, the treatments with severe shoot tip dieback were those with relatively high EC and low pH values (means of four sampling times between July 1997 and March 1988, Table 17). The low growth rates associated with the sand treatments could reflect the relatively low nutrient levels indicated by the EC data but were more likely due to poor aeration indicated by the low air filled porosity (Table 17).

Table 17. Acer palmatum *'Aureum'- The effects of organic matter content on shoot dieback, pH, electrical conductivity (EC) and air-filled porosity of media.*

Treatment	Shoot dieback score	pH	EC ($\mu S\ cm^{-1}$)	% Air-filled porosity
Peat	1.0	5.2	522	8.1
Bark	1.0	6.3	385	11.4
Peat/bark	0.8	5.8	566	6.5
Peat/sand	1.4	6.2	459	2.5
Ver/sand	2.2	7.1	303	1.7
Sand	2.6	7.7	188	0.7
LSD	1.19	0.17	75.8	1.23

Shoot dieback score: 0= plant dead, 1=severe dieback, 2=moderate dieback, 3= no dieback visible.

Daphne

Shoot growth was greatest in the bark and peat/bark mixes but, in contrast to *Acer*, growth was very poor in pure peat (Figure 40). When examined in March 1998, root development was good in all the media except pure peat (Figure 41) and was best in pure bark. EC was highest in peat but only marginally higher than in the peat/sand mix, in which there was twice as much visible root (Figure 43). Similarly, the air-filled porosity data do not indicate that poor aeration is likely to have contributed to poor growth in pure peat (Table 17). The only feature that distinguished peat from all other media was its consistently low pH (Figure 42), suggesting that *Daphne* may be particularly susceptible to acid conditions.

Figure 40. Daphne x burkwoodii *'Somerset' - The effects of organic matter content on total shoot length as measured in July and October 1997.*

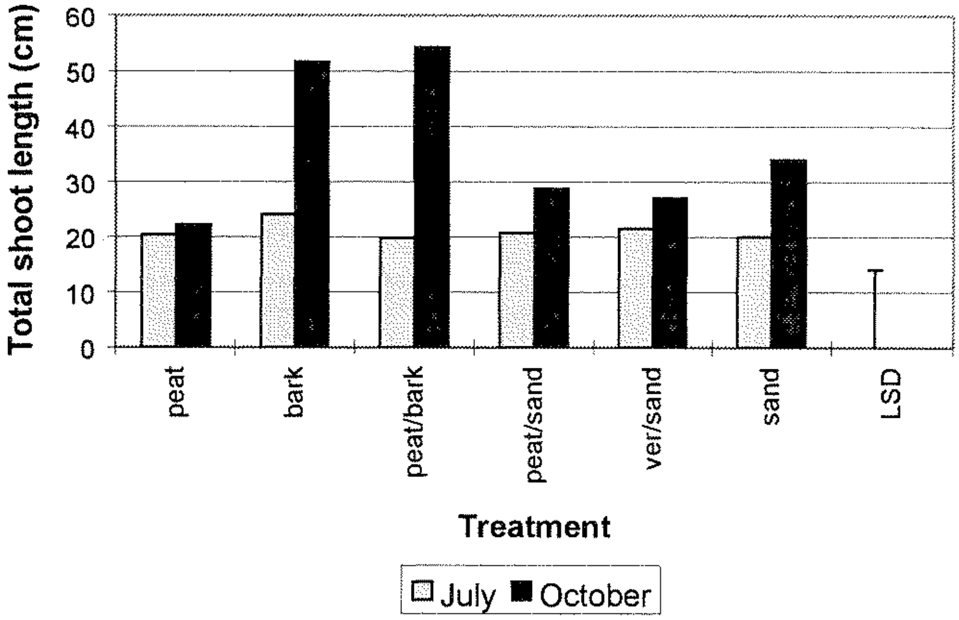


Figure 41. Daphne x burkwoodii *'Somerset' - The effects of organic matter content on percentage root cover on the surface and base of the medium.*

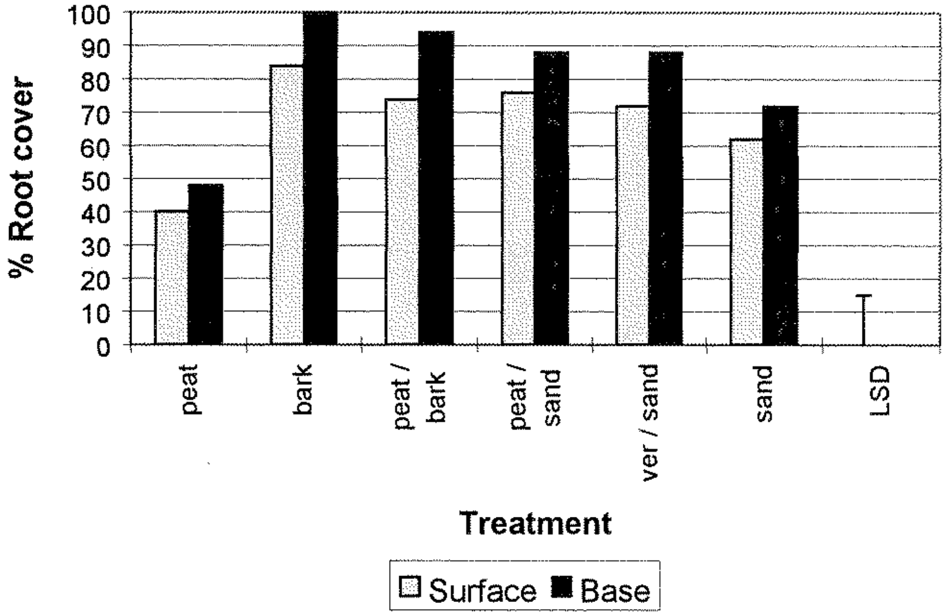


Figure 42. Daphne x burkwoodii *'Somerset' - The effects of organic matter content and sampling date on pH of the media*

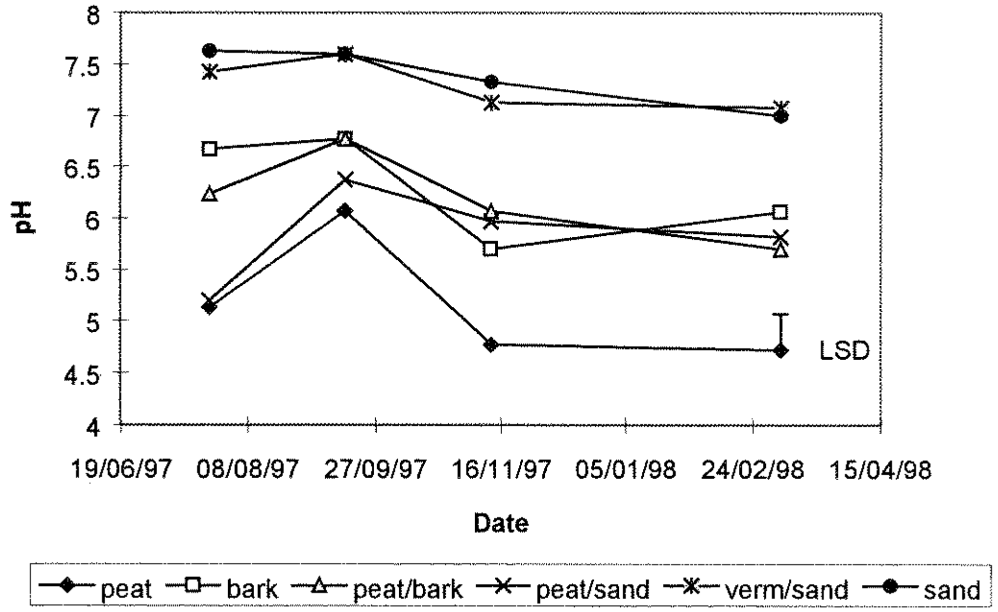
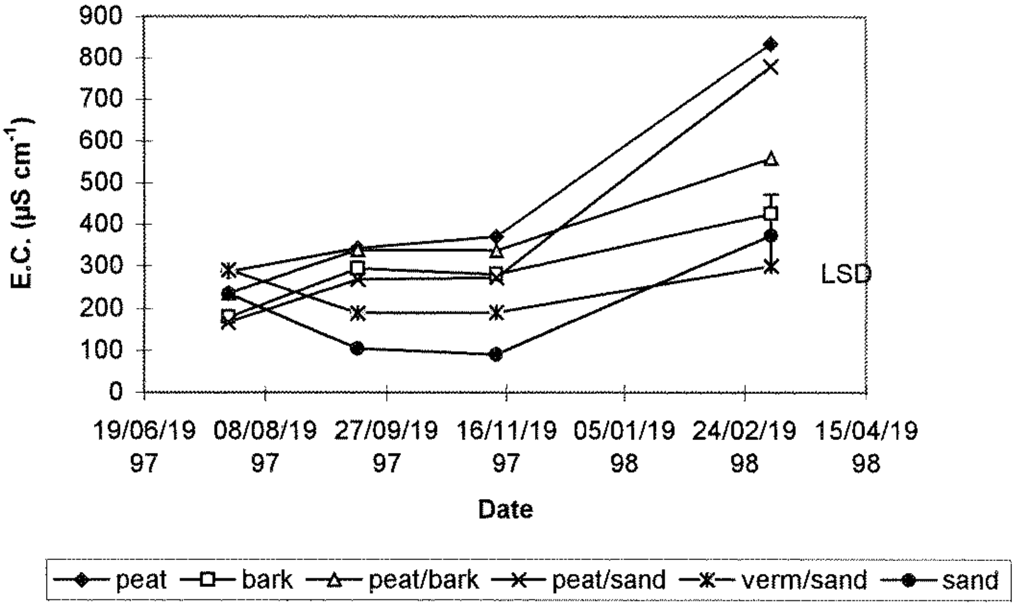


