CONTROLLING WATER USE OF TREES TO ALLEVIATE SUBSIDENCE RISK

Horticulture LINK project 212

Final report – May 2004

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The results and conclusions in this report are based on a series of experiments conducted over a five-year period. The conditions under which the experiments were carried out and the results have been reported with detail and accuracy. However, because of the biological nature of the work, it must be borne in mind that different circumstances and conditions could produce different results. Therefore, care must be taken with the interpretation of the results, especially if they are used as the basis for commercial recommendations.

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INDEX

EXECUTIVE SUMMARY

Background

Buildings and the environment in which we live are key elements of our quality of life. Whether trees grow within a private garden or in a roadside pavement, the general public by and large appreciates them. Justifiably, because trees contribute so much to our environment, as well as their visual contribution to our landscape. But trees and buildings in close proximity can lead to problems, whether restricting light or causing damage by root activity. The planting and management of trees close to buildings therefore needs to be planned and undertaken from a sound science base. In a similar way, the planning process (under the auspices of the Office of the Deputy Prime Minister) needs to take account of the potential for conflict to occur and employ innovative design and careful construction. All this will help to avoid such conflict occurring and ensure that trees are maintained and managed in a sustainable manner.

The Highways Agency (HA) has an extensive roadside estate that extends to 30,000 hectares, which supports more than 25 million trees ranging from London plane to wild service trees. The oldest of these are veteran trees and there is a significant population of trees of about 40 years of age. The estate includes an urban element, although much reduced since the transfer of London's trunk roads to Transport for London. Similarly, there are even more trees on non-highway land, including those on private and public property. Many trees are situated close to structures, buildings, road pavements and footways and controlling soil drying by trees is an important potential management tool in these situations.

Trees are an integral and critical part of urban landscapes that provide important aesthetic and environmental contributions that make towns and cities more pleasant, safer and healthier to live in. Trees can give shelter from noise and wind, reduce chemical and particulate air pollution, provide shade and can add value to nearby properties. Trees also benefit urban ecosystems, by sustaining biodiversity. In addition, they reduce storm water run-off and prevent erosion. Removal of city trees will lead to a decline in the quality of urban landscapes and large-scale felling programmes would not be acceptable to the public at large.

Unfortunately, structural damage is associated frequently with the close proximity of trees to low-rise buildings. Trees can extract water from below the foundations causing some particular clay subsoils to shrink, ultimately leading to failure of the foundations and cracks in the superstructure. The cost of repairing the damage caused by the failure of domestic house foundations, due to subsidence, was during the years preceding this project of the order of £300-£400 million annually. Not all of this can be attributed to the presence of tree roots. However, most of the subsidence incidents in the UK are found to occur in areas with clay soils and in these areas, tree roots are claimed to have an effect on subsidence incidents in 73% of cases (Loss Prevention Council, 1995). Hence the potential for saving on remedial costs, by reducing the need for rectification work, may be around £200 million per annum. Currently, no methods exist that reliably predict which trees may cause damage and not all trees near buildings are implicated. Decreasing water uptake by trees may lessen subsidence risk by conserving soil moisture and reducing clay subsoil shrinkage. Reducing canopy leaf area by pruning may lessen water uptake and cyclical pruning is recommended in a risk limitation strategy for tree root claims developed by the London Tree Officers Association (1995). Tree pruning is perceived as a potentially effective control measure to conserve soil moisture that could prevent excessive removal of trees, but the hypothesis has never been tested on amenity trees in the urban environment.

The aim of this project was to improve the understanding of how isolated amenity trees use water, and to determine whether reduction in canopy leaf area and root-restriction are sustainable ways to control growth and reduce water uptake from soil.

Two different standard arboricultural pruning techniques were used to reduce the crown size of mature trees. The first, 'crown-reduction', reduces crown volume but allows its natural shape to be preserved. This involves an overall reduction of both height and spread by removing the outer portions of all major branches. The second, 'crown-thinning', reduces the number of side lateral branches coming off all of the major branches not affecting the original volume of the crown. In both cases the normal industry standard is to aim to reduce the canopy leaf area by 30% (BS3998: 1989, Lonsdale, 1999). The growth of newly planted amenity trees was controlled by restricting their roots within water permeable geotextile lined pits.

Methods that allowed the measurement of total canopy re-growth following pruning, whole tree transpiration and linking this to prevailing climate conditions and impacts on soil drying were successfully developed and used. Thus, canopy leaf area was measured using a modified hemispherical photographic technique adapted for single trees and shoot extension and individual leaf size were measured directly. Stable carbon and oxygen isotope discriminations were used to provide seasonal estimates of leaf water use and porometry was used to give instantaneous values. An artificial leaf was developed to determine how evaporative demand (factors driving water loss) was modified within the canopy. Whole tree transpiration was determined by quantifying sap flow. Impacts on soil drying by tree roots were measured using a neutron probe.

Mature wild cherry (8 m height) and London plane trees (20 m height) were used as model species for determining the effects of pruning and newly planted Norway maple and lime trees were used to determine the effects of root restriction. Six major experiments were carried out over a period of five years.

Summary of practical implications

- Recommendations on pruning now can be scientifically rather than empirically based as previously.

- Trees recovered their canopy leaf areas to pre-pruning amounts very quickly (1-3 years) following crown-reduction to normal industry standards.

- The re-growth after crown-reduction produced trees with greater leaf area density (m^2 leaf/m^3) because they had larger leaves more closely packed together within a smaller crown volume compared to non-pruned trees.

- Crown-thinning reduced the leaf area density, and generally the trees took longer to recover their canopy leaf area than for crown-reduction.

- Total tree water use (transpiration) was reduced by crown-reduction and unaffected by crown-thinning in the year of pruning.

- Crown-reduction reduced soil drying by trees in the year of pruning, but the effects were generally small and disappeared within the following season, unless the reduction was severe, in which case the effects were larger and persisted for up to two years.

- Crown-thinning did not reduce soil drying.

- For newly planted amenity trees, root restriction was found to be a very effective method to control growth.

Summary of science outputs

- The modified hemispherical photographic image technique adapted for single trees use was shown to provide measurements of total canopy leaf area closely related to those determined using an empirical allometric method based on summation of individual branches.

- Crown-reduction increased subsequent shoot extension on cut branches for a minimum of two years. More new (epicormic) vegetative buds were produced particularly at the cut branch ends. The new shoots grew more rapidly and produced larger leaves with higher nitrogen concentrations and greater abilities to assimilate carbon dioxide than non-pruned or crown-thinned trees.

- Measurements of water loss and carbon isotope discrimination of individual leaves from crown-reduced trees indicated that they had a greater capacity to lose water than those from crown-thinned and non-pruned trees, but were less able to respond to evaporative demand outside of the canopy.

- Differences in soil drying caused by crown-reduction and crown-thinning were related to the changes in total leaf area, leaf size and leaf area density. The boundary layer conductance within the canopies of crown-reduced trees was less than for the non-pruned or crown-thinned trees. This counteracted the intrinsic potential of leaves in crown-reduced trees to lose more water.

- Use of artificial leaves confirmed that environmental conditions within the canopy of crown-reduced trees were less conducive to water loss to the environment outside the canopy.

Recommendations

- For practical soil moisture conservation, severe crown-reduction 70-90% of crown volume would have to be applied. Reduction of up to 50% crown volume is not consistently effective for decreasing soil drying.

- To ensure a continued decrease in canopy leaf area and maximise the period of soil moisture conservation, crown reductions should be repeated on a regular managed cycle with an interval based on monitoring re-growth.

- Crown-thinning is not an effective method to control soil drying by trees.

- More information is needed on the effect of repeated pruning to determine the impact on the root system and soil moisture conservation at the periphery of the root system.

- Root restriction within geotextile membrane lined pits may be used as an effective method for controlling shoot growth, but more knowledge is needed on the long-term integrity of the membrane, the stability and the performance of the tree.