Figure 43. Daphne x burkwoodii *'Somerset' - The effects of organic matter content and sampling date on electrical conductivity (EC) of the media*



Experiment 3 : Medium pH

This experiment examined more directly the effect of medium pH by using lime to adjust the pH of pure peat. There were three treatments:

1. Acid substrate (pH 4-5), pure medium-grade peat
2. Moderately acid to neutral substrate (pH 6-7), peat with 7 g L^{-1} Ca CO_3
3. Moderately alkali substrate (pH 7-8), peat with 400g L^{-1} Ca CO_3

To each substrate Ficote 270, 14:8:8 v/v/v, plus trace elements was added at 4 g L^{-1}. There were 6 replicate plants per treatment for *Daphne* and 3 plants per treatment for *Acer*.

Measured pH levels generally conformed to the intended values apart from a rise of one pH unit in the low pH treatment observed in September 1997, possibly associated with recent overhead watering with mains water at pH 7.8.

Acer

Shoot growth was best at pH 6-7 and this was also the treatment which suffered least from shoot dieback overwinter (Table 18); root growth was satisfactory at low and moderate pH but poor under alkaline conditions (pH 7-8).

Table 18. Acer palmatum *'Aureum' - The effects of media pH on total shoot length, shoot dieback, percentage root cover and electrical conductivity (EC) of the medium.*

PH	Total shoot length (July)	Total shoot length (Oct.)	Shoot dieback score	% Root cover Surface	% Root cover Base	EC (µS cm^{-1})
4-5	42	49	1.3	33	13	502
6-7	44	63	2.3	20	27	485
7-8	44	44	1.0	13	13	562
LSD	27	27	1.04	16	22	132

Shoot dieback score: 0= plant dead, 1=severe dieback, 2=moderate dieback, 3= no dieback visible.

Daphne

Only plants in the medium at pH 6-7 developed any new shoots (Figure 44), whereas good root growth occurred also at pH 7-8 (Figure 45). Plants at low pH also suffered much more leaf loss, as measured by a defoliation score in March 1998 (data not shown).

This experiment therefore confirmed the great importance of correct pH for successful growth of this plant that was suggested by Experiment 2.

Figure 44. Daphne x burkwoodii *'Somerset' - The effects of pH on total shoot length measured in July and October 1997.*

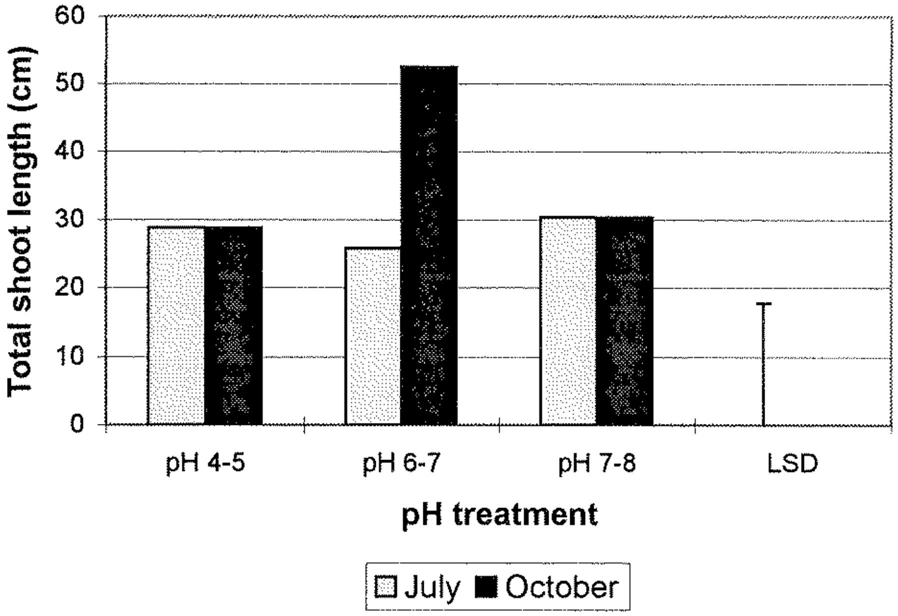
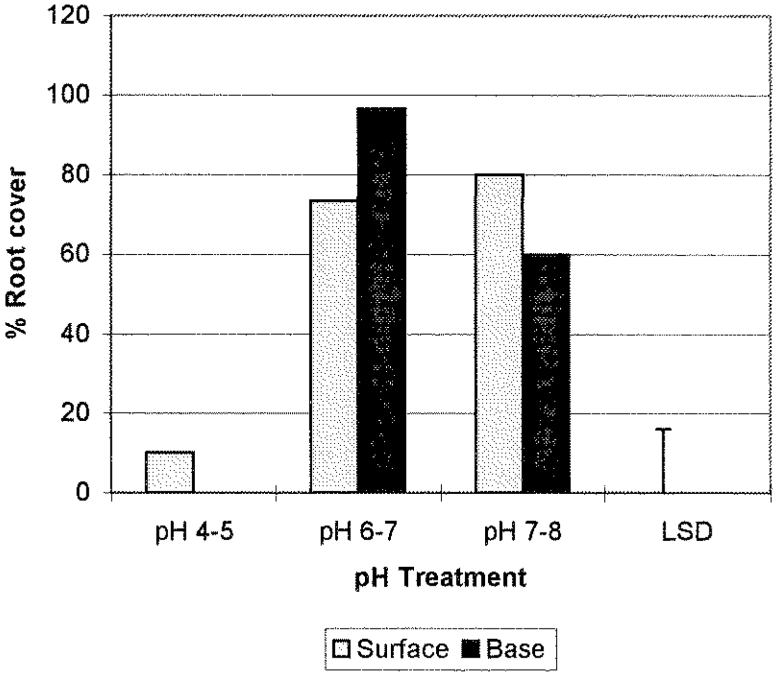


Figure 45. Daphne x burkwoodii *'Somerset' - The effects of pH on percentage root cover on the surface and base of the medium.*



Conclusions

Symptoms related to the loss of quality and viability in *Acer* often include shoot-tip dieback, lesions at the base of stems and leaf wilting (especially at, or shortly after, bud break in the spring). In *Daphne*, poor overall growth, severe wilting and the death of established plants are common complaints.

High levels of nutrients in the medium appear to predispose *Acer* to loss of quality during the dormant phase and as growth restarts in spring, even though the initial effect is to enhance growth. It is difficult to know whether the adverse effect of high levels of salts is direct or indirect. Increased ionic concentrations within stem tissues during winter may directly disrupt cell membranes resulting in cell death. Necrosis of leaf margins and shoot tips is often symptomatic of this form of stress as ions carried in the transpiration stream are concentrated by evaporation in transpiring leaves. Alternatively, high nutritional levels that stimulate active shoot growth late in the summer may suppress lignification and other aspects of 'ripening' of the tissues in preparation for winter. This relatively 'soft' growth is then liable to be more susceptible to abiotic stresses, such as dehydration or cold, and biotic stress such as infection by pathogenic fungi.

In no case was obvious damage of roots in the high fertiliser treatment observed but, as seen in the experiments described in Section A, symptoms of salt stress are rarely distinct except in extreme cases. For the moment, our results suggest that growers would be wise to avoid high levels of fertiliser in the production of *Acer palmatum* cultivars, as short-term benefits may lead to problems in the long term.

No evidence was obtained in support of the idea that the unnaturally high content of organic constituents in modern growing media is responsible for the loss of quality that often afflicts *Acer palmatum* cultivars; indeed the best growth was observed in a mixture of peat and bark. This subject was tolerant of a wide range of pH but 6-7 appeared to be optimal.

For *Daphne*, the most critical factor identified was the pH of the medium. Growing plants at a pH of 6-7 resulted in good growth of shoots and roots. Too low a pH (pH 4-5) inhibited root growth considerably and completely prevented any new shoot growth. At high pH (7-8) roots grew reasonably well but shoots shows lime-induced chlorosis and growth was stunted. These results with *D.* x *burkwoodii* 'Somerset' run counter to the view that *Daphne* is generally tolerant of a wide range of pH (Brickell and Mathew, 1981).

As with *Acer*, there was no evidence that a high proportion of organic matter in the medium had an adverse effect on *Daphne*, as long as steps were taken to adjust the pH to be in the optimum range.

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GLOSSARY : terms, abbreviations and products used

CRF - controlled release fertiliser.

EC - electrical conductivity of a solution. It is a function of the concentration of dissolved ions and as such a simple measure of the concentration of salts in solution. Ficote 140 16-10-10 (and similar)- controlled release fertilisers supplied by Fisons Horticulture Division, Bramford, Ipswich. '140' relates to the expected release period of the product. '16-10-10' indicates the percentage by weight of the major nutrients in the order N - P - K. The value for N is in terms of pure nitrogen but, by convention, the values for P and K relate to the equivalent weight of their oxides $P2O5$ and $K2O$.

FTE - Fritted trace elements. The product used was WM255 which is sintered silicate granules, supplied by Munro South Horticulture, Maidstone, containing the following elements (with minimum percentages, by weight, in brackets): Fe (18.8%), Mn (5.4%), Zn (4.3%), Cu (4.3%), Mo (1.0%), B (1.0%), K (1.2%).

LSD - Least Significant Difference, i.e. the smallest difference between two treatment means that is statistically significant, i.e. there is less than a 5% chance that it occurred by chance rather than because of a real difference between the treatments.

morphology - information about the form of an object. In the context of this report it refers to all aspects of the external appearance of plant shoots and leaves.

necrosis - tissue death. Used in this report to refer to darkening of tissues that indicates that the cells have probably died so that the tissues will eventually break down.

NS - not statistically significant, i.e. the probability that the observed difference(s) were due to chance is greater than 5%.

($P < 0.05$, $P < 0.01$, or $P < 0.001$) - a statement of the statistical probability (P) that the observed differences could have been due to chance. The smaller the value of P, the more certain we can be that the result is 'real'. A value of 0.05 is conventionally taken as the threshold for accepting the result, i.e. that an effect is 'statistically significant'.

pH - a measure of the acidity of a solution. It is actually the negative log of the hydrogen ion concentration and on this scale 7 is neutral; values below 7 are acid and values above 7 are alkaline.

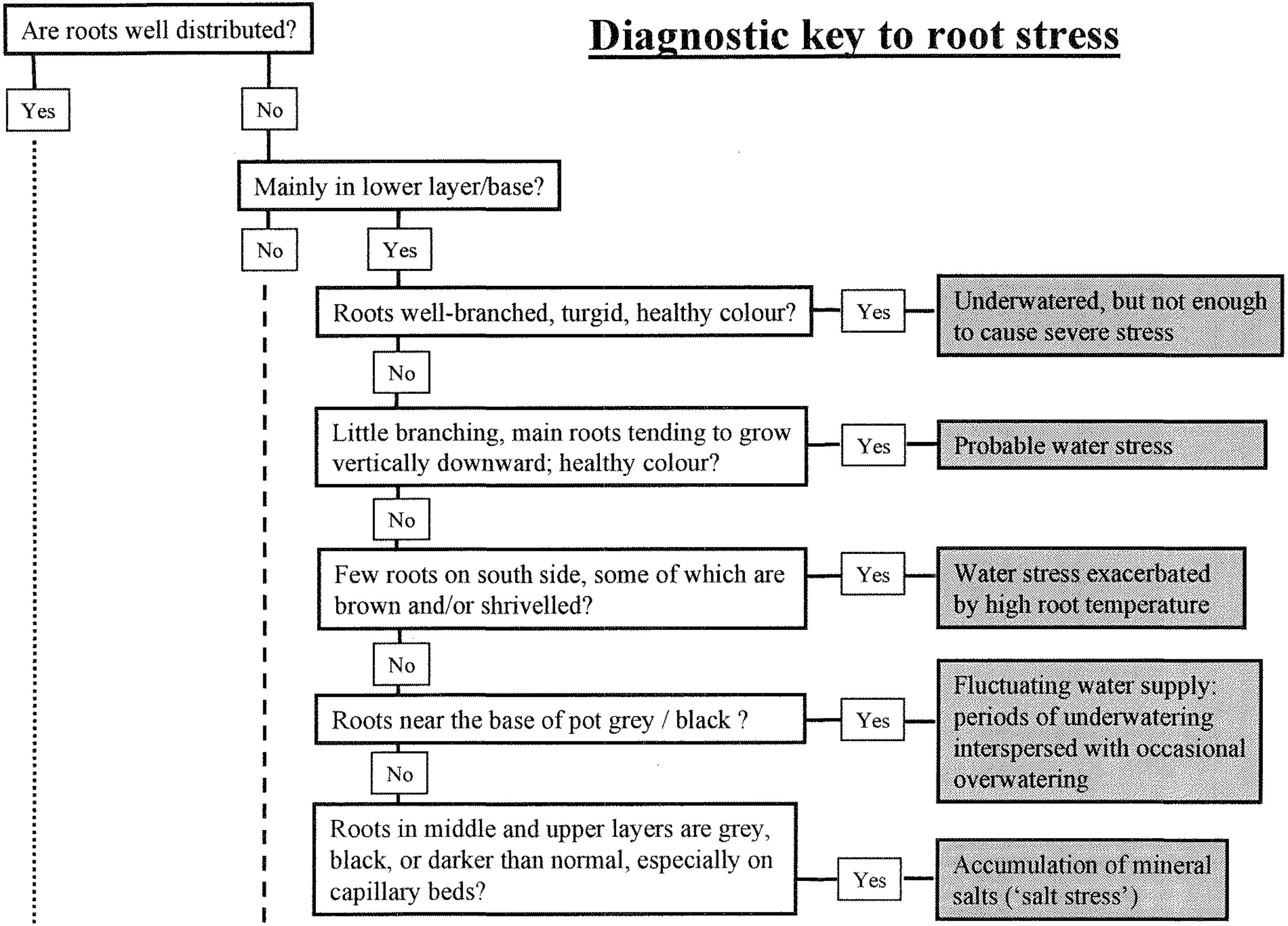
PPFD - Photosynthetic Photon Flux Density. A measure of irradiance confined to the wavelengths of light that are active in photosynthesis (i.e. 400 to 700 nm) and in the units that relate to its action in photosynthesis (i.e. quantum units).

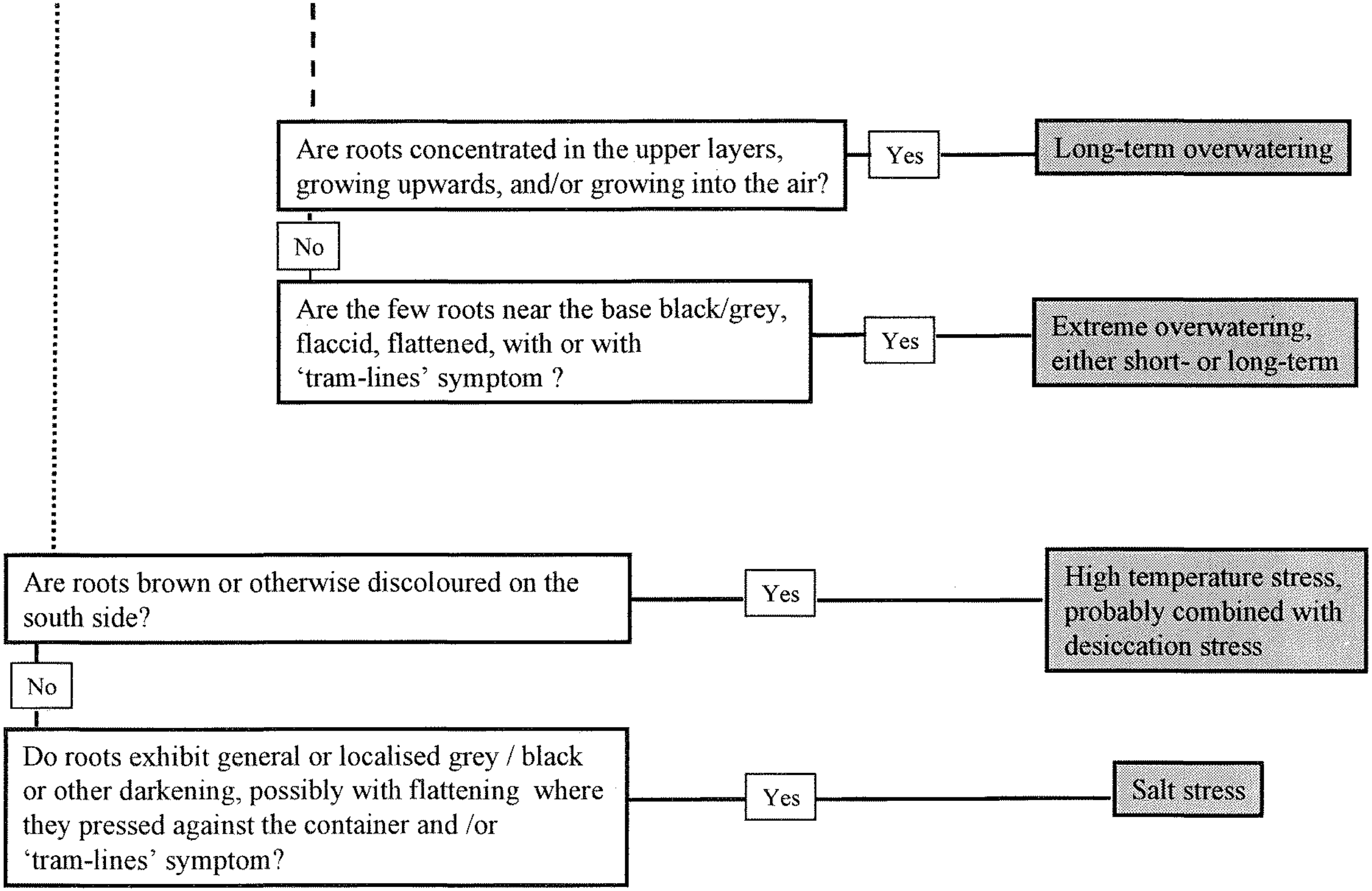
stomatal conductance - A measure of the ease with which water vapour can diffuse out of the lower surface of a leaf. This depends largely on how wide open are the stomata (i.e. stomatal aperture) and the number of stomata per unit area of leaf (i.e. stomatal density).