Tree management implications

If severe crown reduction is required to alleviate subsidence risk, those trees which pose a potential risk must be identified so they can be treated. There are approximately 100 million trees in the urban environment. Of these, a large, but undefined, proportion is in sufficient proximity to a building to pose a perceived risk of damage. However, even in a drought year, the number of actual cases of subsidence is only about 50,000. The risk of a tree causing subsidence damage which is related to species, foundation depth and soil type may therefore be less than 1%. If one could identify this 1% with any reasonable accuracy, they could be pruned accordingly. However,

attempts in the past to develop methods of subsidence risk assessment have not been successful. The Arboricultural Association method of "Subsidence Risk Assessment" was withdrawn, as it was considered to be ineffective. Royal & Sun Alliance's recent efforts to develop a statistically based model TreeRAT (Tree Risk Assessment Tool) also have not been taken beyond an initial prototype stage.

If trees that pose a risk cannot be identified, then one alternative is to treat all trees, regardless of the risk they pose. The environmental consequences of this would be catastrophic; nor could there be economic justification for any such policy as the cost of recurrent pruning would far outweigh alternative methods of remediation. For example, even pruning 1% of the tree population could cost any where between £50-100 million. Thus, pruning universally is unlikely to be a viable method of alleviating subsidence risk.

Some trees, for instance many of the London plane trees in city streets, are pruned on a regular basis as part of their normal management. This project has indicated that there is justification for modifying their pruning regime to reduce the risk of subsidence by reducing rather than thinning the crowns and using techniques which produce compact crowns.

SCIENCE SUMMARY

Background

Previously, water use of amenity trees has been calculated from assumptions made about root distribution patterns and soil drying. These models are empirically based on limited non-replicated data (Biddle, 1998 a,b) or by extrapolation from forest trees. Forest models of tree water use and re-growth following pruning are inappropriate because the chosen species are usually not representative of amenity ones. Furthermore, the surrounding aerial environment, edaphic conditions and spacing between trees all of which influence water use are entirely different to those found in urban situations. Very little published data is available on the actual transpiration of water by amenity trees and even less considers the additional impact of pruning. Thus, a greater understanding of principles which influence water use of isolated trees is vital to enable more accurate predictions of the impact of decreasing canopy size and root restriction treatments for a range of amenity species.

This project had four inter-related primary experimental objectives (described below). These used established and novel methodologies to determine the relationships between canopy leaf area, tree water use and soil drying (Figure I.1). The factors (vapour pressure deficit, radiation, temperature and wind speed) driving water loss external to the canopy were measured by use of automatic meteorological stations. Tree transpiration was determined by measuring sap flux using heat dissipation and heat pulse techniques in the main trunk below the canopy. Canopy leaf area was determined using hemispherical photography. Re-growth potential and regulation of water loss by leaves within the canopy was determined by measurement of assimilation rates, leaf water potential and leaf conductance. Stable isotopes of carbon ($\delta13C$) can normally be used to assess integrated leaf level water use, because different rates of incorporation of ^{12}C and ^{13}C in carbon dioxide (CO_2) leads to discrimination during photosynthetic activity, which normally is proportional to leaf pore (stomatal) aperture. One major approach was to investigate how the relationship between leaf water use and $\delta^{13}C$ was altered within dense individual tree canopies, where an unstirred "boundary layer" of air (see glossary) may limit the transfer of water and CO_2 from and to the leaf respectively. The development of an artificial leaf allowed evaporation rates to be compared with a second stable isotope that of ^{18}O in leaf water, which becomes enriched in residual water during evaporation. The influence of tree water use on soil drying under the canopy was measured directly using a neutron probe and by the use of stable oxygen isotopes in soil and plant water. These methodologies were then used to assess the effectiveness of two practical pruning methods designed to reduce tree water use through the control of canopy leaf area by mature trees.

An alternative method involving root restriction was developed and evaluated for controlling growth of newly planted trees. The data gained during all the phases of this work is integrated to improve the understanding of the principles which influence water use of isolated amenity trees following different pruning treatments. Thus, clear guidance and advice has been provided to the industry and government partners of the consortium.

Summary of scientific objectives and outcomes

Objective 1. Identify the critical components that influence isolated tree water use, determine how these can be best measured in the canopy, and how these components change in response to canopy manipulation

- Pruning significantly affected the total crown leaf area and shoots regrowth (Experiment 1).
- Hemispherical photography was an effective method to determine the impact of pruning on changes in canopy leaf area in isolated single trees (Experiment 1).
- Leaf water relations (conductance to water vapour, water potential) and assimilation rates of new shoots were influenced by pruning (Experiment 1).

- Carbon isotope (^{13}C) discrimination showed that increased stomatal conductance (water use efficiency) in crown-reduced trees occurred over the whole growing season (Experiment 1).
- Differences in tree water use directly after pruning were not explained by either measurements of leaf conductance or the environmental conditions outside the canopy (Experiment 1).
- Thermal dissipation probes were suitable for determination of sap flux in trees with an approximately 50 cm girth, but do not provide a good estimate in large amenity trees >1.5 m girth (Experiment 5).

Objective 2. Identify the main sources of water within the soil profile for tree water extraction and the influence of species and season

- Measurement of isotopic (^{18}O) signals in soil water were not reliable for distinguishing between soil water extracted from near the surface (10-20 cm depth) compared to that deeper in the soil (60-70 cm depth). (Experiments 1 and 2).
- The principle effects of crown reduction and crown thinning on soil moisture conservation were similar irrespective of trees size, age and species, although the magnitude of the effects varied from year-to-year and experiment-to-experiment (Experiments 1-5).
- Crown-reduction decreased soil drying by trees in the year of pruning, but the effects were generally small and disappeared the following season, unless the reduction was severe (>70% crown volume), in which case the effects were larger and persisted for up to two years (Experiment 1-5).
- Crown-reduction and crown-thinning had limited effects on conserving soil moisture under either soil droughted or non-droughted trees (Experiment 4).
- Crown-reduction had a greater effect than crown-thinning on conserving soil moisture towards the horizontal periphery of the root system (Experiment 5).
- Crown-reduction allowed faster and more complete recharging of soil water during a drier than average autumn and winter (Experiment 5).
- Severe crown-reduction (90% crown volume decrease) substantially decreased soil drying during a summer with less than average rainfall (Experiment 5).

Objective 3. Test the hypothesis that canopy manipulations by crown pruning and root restriction provide an effective way to control cherry and London plane trees growth and water use

- Wild cherry trees with crown volumes reduced by approximately 25-50 % completely recovered their canopy leaf areas within 1-2 growing seasons, due to increased terminal and lateral shoot growth (Experiments 1, 2 and 4).
- London plane trees with crown volumes reduced by approximately 70% completely recovered their canopy leaf areas within three growing seasons (Experiment 5).
- Crown-reduction produced trees with more compact canopies containing larger leaves, more closely packed together and with greater concentrations of nitrogen, (Experiments 2, 4 and 5).
- Crown-reduction extended the duration of leaf development by prolonging shoot extension and delaying leaf senescence during the year of pruning (Experiment 3).
- Crown-thinned wild cherry and London plane trees recovered their canopy leaf areas due to increased numbers of leaves, particularly on lateral shoots (Experiments 1, 2, 4 and 5).
- Terminal shoot growth, mean leaf size and leaf nitrogen concentrations were not affected by crown thinning (Experiment 2, 4 and 5).
- Crown-thinning did not influence the timing of seasonal growth (Experiment 3).
- The effects of canopy removal on reducing tree water use (sap flux) in wild cherry trees were variable and the maximum effects persisted for only four months after application (Experiments 2 and 4).
- Crown-reduction and crown-thinning increased (less negative) leaf water potentials compared to non-pruned trees in the year after pruning (Experiments 2, 4, and 5).
- Pruning did not affect leaf water relations within the canopy beyond the year of application (Experiment 2 and 4).

- Potential water loss (leaf conductance) from crown-reduced wild cherry and London plane trees was greater than for crown-thinned and non-pruned trees (experiment 2, 4 and 5).
- Higher ^{13}C discrimination in crown-reduced trees was not associated with low water use efficiency, since transpiration rates were reduced by low boundary layer conductance in the dense canopy (Experiments 2, 4 and 5).
- Root restriction within geotextile lined planting pits provided an effective way to control tree size and vigour (Experiment 6).
- The shoot extension growth was closely correlated with the volume of the planting pit (Experiment 6).

Objective 4. Determine the influence that changes in the environment have in the control of water use in isolated trees, and develop an understanding that enables responses to canopy manipulation of different species to be predicted

- Boundary layer conductance was identified as a key factor determining tree response to evaporative demand following pruning (Experiments 1 and 2).
- Evaporative enrichment of [18]O in leaves was shown to provide a good estimate for overall leaf water use (Experiment 2).
- The combined use of ^{13}C and [18]O as a diagnostic tool for determining the physiological basis of the interaction between boundary layer conductance and leaf level gas exchange was an important development associated with this project.
- The use of ^{13}C and [18]O isotopes to distinguish assimilation (carbon) limitation and water loss in leaves of shoots following pruning produced similar results in London plane and wild cherry trees (Experiments 4 and 5).
- The fastest rate of evaporation occurred from leaves within the canopies of crown-thinned trees and the slowest in the canopies of crown-reduced trees (Experiments 2 and 4).
- Leaves of crown-thinned trees were shown to be more closely coupled to conditions in the external atmosphere causing greater water loss thus had a lower Ω factor (Jarvis & McNaughton, 1986) than non-pruned trees (Experiments 2 and 4).
- The lower boundary layer conductance in crown-reduced trees moderated the effect of greater stomatal aperture (c.f. non-pruned and crown-thinned trees) by limiting water use through restricted diffusion away from the leaf surface into the bulk atmosphere (Experiments 2 and 4).

Summary of experiments and results

Two different commonly practised dormant season pruning techniques were used to reduce canopy leaf area. One was 'crown reduction', in which the potential leaf canopy is lessened by removing the outer portions of all major branches, this reduces the total volume of the crown. The other was 'crown thinning', in which the number of side branches coming off all of the major branches is reduced (not affecting the original volume of the canopy). For controlling tree growth at planting, restriction of roots within pits lined with a geotextile permeable to water and mineral nutrients, but not to roots was used.

Experiment 1. Validation of approaches: isolated tree water use as a function of crown pruning (Objective 1)

Comparing the effects of crown reduction and crown thinning on the first year re-growth and water use of wild cherry (Prunus avium, Plena) trees

The experiment used twenty-four 15-year-old trees (8 m height) spaced 9 m within the tree row and 5.5 m between rows. A 30% (height decrease by visual estimate) crown reduction was applied by professional arboriculturists to one third of the trees on 10 April 1999 and 30% (lateral branch removal by visual estimate) crown-thinning was applied to another third of the trees on 17 April 1999, the remaining trees were non-pruned controls (Figure I.2). The crown-reduction actually

decreased canopy volume by 47%. The experimental design was a randomised complete block with three treatments and eight replicates. Each tree formed a single plot.

- Hemispherical photography was developed and validated as a very effective way to measure and compare the impact of pruning on changes in canopy leaf area in isolated single trees.
- Crown-reduction caused increases in leaf conductance to water vapour and assimilation rates (photosynthesis) and less negative leaf water potential (drought stress) on new shoots.
- Carbon isotope (^{13}C) discrimination showed that increased stomatal conductance in crown-reduced trees was a feature present over the whole growing season.
- Crown-reduction conserved soil moisture compared to non-pruned trees directly after pruning, whereas crown thinning had no effect.
- Differences in tree water use directly after pruning were not explained by either measurements of leaf conductance or the impact of the environmental conditions outside the canopy.

Experiment 2. Long term impact of crown pruning and environment on tree water use
(Objectives 2, 3, 4)

Determining the effects of crown reduction and crown thinning on the growth and water use of wild cherry (Prunus avium, Plena) trees and the optimum period before re-pruning

Another group of 24 trees adjacent to those described in experiment 1 and of the same age and similar size was used for this experiment. The canopy pruning treatments were completed on 11 March 2000. A 30% (height decrease) crown reduction was applied to eight trees and a 30% (lateral branch removal) crown-thinning was applied to another eight trees. These treatments were compared with eight non-pruned control trees. Crown-reduction actually reduced canopy volume by 39%. The experimental design was a complete randomised block with three treatments and four replicates, each treatment plot contained two trees.

One of each of the two trees in each of the crown-reduced plots was re-pruned back to its original branch lengths on 17 April 2002. One of each of the two trees in each of the crown-thinned plots was thinned again at the same time and the control trees remained non-pruned. So the effects of repeated 2-year cycle pruning and pruning once only were compared directly (Figure I.3). The experimental design of the modified experiment was a split plot with pruning treatment as the main plot and frequency of pruning as the sub-plot, each treatment contained four replicates.

- A two year pruning cycle with crown-reduction or crown-thinning did not conserve moisture at anytime after pruning.
- Crown-reduced trees recovered their canopy leaf areas within one growing season; leaf areas rapidly increased with little change in crown volumes (i.e. leaf area density increased).
- Increases in leaf nitrogen content, leaf size, number of leaves per shoot and shoot length were associated with the greater leaf area density found in crown-reduced trees.
- Evaporative demand within the canopy of crown-reduced trees was decreased compared to non-pruned or crown-thinned trees because the atmosphere within the canopy was more saturated with water vapour.
- Crown-thinned trees required two growing seasons after pruning to recover their canopy leaf area. Terminal shoot growth and mean leaf sizes were not affected by the pruning.
- Leaves of crown-thinned trees were shown to be more closely coupled to conditions in the external atmosphere causing greater water loss than non-pruned trees.
- Greater ^{13}C discrimination in crown-reduced trees was not associated with greater water use, but increased stomatal opening, allocation of nitrogen to chlorophyll and assimilation rates.
- Measurement of isotopic (^{18}O) signals in soil water were not reliable for distinguishing between soil water extracted near the surface (10-20 cm depth) and deeper (60-70 cm depth), however evaporative enrichment of ^{18}O in leaves was shown to provide a good estimate for overall leaf transpiration.

Experiment 3. Effects of crown pruning on the phenology of shoot re-growth
(Objective 3)

*Determine the effect of crown reduction and crown thinning on shoot development, extension and longevity on wild cherry (*Prunus avium*) trees*

The experiment contained nine 15-year-old trees (8 m height) spaced 9 m within the tree row and 5.5 m between rows. A 30% (height decrease) crown reduction was applied by professional arborists to one third of the trees and 30% (lateral branch removal) crown-thinning was applied to another third of the trees on 23 March 2001, the remaining trees were non-pruned controls. The experimental design was a randomised complete block with three treatments and three replicates. Each tree formed a single plot. Unfortunately, extensive long-term flooding in the previous winter had damaged the trees in one block and these trees were excluded.

- Crown-thinning did not influence the timing of seasonal growth (phenology).
- Crown-reduction extended the duration of leaf development by prolonging shoot extension and delaying leaf senescence during the year of pruning.

Experiment 4. Impact of drought on response to crown pruning
(Objectives 2, 3, 4)

Interaction of soil drought and pruning on the re-growth and water use of wild cherry (Prunus avium) trees

This experiment was designed to determine the interaction between soil drought and crown-reduction and crown-thinning treatments on tree water use. Twenty-four 11-year-old trees (7.4 m height) spaced at 8 m between trees and 6 m between rows were used. Eight trees were crown-reduced (30% height decrease by visual estimate) and another 8 were crown-thinned (30% lateral branch removal) on 13 April 2002. In addition, half of the crown-reduced trees and half of the crown-thinned trees had sloping plastic covers 5 m $\times$ 8 m placed over the ground around them to deflect rain. The installation was completed during the period 8-12 April 2002 (Figure I.4). The design of the experiment was a split-plot with pruning treatment as the main plot and soil drought as the sub-plot, each treatment contained four replicates.

- Crown-reduction and crown-thinning had limited effects on conserving soil moisture under either soil droughted or non-droughted trees.
- Crown-reduced trees recovered their canopy leaf areas and greatly increased their leaf area density within one growing season due to increased terminal and lateral shoot extension and leaf size.
- Crown-thinned trees recovered their leaf areas within two growing seasons due to increased numbers of leaves, particularly on lateral shoots.
- Crown-reduction and crown thinning decreased daily sap-flux for only four months after application. Crown-reduction had a greater effect than crown-thinning.
- The fastest rate of evaporation occurred from leaves within the canopies of crown-thinned trees and the slowest in the canopies of crown-reduced trees.
- The lower boundary layers conductance in the canopy of the crown-reduced trees moderated the effect of greater stomatal aperture (c.f. non-pruned and crown-thinned trees) by limiting the rate at which transpired water vapour diffused away from the air immediately surrounding the leaf surface into the bulk atmosphere.
- Crown-reduction and crown-thinning increased (less negative) leaf water potential compared to non-pruned trees in the year after pruning.
- Pruning did not affect leaf water relations within the canopy beyond one growing season.
- The drought applied in this experiment had no apparent effect on leaf water relations or tree stress levels.