APPENDIX 1.

A simple key for the preliminary diagnosis of stress factors from the appearance of roots as seen on the surface of the root-ball after removal from of a container

Diagnostic key to root stress




Are roots concentrated in the upper layers, growing upwards, and/or growing into the air?
Yes
Long-term overwatering
No
Are the few roots near the base black/grey, flaccid, flattened, with or with 'tram-lines' symptom ?
Yes
Extreme overwatering, either short- or long-term
Are roots brown or otherwise discoloured on the south side?
Yes
High temperature stress, probably combined with desiccation stress
No
Do roots exhibit general or localised grey / black or other darkening, possibly with flattening where they pressed against the container and /or 'tram-lines' symptom?
Yes
Salt stress

APPENDIX 2 : Photographs

Plate 1. Root system of an *Elaeagnus pungens* 'Maculata' plant grown for ten weeks under the 'overhead-dry' treatment.

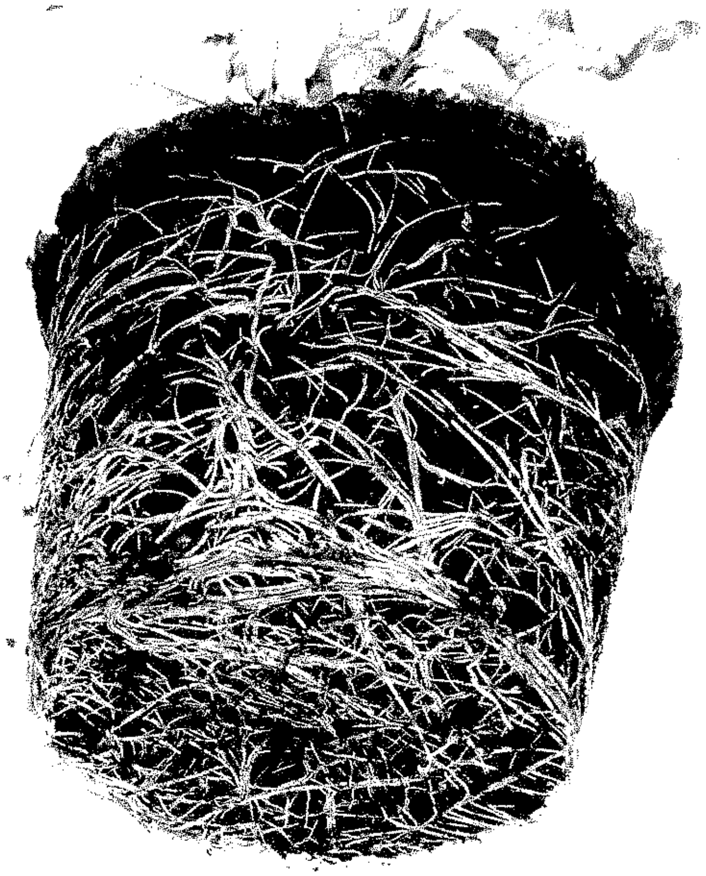


Plate 2. Root system of an *Elaeagnus pungens* 'Maculata' plant grown for ten weeks under the 'overhead-control' treatment.

Plate 3. Root system of an *Elaeagnus pungens* ‘Maculata’ plant grown for ten weeks under the ‘overhead-wet’ treatment.

Plate 4. Root system of an *Elaeagnus pungens* 'Maculata' plant grown for ten weeks under the 'capillary-control' treatment.

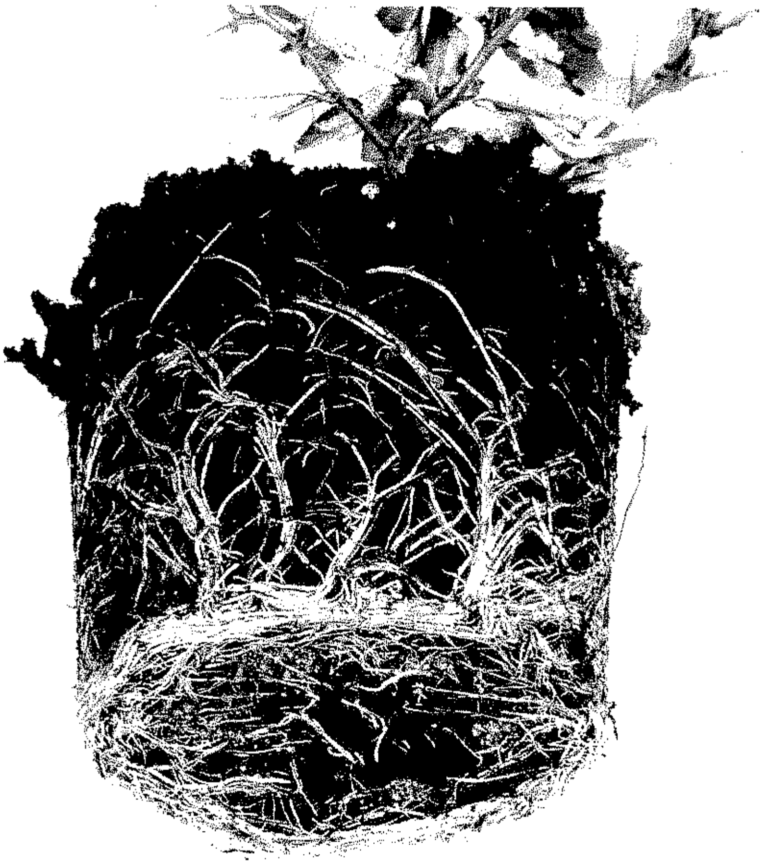


Plate 5. Root system of an *Elaeagnus pungens* 'Maculata' plant grown for ten weeks under the 'capillary-salt' treatment.

Plate 6. Root system of a *Berberis stenophylla* plant grown for ten weeks under the ‘overhead-dry’ treatment.

Plate 7. Root system of a *Berberis stenophylla* plant grown for ten weeks under the ‘overhead-control’ treatment.

Plate 8. Root system of a *Berberis stenophylla* plant grown for ten weeks under the 'overhead-wet' treatment.

Plate 9. Root penetration of the top surface of a pot containing a *Cornus alba* 'Elegantissima' plant grown for ten weeks under the 'overhead-wet' treatment.