Experiment 5. Assessing the impact of crown pruning on large amenity trees
(Objectives 2, 3, 4)

Determining the effects of crown reduction and crown thinning on the growth and water use of mature London plane (Platanus hispanica) trees and optimum interval between re-pruning

This experiment contained twenty-four 28-year-old London plane trees (20 m height) within a single row spaced at 12 m between trees. The pruning treatments were applied 24 January-15 February 2000. Crown-reduction (30% height decrease by visual estimate) was applied to eight trees and crown-thinning (30% lateral branch removal) to another eight trees (Figure I.5). The crown-reduction treatment reduced canopy volume by 72%. The experimental design was a complete randomised block with three treatments and four replicates, each treatment plot contained two trees.

Four of the crown-reduced and three each of the crown thinned-trees and previously non-pruned trees were severely crown reduced (60% height decrease, 90% volume reduction) on 14 and 17 April 2003. One of each of the two trees in each of the respective plots was reduced (Figure I.5). Thus, the effects of severe crown reduction in 2003 and single crown-reduction and crown-thinning in 2000 were compared. The design of the modified experiment was a split plot with pruning treatment as the main plot and re-pruning as the sub-plot.

- Crown-reduction and crown-thinning conserved soil moisture for three growing seasons after application, but crown-reduction had a greater effect towards the horizontal periphery of the root system.
- Crown-reduction allowed faster and more complete recharging of soil water during a drier than average autumn and winter.
- Severe crown-reduction (90% volume decrease) substantially decreased soil drying during a summer with less than average rainfall.
- Differences in soil drying after severe crown-reduction did not correlate with sap flux measured by the thermal dissipation probes. Therefore, their suitability for use on large trees is open to question.
- Crown-thinned trees recovered their total canopy leaf areas within two growing seasons and crown-reduced trees recovered theirs within three growing seasons.
- Crown-reduction produced trees with more compact canopies containing larger leaves, with greater concentrations of nitrogen, more closely packed together, whereas as crown-thinning had no impact on leaf size and nitrogen concentration.
- Potential water loss (leaf conductance) from leaves of crown-reduced trees was greater than in those of crown-thinned and non-pruned trees.
- The use of ^{13}C and ^{18}O isotopes to distinguish carbon limitation and water loss produced similar results in London plane to those found for wild cherry trees.

Experiment 6. An alternative approach to controlling growth in newly planted amenity trees
(Objective 3)

Effect of root restriction on the growth of 'heavy standard' Lime (Tilia cordata) and Norway maple (Acer platanoides) trees following planting

Lime (girth 14.5 cm) and Norway maple (girth 13.4 cm) trees were planted as separate experiments at 8×8 m spacing between 27 April and 12 May 2000. Three different sizes of pit; $0.6 \times 0.6 \times 0.7$ m (250 litre), $1.2 \times 1.2 \times 0.7$ m (1000 litre) and $1.7 \times 1.7 \times 0.7$ m (2000 litre) were dug (Figure I.6). These pits were lined with the geotextile Terram 3000 (Exxon Chemical Geopolymers Ltd, Mamhilad Park, Pontypool, Gwent). The geotextile was of sufficient width that no seams were necessary. The experimental designs were separate randomised complete designs with four treatments (including unrestricted control trees) and six replicates. Each treatment plot contained one tree.

- Root restriction provided an effective way to ensure that tree vigour was controlled.
- The shoot extension growth was closely related to the volume of the planting pit, thus giving potential to predict tree size and therefore water use.
- A lined planting pit of 2000 l was ample to support full growth for four growing seasons whilst preventing root intrusion to surrounding soil.

Experimental sites

Experiments 1, 2, 3, 4 and 6 were carried out at Rocks Farm, Horticulture Research International (HRI), East Malling, on a soil of the Langley series, which has approximately 1.5-2.0 m silty loam overlying ragstone. The soil is stony at > 1 m depth. Experiments 4 and 6 were planted on soil > 2 m depth. Experiment 5 (London plane trees) was carried out at HRI, East Malling on a soil of the Malling series, which has 0.7-1.2 m fine sandy loam overlying clay loam

Technical innovations developed during the project

- Evaluation of stable isotope discrimination for leaf water use and canopy environment

The combined use of ^{13}C and ^{18}O as a diagnostic tool for determining the physiological basis of the interaction between boundary layer conductance and leaf level gas exchange was an important development, supported by data from both wild cherry and London plane trees. The interactions between ^{13}C and ^{18}O have provided tremendous scientific insight into the co-limitation of carbon gain and water use in isolated tree canopies, and suggest that ^{13}C alone cannot be used as direct indicator of plant water use efficiency in these circumstances. For the benefit of the industry, it may be possible to rank tree water use from ^{13}C and ^{18}O as a means for selecting suitable trees with reduced water use for replanting in urban environments.

- Development of 'artificial leaves' for assessing canopy micro-environment

This will enable improved prediction of water extraction by trees because it allows the effect of canopy shape, size and volume on evaporative demand to be determined. Thus, allowing more appropriate pruning practices to be developed that conserve soil moisture. East Malling Research is an academic partner in a new HortLINK project that is considering the commercial exploitation of artificial leaves for improving irrigation efficiency in hardy ornamental stock production.

- Use of hemispherical digital photography for single trees

This provides a rapid method for quantitative assessment of tree size and the impact of pruning on canopy leaf area that was impossible to do previously. Since canopy leaf area is also related to the condition of the tree, hemispherical digital photography could be developed further as a method of assessing tree health and any (non-pruning induced) decline occurring over time.

- Improved design of heat dissipation probe for measuring sap flow

The Granier type heat dissipation probe is widely used in forest tree research. The commercially available probe has only a short life (2-3 months), and is generally not economically repairable following failure. Thus, making it unreliable and expensive to use on long-term experiments. The improved design developed here was more robust, as probes withstood multiple insertions and extractions for up to three years and therefore substantially reduces the cost of this type of work.

- Redesign of Profile probe

Since the neutron probe has a radioactive source, it requires a licence for its use to satisfy health, safety and security issues. Registered users are generally restricted to Universities, other research

based organisations and irrigation advisors. Therefore, it is not used frequently for site investigations of tree implicated subsidence. The Profile probe developed by Delta-T Devices provides a non-radioactive method for measuring soil water content and does not require a licence. Thus, it can be used by any aboricultural, engineering and environmental consultants dealing with subsidence issues. Since a high proportion of subsidence claims are related to trees on clay soils, the redesigned (Mark II) probe, developed as a result of this project, will give greater accuracy in use on clay soils and provide a useful and inexpensive tool for consultants investigating soil drying by trees.

Knowledge transfer during the project

The fifth objective of this project was to 'Deliver prevention and risk management guidelines for all interested parties based on scientific principles and amenity tree values to enable economic and long-term control of water use'.

Oral presentations (presenter in brackets, excluding co-authors)

1. 19 March 1999, Controlling water use of trees to alleviate subsidence risk. London Tree Officers Association, Kingston-upon-Thames (N A Hipps)
2. 7 September 1999, Trees, water use and subsidence, 33rd National Arboriculture Conference, Technical Seminar and Trade Exhibition, Keele University (N A Hipps)
3. 5 May 2000, Controlling water use of trees to alleviate subsidence risk London Tree Officers Association, Kensington Town Hall (N A Hipps)
4. 22 June 2000, Trees the Infinite Sponge. 'Trees 2000 Challenges for the future'. Keele University. Organised by the Tree Advice Trust & Institute of Chartered Foresters, (N A Hipps)
5. 19 September 2000 Controlling water use of trees to alleviate subsidence risk. Arboricultural Association Annual Conference 'Reaching Out' Exeter University (T Ball)
6. 7 June 2001. Trees and subsidence, Capel Manor College/ Middlesex University
7. Lecture to MSc students studying Resource Management/ Arboriculture and Countryside Management (N A Hipps)
8. 20 December 2001 Boundary layers and leaf-canopy coupling II: A canopy level approach. British Ecological Society Winter Meeting, University of Warwick. (J Dunn)
9. 15 May 2002 Controlling water use of trees to alleviate subsidence risk, Midland Tree Officers Group, South Staffs District Council at Codsall nr Wolverhampton (N A Hipps)
10. 25 July 2002 Does pruning reduce tree water use? London Tree Officers Association, Town Hall, Royal Borough of Kensington and Chelsea (N A Hipps & J Dunn)
11. 20 November 2002 Does pruning reduce water use? Kent Nursery Stock Committee, HRI, East Malling (N A Hipps)
12. 14 January 2003, Does pruning reduce tree water use? Royal and Sun Alliance, Berkley Square, London (N A Hipps & H Griffiths)
13. 20 May 2003, Does pruning reduce tree water use, Association of British Insurers, London. (N A Hipps & H Griffiths)
14. November 2003, Moving the boundary: use of ^{13}C and ^{18}O to go beyond stomatal limitation SIBAE-BASIN joint international conference on stable isotopes, Orvieto (H Griffiths)
15. November 2003, From urban subsidence to tropical cloud-forest epiphytes: integrating ^{18}O signals across biomes IAEA Discussion meeting on establishing a network for ^{18}O measurements in water (H Griffiths)

Posters

1. 31 March 2003. The effects of two different canopy manipulations on leaf water use and photosynthesis as determined by gas exchange and stable isotope discrimination. Society for Experimental Biology Annual Main Meeting 31 March–4 April 2003, Southampton, (J M Dunn, N R Betson, C J Atkinson, H Griffiths & N A Hipps)
2. 27 February 2004. Controlling water use of trees to alleviate subsidence risk. HortLINK Conference

Popular articles

1. Research and rescue. *Horticulture Week*, 26 July 2001, pp 14-16 (E McMillan, & N A Hipps)
2. Finding the root cause *Horticulture Week*, 26 February 2003, pp 18-21 (J Abbott)

Fact sheet and web site

1. 1999. N A Hipps, & G Poole Controlling water use and subsidence risk. LPC, Borehamwood, Herts
2. 1 July 2002. http://www.hri.ac.uk/research/Hlp212/index.htm website giving latest results from the project (N A Hipps)

Future

Oral presentations

1. 16 July 2004. London Tree Officers Group (N A Hipps)
2. 20 September 2004. National Arboriculture Conference, University College, Chester. (N A Hipps)
3. April 2005. Research for amenity Trees Conference (speaker to be confirmed)

Publications

1. Article to summarise results of final report on East Malling Research website with link from Office of Deputy Prime Minister and Arboricultural Association (to go live September 2004)
2. Four Arboriculture Research and Practice Notes:
   Root restriction and shoot growth (September 2004)
   Shoot pruning to manipulate water uptake (November 2004)
   Canopy characteristics as a measure of tree vitality (March 2005)
   Loss of water from trees – the importance of the boundary layer (February 2005)

Scientific papers in preparation

1. Dunn J, Davies M, Fordham M, Dodds P, Betson N, Atkinson C J, Hipps N and Griffiths H. Water use by isolated tree canopies: integration of leaf level water use and stable isotopes to characterise boundary layer limitations. For submission to *Plant Cell Environment*, September 2004
2. Hipps N, Davies M, Dunn J, Dodds P, Atkinson C J, Hipps N, Griffiths H The effects of two contrasting pruning regimes on water use by large urban trees (London plane). For submission to *Tree Physiology*, October 2004
3. Dunn J, Hipps N, Griffiths H and Atkinson CJ Impact of crown pruning regimes on the phenology, leaf morphology and physiology of *Prunus avium*. For submission to *Tree Physiology*, November 2004

Exploitation plans and further dissemination of information

It is the intention of the consortium that all positive results with practical application emerging from the project should be exploited by any party, whether a consortium member or not, if the result of such exploitation is a reduction in the impact of tree-related subsidence. One of the main aims of the project is to engage support for and use of all valid findings from the research, through wide discussion of the results amongst interested parties. This will be encouraged by (1) writing articles for the technical journals of a number of business sectors (local government, insurance, amenity and environment, transport etc.) and (2) making presentations at key meetings and conferences. Each of the consortium members will lead this activity within their own sector.

ABI, and member insurance companies

- Insurers will be encouraged to use the information in advising existing and new clients about best practice risk management and when instructing professional advisers (such as loss adjusters and engineers) in connection with claims.
- Guidance documents will be prepared and published incorporating the results into market codes of practice.
- ABI will issue guidance to members outlining the steps that might be taken to exploit the results of the project by December 2004.

Arboricultural Association and Professional Arboriculturists

- Recommendations on pruning can now be scientifically rather than empirically based.
- The information will be used to update management advice for trees that will reduce the impact of tree-related subsidence.
- Four advice notes are in preparation and will be issued through the Tree Advice Trust within the Arboricultural Notes series. These will be released from November 2004 to April 2005. These are widely publicised and made available to tree officers in local authorities, town planners, education establishments, research institutes, other professionals and the general public.
- Guidance on the management of trees is given in two British Standards:
    BS 5837:1991 Guide for trees in relation to construction, and
    BS 3998:1989 Recommendations for tree work.
  It is British Standard's practice to review such documents regularly and as appropriate convene panels to recommend necessary amendments. Currently, both documents are undergoing extensive rewriting to reflect current practices, new knowledge and innovations. The timely completion of this Horticulture Link programme will influence the thinking of panel members and thus future tree management through the texts of the Standards.
- London Tree Officers' Association leaflet on subsidence damage to trees is undergoing revision and the results of the Horticulture Link programme will be included.
- The results will be brought to the attention of the National House-Building Council (NHBC) for consideration when their Standards are being revised.

BRE

- The research will be exploited by utilising the understanding, techniques and devices developed, as well as partnerships forged during the project to further its engineering research into subsidence and technical advice.
- BRE will draw attention to the outputs of this project as appropriate in its provision of advice to government and private sector clients. We strongly advocate the adoption of best practice in the risk management of trees where they pose a threat to buildings, as defined by current guidance and the outputs of this research, to develop the best outcome for both tree and building.

BRE will develop ideas further to:

- Offer guidance for builders on choice of tree, location and root barriers.
- Give guidance for town planners and developers on planting regimes and specification of species, location and root barriers.
- Provide standards for testing and certification of root barrier systems, geotextiles and installation process.
- Transmit results to Local Authorities through ALARM and LGA.

Cambridge University

- A grant application to NERC is in preparation. The interplay between ^{13}C and ^{18}O will be used to investigate coupling within forest canopies underpinned by the principles discovered in the HortLINK project.
- Collaborating with a mass spectrometer manufacturer (confidential) for the development of a portable instrument to measure instantaneous gas exchange and allow the ^{13}C and ^{18}O exchanges to be captured in real time as a function of changing boundary layer conditions in the field.
- Broadening the application of combined ^{13}C and ^{18}O analysis for investigating water use in tropical forest epiphytes as a function of altitudinal gradients and climate change in cloud forests, and also for investigating diffusion limitation in Antarctic bryophytes. Ongoing.
- Three papers for the international scientific journals (see future publications).
- Joel Dunn will provide a PhD thesis by November 2004 that will be available for public inspection and loan through The British Library and/or Cambridge University.

Delta-T Devices

- This project has shown the potential future use by professionals of the company's systems for tree water use and canopy leaf area measurements. As a result, the single trees analysis algorithms were developed and evaluated for the Hemispherical digital photographic image analysis system (marketed as HemiView). The single tree analysis facility is now incorporated into the HemiView system marketed by Delta-T Devices.
- A major redesign of the Profile Probe (a device that determines soil moisture content by measuring dielectric constant) has been undertaken to overcome problems identified by the academic partners with its use in certain clay soils. Many of the problems associated with the current device have been overcome due to close cooperation between Delta-T Devices and East Malling Research and a MKII device is now well on the way to becoming available and will be marketed in autumn 2004.
- The assessment of the thermal dissipation probes by East Malling Research has helped considerably in establishing the value of their use in the UK market. Delta-T Devices and East Malling Research are discussing collaboration over the design of a more robust and reliable Thermal Dissipation Probe for use in the UK market.