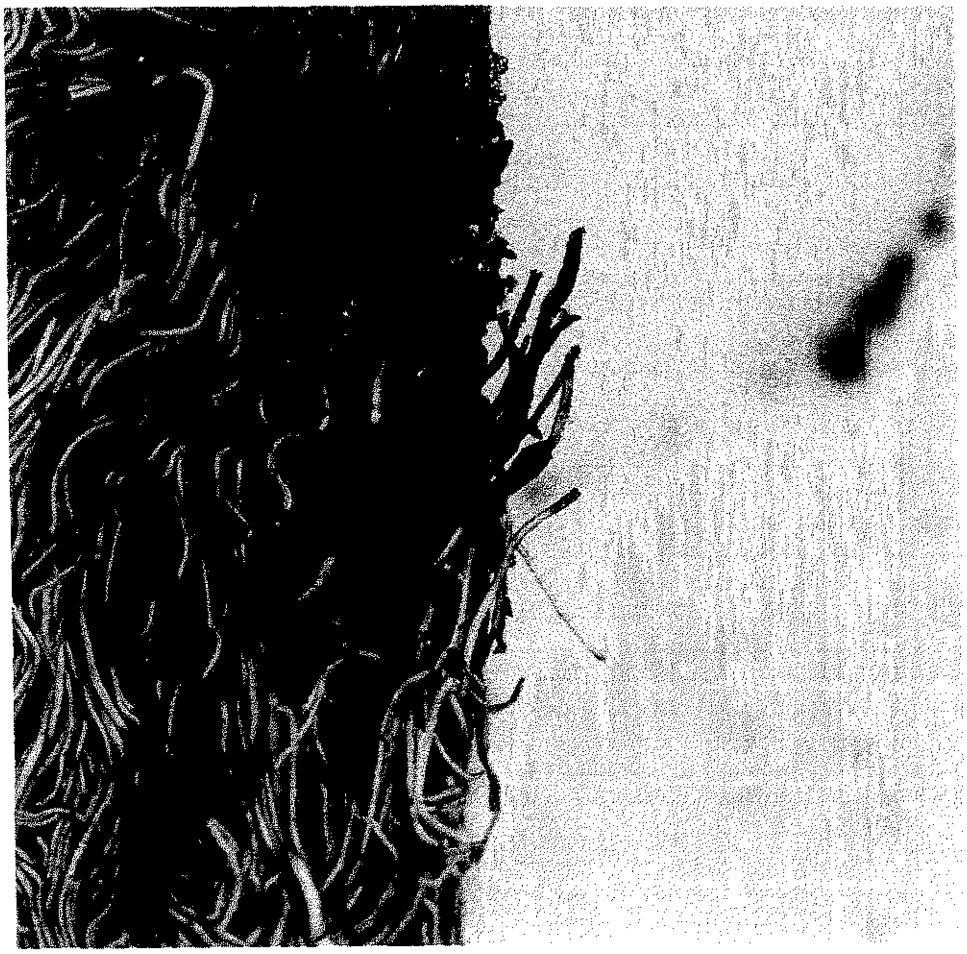


Plate 10. Root blackening on the bottom edge of the rootball (lower zone) of a *Cornus alba* 'Elegantissima' plant grown for ten weeks under the 'capillary-salt' treatment.

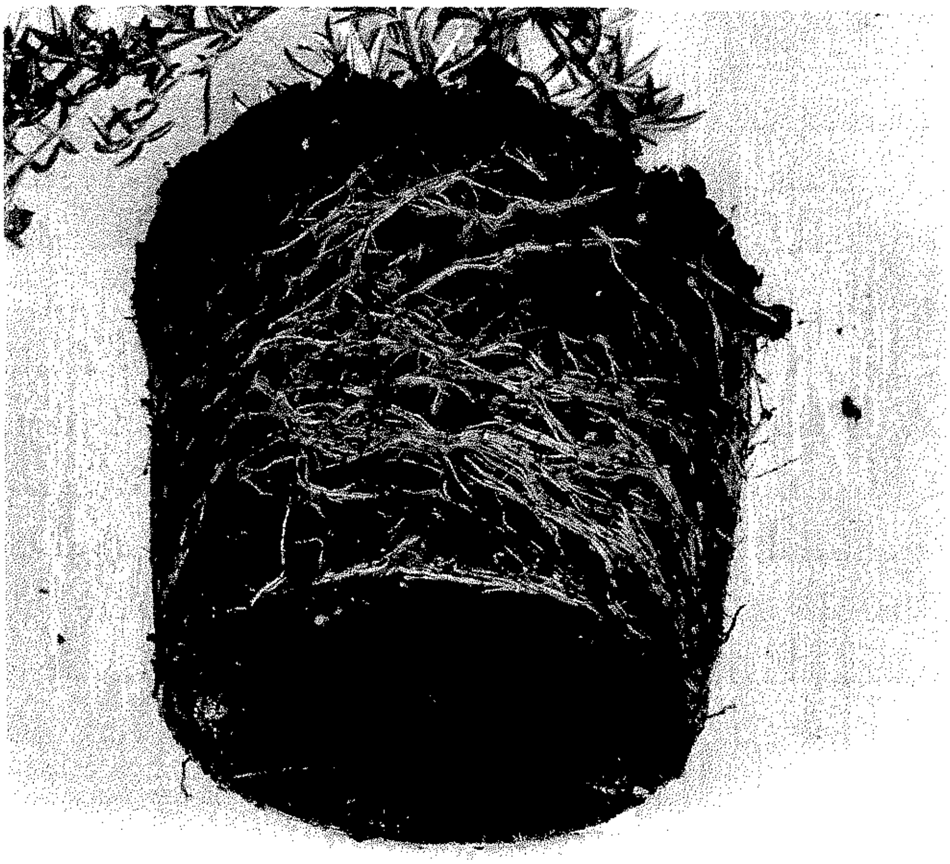


Plate 11. Root system of a *Berberis stenophylla* plant grown for ten weeks under the 'capillary-control' treatment.

Plate 12. Root system of a *Berberis stenophylla* plant grown for ten weeks under the 'capillary-salt' treatment.

Plate 13. Root system around the base of a *Garrya elliptica* 'James Roof' plant grown for ten weeks under the 'capillary-salt' treatment.

Plate 14. Root system around the lower zone of a *Garrya elliptica* 'James Roof' plant grown for ten weeks under the 'overhead-wet' treatment.

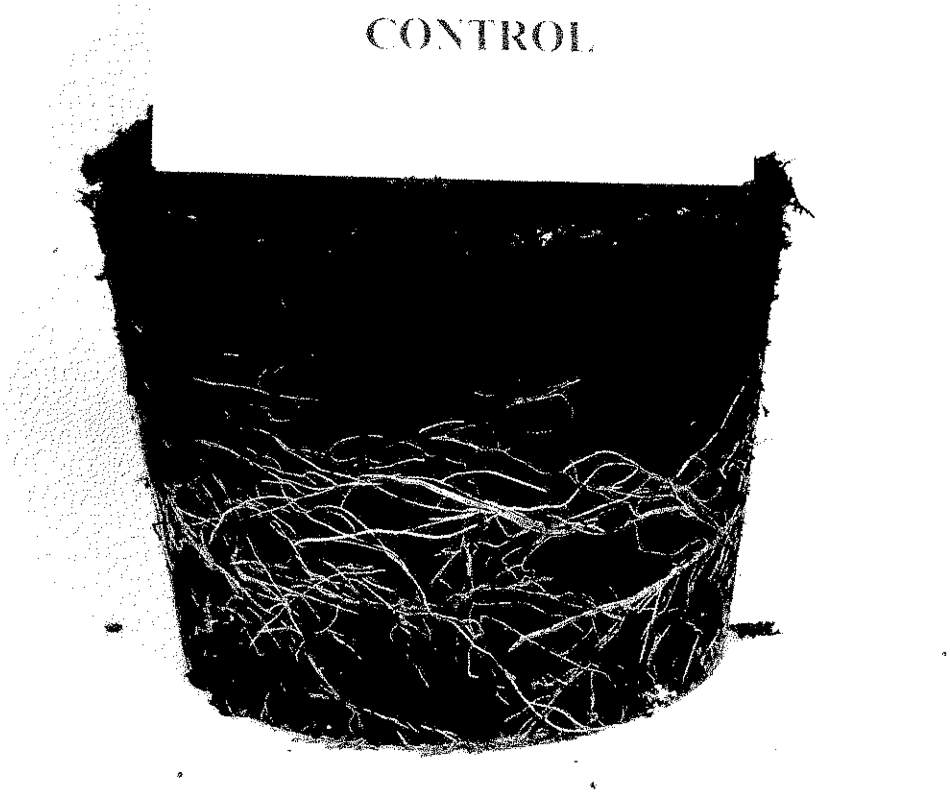


Plate 15. Typical root system of a *Daphne* x *burkwoodii* 'Somerset' plant.

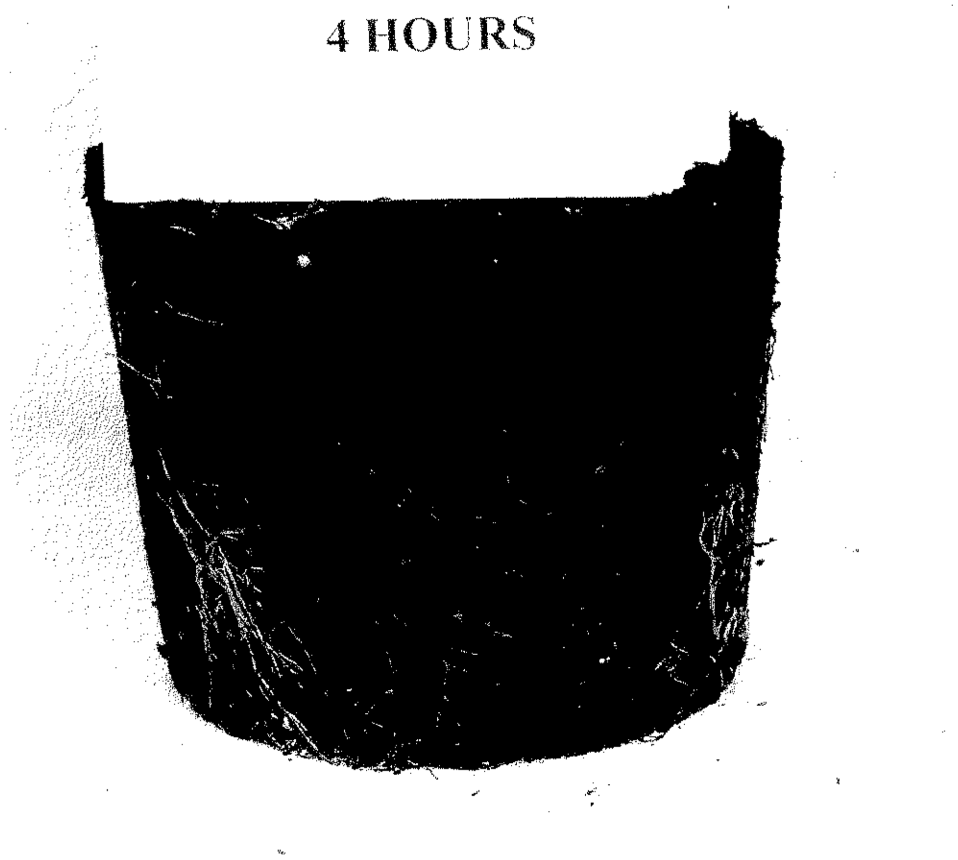


Plate 16. Root system of a *Daphne* x *burkwoodii* 'Somerset' plant exposed to a four hour high-temperature treatment.

Plate 17. Detailed view of root discolouration of a *Daphne* x *burkwoodii* 'Somerset' plant exposed to a four hour high-temperature treatment.

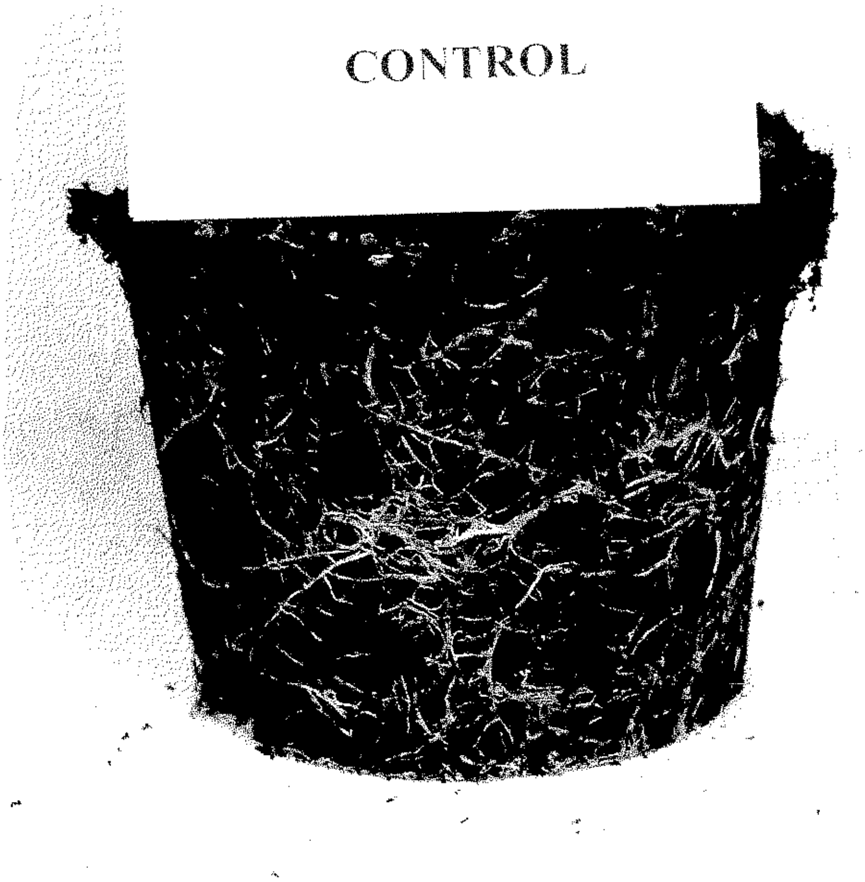


Plate 18. Typical root system of a *Garrya elliptica* 'James Roof' plant.

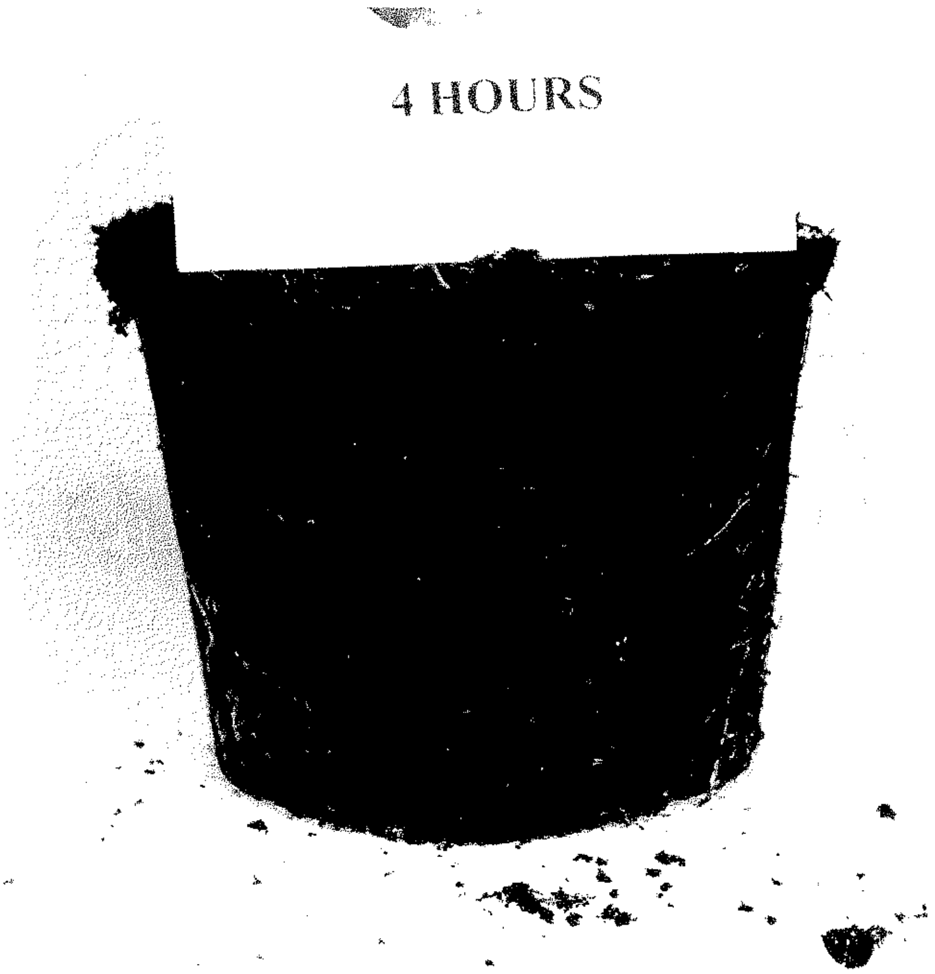


Plate 19. Root system of a *Garrya elliptica* 'James Roof' plant exposed to a four hour high-temperature treatment.