East Malling Research

- East Malling Research is already gaining additional value from HortLINK 212 by exploiting the knowledge gained about methodologies, techniques and the principles of measuring water use and canopy growth in trees in other projects. This includes research relating to controlling water use in fruit and hardy ornamental shrubs commissioned by Defra Horticultural Development Council and Horticulture Link.
- Article to summarise results of final report for the East Malling Research website are being developed with links from Office of Deputy Prime Minister and Arboricultural Association and Horticulture Link. These will go live by October 2004.
- Three papers are in preparation (jointly with University of Cambridge) for peer reviewed international scientific journal based on the results of the project. (See Cambridge University and future publications.)

- Results will be presented to the London Tree Officers Group 16 July 2004 and the Arboricultural Association Annual Meeting 20 September 2004.
- Article for the amenity horticulture trade journal *Horticulture Week* in preparation.

East Malling Trust for Horticultural Research

- East Malling Research will publicise the HortLINK 212 results through an article in the annual report of the East Malling Research Association published in December 2004. This report is presented to all members of the association and membership is open to companies and private individuals.

Highways Agency

- The Highways Agency (HA) will disseminate the findings of the HortLINK 212 Project to its Environment Group advisers initially (September 2004) and will prepare new guidance for incorporation in Volume 10 of the Design Manual for Roads and Bridges (DMRB) for publication in May 2005. This will require changes to both the Landscape Management Handbook and the Good Roads Guide Series of Advice Notes.
- The results of the HortLINK 212 Project will be presented to the Trunk Road Management Conference in September 2004 for the attention of the HA's Managing Agents and their associated consultants. It is anticipated that the project's recommendations will improve the cost effectiveness of the management of mature/older highway trees particularly in urban situations and in the built environment.

Office of the Deputy Prime Minister

- The Department will review opportunities and priorities for arboricultural research in the light of the outcomes and recommendations of this project.
- The outcomes of the project will inform the development and revision of policy and guidance in respect of tree planting and management, particularly in relation to urban trees.
- Encouragement will be given to disseminate and discuss the implications of the project findings.

Future R&D

The following proposals have been submitted to the Association of British Insurers and the Office of the Deputy Prime Minister for consideration.

Proposals:

1)   *Identifying the risk posed by the existing trees and shrubs and predicting future risk of newly planted trees and shrubs to existing buildings*

a)   *Objective*

Improve knowledge of urban tree water use and the likely impact of climate change.

Methodology
Use modern analytical techniques (including stable isotope technology) to improve the scientific basis to characterise the differences in urban tree and shrub water use *in situ* on a wide range of species and relate this to environmental conditions and growth.

Benefits
To provide knowledge and improved models for Insurers to assess the prediction of risk of subsidence caused by trees in an urban setting and therefore assist Government sustainability plans by reducing the number of trees removed because of perceived subsidence risk.

b)   *Objective*

Improve understanding and prediction of root proliferation and distribution in urban tree species and relate root distribution to water use.

Methodology
Use 'urban tree' plantings at East Malling research to determine root distribution and growth rates. Develop potential use of ground penetrating radar (GPR) and other techniques to non-destructively survey growth of *in situ* urban tree root systems and relate to tree size, age and species.

Benefits
To provide a simple non-intrusive means by which Arboriculturists and other advisors can assess in more detail whether trees that have been identified as posing a subsidence risk to a given building are likely to do so.

c)   *Objective*

Improve understanding of the urban tree-soil-building system as a determinant of subsidence damage.

Methodology
Use analytical techniques to determine patterns of soil water drying and foundation response in urban situations compared with green field sites. Include modern methods of construction.

Benefits
To provide information and improved models for Insurers and developers to assess the prediction of risk of subsidence caused by trees in an urban setting.

2)   Managing the risk of existing trees and shrubs to existing buildings

a)   Objective

Develop more effective remedial actions than those currently recommended to control tree water use.

Methodology
Investigate effects of novel treatments on tree water use e.g. use of vertical soil membranes, pollarding shoots, ring barking to reduce subsequent growth and determine the impact of frequently repeated pruning. Relate this to soil drying capacity and extent.

Benefits
Provision of alternatives to tree removal to prevent subsidence damage.

3)   Managing the future risk of newly planted trees and shrubs, both to new and existing construction

a)   Objective

Provide guidelines on management of new urban vegetation, i.e. how, what and where to plant.

Methodology
Literature review and experiments on the influence of species on water use in relation to tree and shrub size, root location and shoot growth rate. Determination of minimum volumes to completely restrict roots and ensure tree survival given the potential for climatic changes. (The latter will also provide a feasibility for the more frequent replacement of urban trees).

Benefits
Enable sustainable methodologies for selection and planting of new trees that will not pose a risk of vegetation-induced subsidence.

b)   Objective

Develop novel approaches that could be included particularly in new developments to manage drainage ('grey') water to control and influence root growth away from foundations or other construction.

Methodology
Use field based experiments near new and old buildings to determine the potential to manage the interactions between root distribution, soil water placement and soil drying. Determine the effects selective root killing on young and mature trees.

Benefits
To enable the Insurance industry and others to influence regulators and developers to provide drainage systems that minimise subsidence risks to new and existing constructions.

Potential collaborators for proposals 1-3: East Malling Research, Prof. H Griffiths (University of Cambridge), BRE, Arboricultural Consultants (several have expressed an interest).
Potential Co-funders: Office of the Deputy Prime Minister (ODPM), Highways Agency (HA), Association of British Insurers.

4)   Novel use of stable isotopes to determine water use in forest canopies

A grant application to NERC is in preparation by Prof. H Griffiths, University of Cambridge. The interplay between ^{13}C and ^{18}O will be used to investigate coupling within forest canopies underpinned by the principles discovered in the Horticulture LINK project.

5)   Development of a portable mass spectrometer for field use

Prof H Griffiths, University of Cambridge is collaborating with a mass spectrometer manufacturer for the development of a portable instrument to measure instantaneous gas exchange and allow the ^{13}C and ^{18}O exchanges to be captured in real time as a function of changing boundary layer conditions in the field.

Figure I.1: Factors influencing soil drying by trees and methods of measurement

Evaporative demand outside canopy
- Vapour pressure deficit (Humidity)
- Temperature
- Wind speed
- Solar radiation

Evaporative demand within canopy
- Artificial leaf

Growth
- Shoot extension
- Leaf size
- Seasonal phenology

Leaf water relations and photosynthesis
- Gas exchange (H_2O and CO_2)
- Water potential
- Stable carbon and oxygen isotopes

Canopy leaf area
- Hemispherical photographic images

TRANSPIRATION
- Thermal dissipation probes
- Heat pulse probes

Water location
- Oxygen isotopes

Soil moisture
- Neutron probe
- Soil dielectric constant (Profile probe)

Figure I.2: Crown pruning treatments applied to wild cherry trees in 1999 (Experiment 1)

Non-pruned

Crown-reduced

Crown-thinned

Figure I.3: Crown-reduction and crown-thinning treatments applied to wild cherry trees in 2000 and repeated in 2002. Showing the effects on canopy growth in summer 2002. (Experiment 2)

Figure I.4: The application of crown-reduction and crown-thinning to wild cherry trees in 2002 for soil-droughted trees (Experiment 4). The plastic covers excluded rain around the tree

Non-pruned

Reduced

Thinned

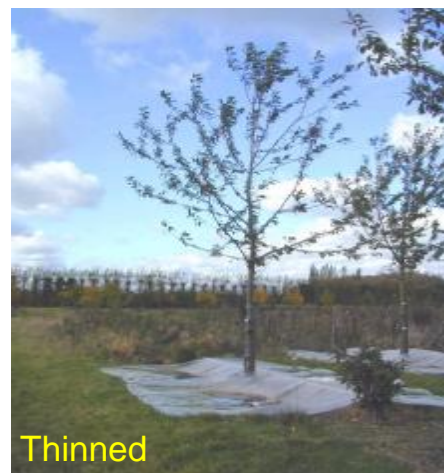Figure I.5: Crown-reduction and crown-thinning treatments applied to London plane trees in 2000 and 2003 (Experiment 5)

Figure I.6: Root restriction effects on lime trees two years after planting. Volume of restriction is given in Figure

APPENDIX – SCIENTIFIC REPORT

Objective 1:

Identify the critical components that influence isolated tree water use, determine how these can be best measured in the canopy, and how these components change in response to crown pruning.

Rationale

The results in this section are based exclusively on the data derived from Experiment 1. The aim was to validate approaches to measure isolated tree water use and growth as a function of pruning and provide preliminary identification of critical physiological and environmental factors that influence water loss from trees.

Since trees are pruned in an attempt to reduce leaf area and subsequent water use, it is important to measure canopy leaf area to determine the impact of the pruning treatment. Hence, our development of image analysis with hemispherical photography for this purpose. The components of canopy growth (i.e. shoot length and individual leaf area) also need to be measured to understand responses at the whole tree level.

Water is drawn to the leaves by a gradient of water potential (Ψ) between the soil and the leaf and ultimately the atmosphere. The water travels in the direction of more negative potential. Therefore, as soil dry and its water potential declines to maintain the hydraulic link and movement of water to the shoot, leaves must have a more negative water potential than the soil. Thus, leaf water potential can be used as a measure of the drought stress experienced by the trees. Pre-dawn measurements of leaf water potential, made when leaf water status has come in to equilibrium with soil water potential (i.e. no transpiration loss from the leaves), gives an estimate of soil water available to the tree. The lower the leaf water potential values (i.e. more negative) the less water there is available in the soil for the tree to use.

In the presence of light, trees lose water by transpiration via the stomatal pores on the leaf. The amount of water lost is partly dependent on the aperture of the stomata (measured as leaf conductance). As the stomata open in response to light, transpiration rate often also increases causing the leaf water potential to become more negative as the water deficit within the plant increases. Evaporative demand is driven by atmospheric humidity (vapour pressure deficit), sunshine (radiation), temperature and wind speed. Thus, water vapour loss from leaves may be closely coupled to the prevailing micro-climate in their immediate vicinity. Therefore, to understand the factors driving tree water use it is necessary to measure leaf water potential, leaf conductance to water vapour and the external climatic conditions.

Scaling up water use from individual leaves to an entire canopy should be undertaken with caution (e.g. Hinckley *et al*., 1994, Whitehead, 1998). It is necessary to elaborate upon the coupling with the atmosphere within and outside the canopy, leaf size, wind speed and stomatal conductance (Wullschleger *et al*. 1998). To overcome problems of extrapolation from single leaf measurements within natural and managed eco-systems, an increasing number of studies are reporting on direct measurements of whole tree water use by continuous measurements of sap flow through the tree (Wullschleger *et al*., 1998). Sap flow (i.e. the total quantity of water moving from the roots through the trunk to the leaves) is determined by a number of methods including use of heat balance, heat pulse and thermal dissipation techniques (Smith and Allen, 1996). Therefore, development of these methods for use on amenity trees has been included in the first experiment.

Ultimately, the integration of the factors driving water loss within and outside the canopy result in soil drying by the roots. This was measured directly within the rooting zone using a neutron probe. Growth and water relations data at the leaf level has been integrated with that from the

from the entire canopy to provide an indication of the critical canopy components influencing water use following pruning.

<u>Validation of approaches: isolated tree water use as a function of crown pruning (Experiment 1, Objective 1)</u>

Soil drying by wild cherry trees in April 1999

Methods

Soil moisture was determined using a neutron probe (Didcot Instruments, Abingdon UK) approximately every two months from May 1999 until September 2000. Access tubes were inserted to a depth of 1 m, at 1 m distance from each of the 24 experimental tree. Soil moisture deficits (mm) were referenced to 9 February 2000 when the soil was at field capacity. The neutron probe had been field calibrated in soil of the same type.

The Profile Probe (Delta-T Devices, Cambridge) which determines soil moisture content by measuring its apparent dielectric constant was found to be very inaccurate in the clay found in soils at HRI East Malling and therefore was not used.

Results and conclusions

Total soil moisture deficit throughout the whole soil profile increased during spring and summer 1999 as the trees lost water by evapotranspiration and the differences between treatments also increased during this period (Figure 1.1). The differences in moisture deficit in soil under trees receiving the different pruning treatments were generally <30 mm. The soil remained wetter under the crown reduced trees than under the crown thinned or non-pruned trees between May and November 1999; differences between treatments were significant in July (P=0.059), September (P=0.095) and November (P<0.05). The largest differences in soil moisture occurred near the surface at 20-60 cm depth (Figure 1.1) Non-pruned and crown-thinned trees had similar moisture deficits throughout the year because soil moisture contents were the same. Re-hyrdation occurred more rapidly in soil under the crown-reduced trees.

In the second year after pruning, between July and September 2000 the soil moisture deficit under the non-pruned trees was greater than under either the crown-reduced or crown-thinned trees, but the differences were not significant (P>0.30). The maximum soil moisture deficit was reached during August. Again the differences in moisture deficit were mainly due to differences in soil moisture content at 20-60 cm depth.

- Soil remained significantly wetter near the surface (20-60 cm depth) under the crown-reduced trees and reached field capacity quicker in the winter than either crown-thinned or non-pruned trees in the year of pruning.
- Differences in soil moisture deficit between soil under crown reduced trees and the other treatments were small (< 30 mm).
- The effects of crown reduction on soil moisture deficit disappeared in the second year after pruning.

<u>Measuring physiological processes which determine leaf water loss in wild cherry trees pruned in April 1999</u>

Leaf water potential (ψ) and conductance (g)

Methods

A Scholander-type pressure chamber (Chas W. Cook & Sons Ltd, Birmingham, UK, or Skye Instruments, Llandidnod Wells, Wales) was used for Ψ. When measuring Ψ, desiccation was prevented by enclosing the leaf in a humidified polythene bag immediately after cutting from the shoot. Abaxial leaf conductance to water vapour (g_l mmol^{-2} s^{-1}) was measured on well exposed mature leaves using a continuous-flow diffusion porometer according to Day (1977). The effects of crown reduction and thinning treatments on leaf water relations were compared with non-pruned trees. Diurnal time courses of leaf water potential and leaf conductance were measured on four occasions between July and September 1999 and on three occasions between July and September 2000.

Results and conclusions

A representative diurnal time course for 29 July 1999 is shown in Figure 1.2. Pre-dawn leaf water potentials (Ψ) early season were high (<0.03 MPa) indicating that none of the trees were expressing signs of experimentally induced drought stress. As the soil dried, Ψ reduced and differences were apparent. The non-pruned trees had the most negative leaf Ψ and crown-reduced trees had the least negative, and the crown-thinned trees were similar to the non-pruned trees. This confirms that soil was drier under the non-pruned and crown-thinned compared to the crown-reduced trees.

At midday, when climatic conditions induce high evaporative demand, transpiration rates are high and leaf water potential falls below soil water potential to maintain transpiration. Under these conditions the crown-reduced trees had less negative Ψ than either of the other treatments, confirming they were experiencing less drought stress. Similar effects were observed for all other midday measurements.

The leaf conductance (i.e. potential of leaves to lose water) increased early in the day to a maximum at midday and then subsequently declined. This pattern is closely linked to the changes in photosynthetically active radiation (PAR). The leaf conductance of the crown-reduced trees was greater than for the non-pruned and crown-thinned trees which were similar. Thus, the crown-thinned and non-pruned trees were regulating their water loss by partially reducing their stomatal apertures. The combination of greater leaf conductance and higher leaf water potential suggests that crown-reduced trees have an increased potential to lose water.

In the following year, carry over effects of pruning treatments on leaf water relations was short lived. A representative daily time-course for the 20 June 2000 is shown in Figure 1.2. Significant differences in pre-dawn leaf water potentials were not detected, suggesting that there were no treatment differences in water availability within the soil profiles. Similar results were obtained for all the other measurement dates. The less negative pre-dawn water potential values (0.25 MPa c.f. <0.5), indicated higher soil water availability in 2000 than in 1999. Furthermore, the less negative midday leaf Ψ (found on other dates as well) also suggest that 2000 was wetter than 1999. The vapour pressure deficit, that drives water vapour loss from leaves was half that in the first year. Thus, evaporative demand was probably much lower on this measurement day.