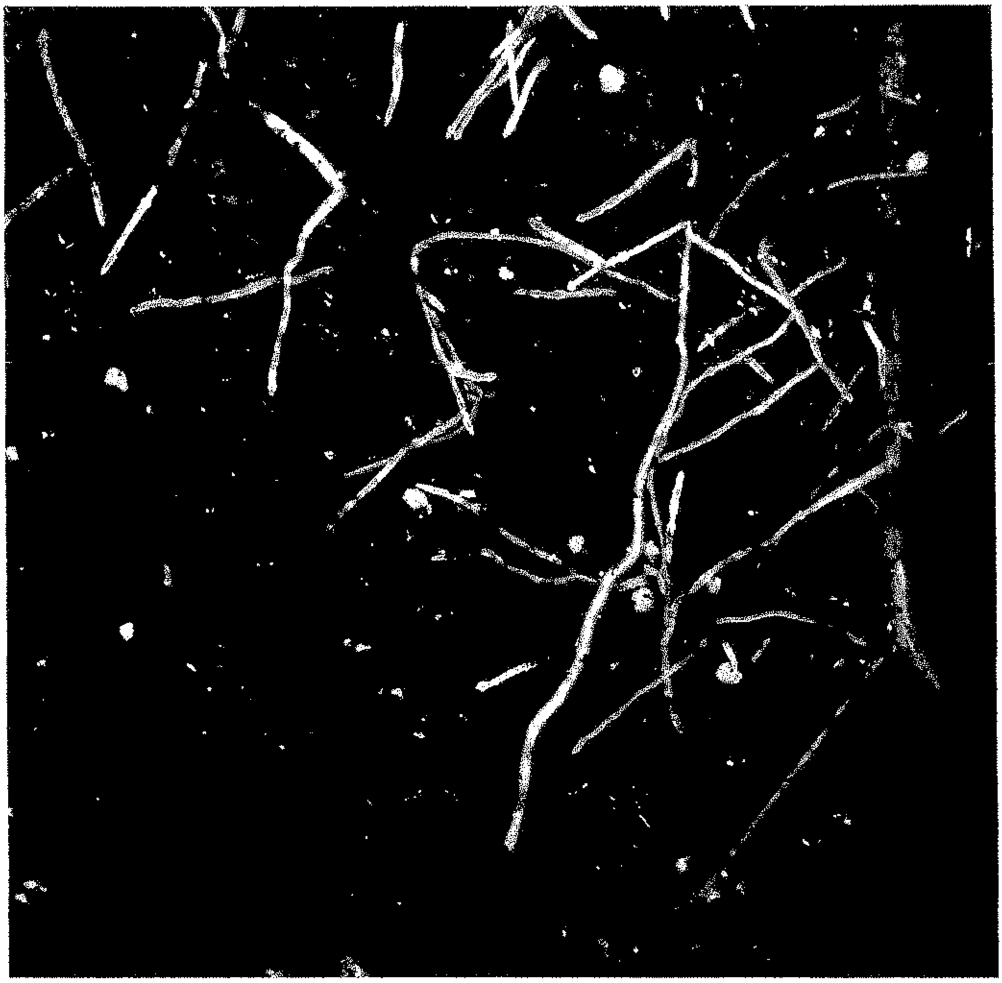


Plate 20. Close-up of roots in the 36 g L^{-1} CRF treatment layer in a root box, photographed after the glass panels had been removed at the end of the experiment, showing thick main roots and many fine lateral roots which have grown through groups of CRF granules without suffering any visible injury from 'salt stress'.

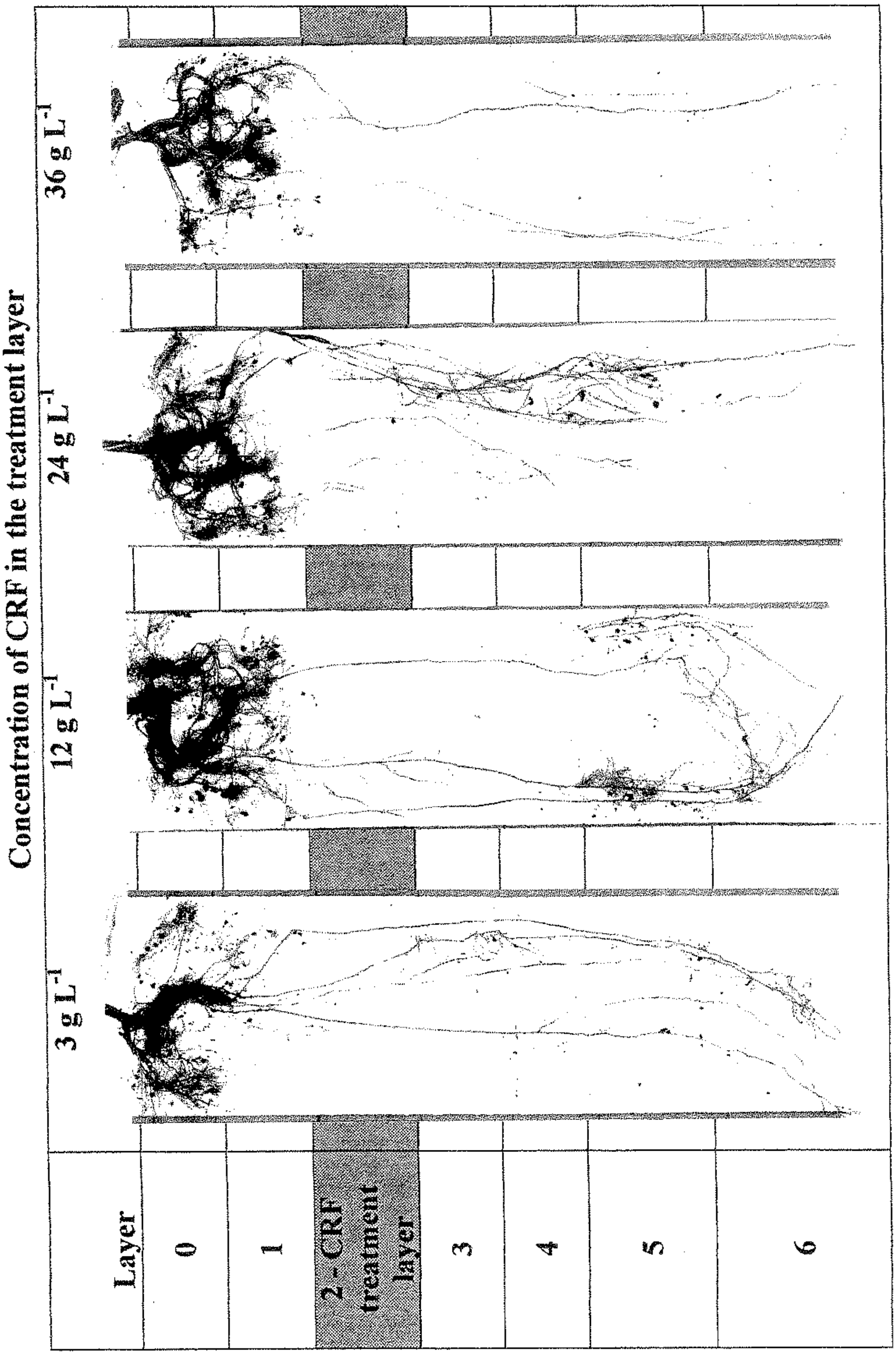


Plate 21. Photographs of the root systems of *Elaeagnus pungens* 'Maculata' extracted from the root boxes. The concentration of CRF was 3 g L^{-1} in all the unshaded layers but varied in the shaded layer from 3 to 36 g L^{-1}.

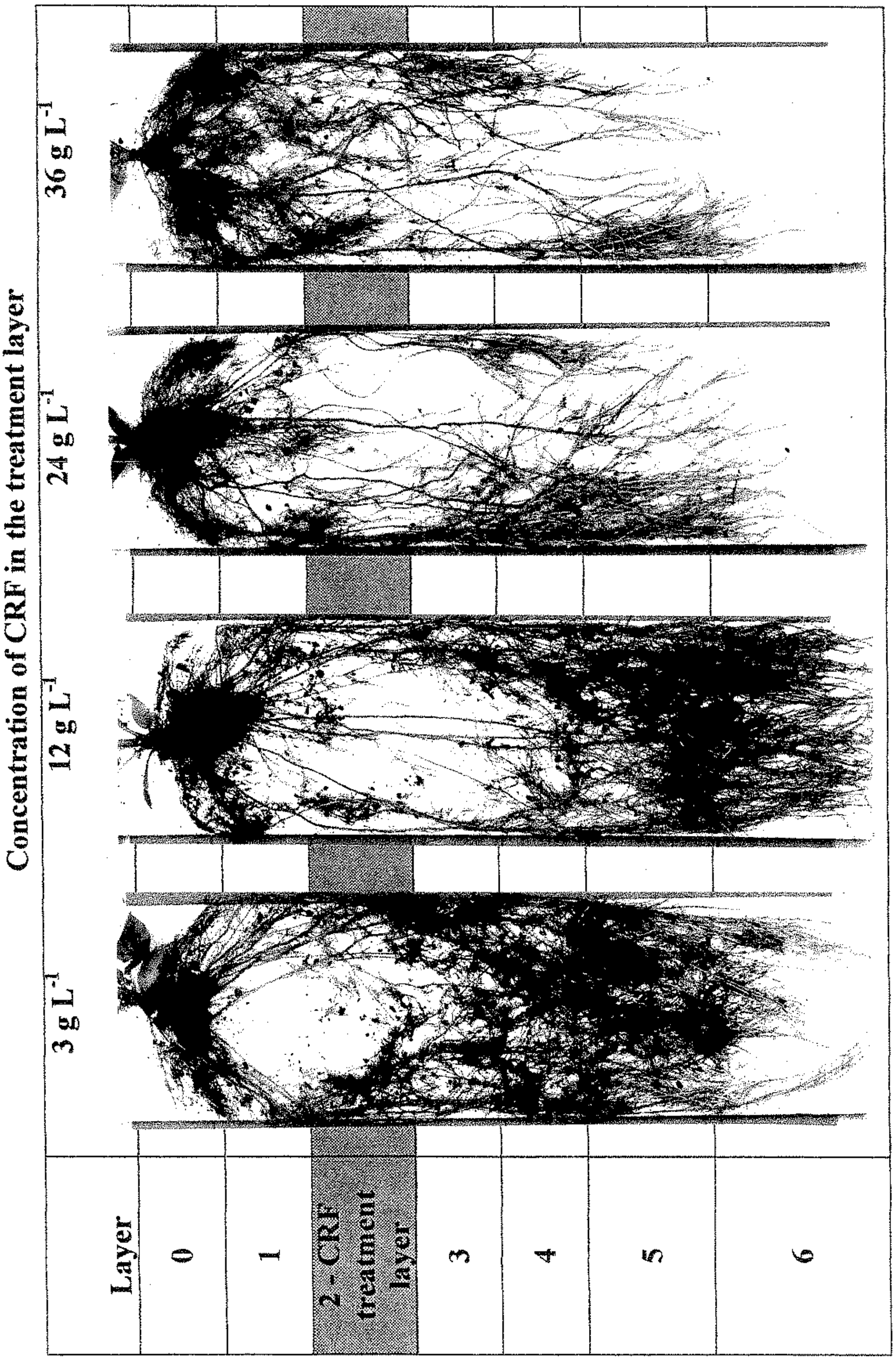


Plate 22. Photographs of the root systems of *Viburnum tinus* 'Eve Price' extracted from the root boxes. The concentration of CRF was 3 g L^{-1} in all the unshaded layers but varied in the shaded layer from 3 to 36 g L^{-1}.

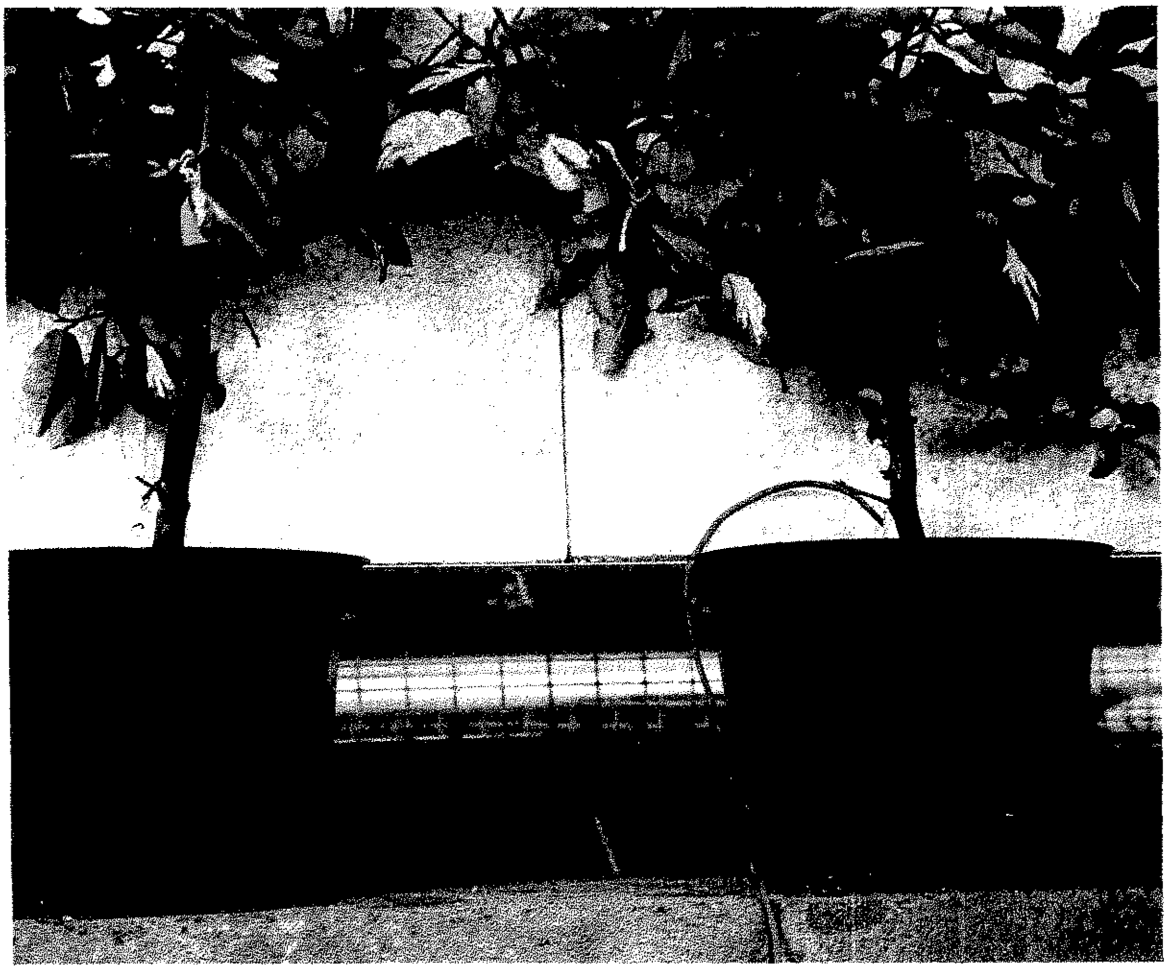


Plate 23. Plants of *Elaeagnus pungens* 'Maculata' in the apparatus used to simulate the most extreme heat stress on roots in containers that could occur under outdoor conditions in the UK.

Plate 24. Roots of *Elaeagnus pungens* 'Maculata' after three severe heat stress cycles in which roots on the side of container facing the heat source were exposed to temperatures of 45 – 55 °C using the apparatus shown in Plate 23. The upper photograph is a side view with the heated side on the right. The lower photograph is a face view of the heated side.

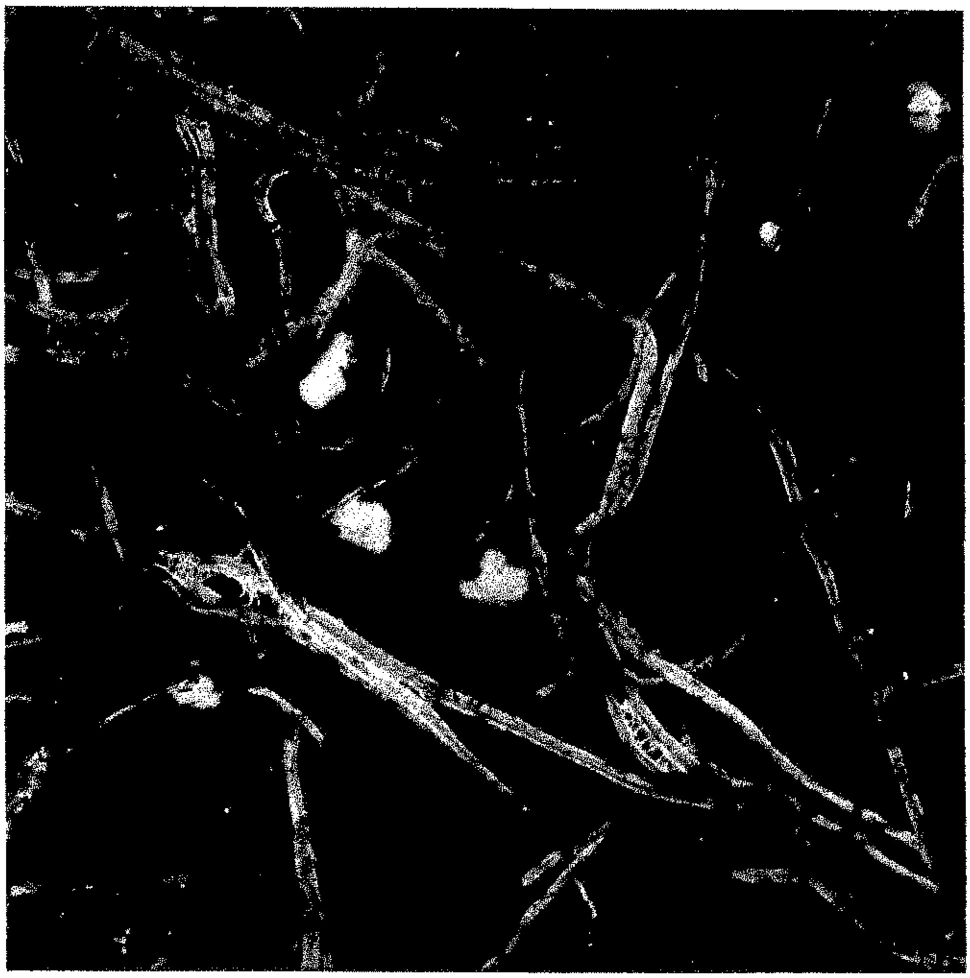


Plate 25. Roots of *Garrya elliptica* 'James Roof' showing the 'tram lines' symptom in response to salt stress (15 g L^{-1} Ficote 180 [16:10:10] in the medium). The distinctive striated appearance seemed to be due to darkening of the central tissues of the root (i.e. the stele) relative to the outer tissues (i.e. principally the cortex). The symptom was often associated with roots becoming flattened against the wall of the container.