- Pre-dawn leaf water potential indicated that the roots of crown-reduced trees were experiencing wetter soils than those of crown-thinned and non-pruned trees.
- Measurement of leaf water potential indicated that during the period of maximum evaporative demand from the tree canopy, crown-reduced trees were less drought stressed than non-pruned or crown-thinned trees.
- Crown-reduced trees had greater potential to lose water (leaf conductance) from their leaves than either non-pruned or crown-thinned trees.
- The effects of pruning on leaf water potential and conductance did not persist into the second year after pruning.

Boundary layer conductance

Leaf and canopy boundary layers have been shown to have a significant influence on whole tree transpiration rates (Wullschleger *et al*., 2000, Meinzer, *et al*., 1997). Boundary layers have the capacity to moderate the effect of stomatal aperture on transpiration rates by limiting the rate at which transpired water vapour can diffuse away from the air immediately surrounding the leaf surface into the bulk atmosphere (Wullschleger *et al*., 1998, Meinzer, *et al*., 1997). This diffusive rate limitation can lead to the development of a layer of air surrounding the leaf, which has a higher water vapour pressure than the air of the bulk atmosphere, effectively decoupling the leaf from the major transpirational driving force (Jarvis and McNaughton, 1986). The extent of any decoupling effect will depend more on the ratio of stomatal (g_s) to boundary layer (g_b) conductance than the absolute boundary layer conductance, with a high ratio (high g_s, low g_b) increasing the degree of decoupling and a low ratio (low g_s, high g_b) decreasing the degree of decoupling (Wullschleger *et al*., 1998).

Canopy shape and architecture have been shown to significantly alter canopy boundary layer conductance (Schuepp, 1993). Crown reduction and crown thinning both have profound effects on canopy architecture. Crown reduction leads to the development of dense closed canopies while crown-thinning produces more diffuse open canopies (see Objective 3). For this reason measurements of boundary layer conductance have been conducted throughout the project.

Boundary layer conductance may be measured indirectly via estimates produced using various mathematical relationships (for examples see Meinzer, *et al*., 1997) or directly using custom made sensors (see Daudet *et al*., 1998, and Roberts *et al*., 1990). In 1999 prototype sensors similar to those described in Roberts *et al*. (1990), which combined an artificial leaf component with integrated wet and dry thermocouples, providing measures of both evaporation rate and absolute humidity, were constructed for determination of boundary layer conductance.

A modified artificial leaf was developed in spring 2000 specifically for this work and used to determine the rate of water loss, and hence boundary layer conductance. The artificial leaf consisted of a reservoir of water that supplied, by capillary action, a black wicked evaporative surface (the 'artificial leaf') of about 5cm^3. Each sensor was supplied with two thermocouples for the accurate measurement of temperature; the first at the 'artificial leaf' (the wet or evaporative temperature) and the second the dry or surrounding air temperature. The weight of the whole device (including the water reservoir) was used to determine the mass of water lost from the artificial leaf over time by frequent weighing. From the measured weight of water loss the rate of evaporation from the 'artificial leaf' can be calculated. The change in weight of the artificial leaf, combined with the temperature depression (due to evaporation) of the wet surface of the 'artificial leaf' compared to the dry air temperature, was used to estimate leaf boundary layer conductance.

The artificial leaf was redesigned in 2001 to improve its ability to estimate boundary layer conductance. The new artificial leaf sensor had leaves that were similar in shape to natural cherry leaves as leaf area and shape can influence boundary layer conductance. In addition, they had an integral light sensor for determining radiation.

Measurement of boundary layer conductance in wild cherry trees pruned in April 1999

Methods

Boundary layer conductance within the canopies of the different treatments was quantified in July 1999. Four sensors were placed in the canopy of an individual tree from each treatment. Sensors were arranged within the north side of the canopy at a height of 3 m. Results of this preliminary investigation suggested that crown reduction decreased boundary layer conductance while crown thinning increased boundary layer conductance when compared to the non-pruned control.

Results and conclusions

- Sensors were successfully developed to quantify boundary layer conductance within the canopy.
- Initial results suggest that both crown-reduction and crown-thinning alter canopy boundary layer conductance.
- Lower boundary layer conductance in the canopy of the crown-reduced tree suggests that canopy water loss may be diminished by this pruning treatment.

Instantaneous measurements of leaf assimilation rate (photosynthesis) and water use in wild cherry trees pruned in April 1999

Methods

A Ciras 1 combined infra-red gas analyser and porometer (PP Systems, Hitchin, Herts) was used with a broad leaf chamber (2cm ×6cm) to determine instantaneous gas exchange characteristics. It measures CO_2 assimilation rate, (A, µmol m^{-2} s^{-1}), transpiration, (E, mmol m^{-2} s^{-1}) and stomatal conductance, g_s, (mmol m^{-2} s^{-1}) allowing derivation of external: internal partial pressures of CO_2 (c_i/c_a) and instantaneous water use efficiency (A/E). Well-exposed fully expanded leaves third/fourth from the shoot tip were sampled. Assimilation rates were measured on 27 July and 9 September 1999.

Results and conclusions

On both sampling dates in July and September, the crown-reduced trees had consistently higher rates of CO_2 assimilation and stomatal conductance than either non-pruned or crown-thinned trees (Figure 1.3). Instantaneous water use efficiency (A/E) was similar for all treatments on both sampling dates.

- Individual leaves of the crown-reduced trees had higher rates of assimilation and stomatal conductance than leaves on trees of either the crown-thinned or non-pruned trees. (This contradicts expectations from the wetter soil found under the crown-reduced trees.)

Utilising stable isotopes to determine long-term and instantaneous measurements of leaf water use and assimilation

Background

Stable isotopic discrimination of carbon ($^{13}C/^{12}C$) and water ($^{18}O/^{16}O$) provide markers of photosynthetic carbon gain and water loss, which can either be measured in leaf organic material or from a comparison of soil, leaf and twig water signal. Traditionally, discrimination against $^{13}CO_2$ has also been used to infer leaf water use, since both transpiration and isotope exchange are independently related to the stomatal aperture, via the ratio of internal: external CO_2 concentrations across the leaf (c_i/c_a). Thus, a higher leaf conductance to water vapour is normally associated with higher discrimination against $^{13}CO_2$, with leaves containing a smaller proportion of the heavy isotope (i.e. depleted) (Farquhar *et al*. 1989; Griffiths *et al*. 1999). The heavy isotope of water ($H_2^{18}O$) evaporates less readily than the normal isotope, leaving the leaf enriched in ^{18}O as a function of the evapotranspiration rate, leaf temperature and atmospheric humidity (Farquhar *et al*. 1998).

However, we found that the crown-reduction treatment caused distinct boundary layer conductance effects within the canopy which were unexpected, and allowed the combined use of ^{13}C and ^{18}O to provide an innovative diagnosis for water use and coupling within canopies. Analyses were therefore undertaken of organic material isotope signals for ^{13}C and ^{18}O, as well as that of leaf water ^{18}O.

Isotopic analysis and leaf N content in organic material of wild cherry trees pruned in April 1999

Methods

Plant material was finely ground in a ball mill and samples stored in glass vials to prevent static. For ^{13}C, 1 mg of each sample was weighed into a tin cup, which was then crimped with tweezers and stored in a micro-titre plate. Samples were analysed following combustion in a C:N analyser, with the CO_2 interfaced with a Europa 20/20 continuous flow mass spectrometer either at the Newcastle-upon-Tyne stable isotope facility or by Dr C Scrimgeour at Scottish Crop Research Institute, Dundee. This procedure also provided the total N content of leaf samples, an additional physiological marker not included in the original proposal. Samples for ^{18}O in leaf material were analysed by pyrolysis at 1300 ^{o}C to CO, at the stable isotope facility, Research School of Biological Sciences, Australian National University, Canberra. Whilst data are measured as an isotope ratio ($\delta^{13}C$) with the mass spectrometer, it is more practical to express data as discrimination (Δ), which reflects the fractionation (partitioning) during the biological transformation from source (CO_2 in air) to organic material. Discrimination is therefore a positive term, with high values associated with high discrimination against ^{13}C, when leaf material is relatively depleted in ^{13}C. The physiological range of discrimination in C3 plants, such as the trees under this investigation, is likely to be shifts of between 1-3 parts per thousand (‰).

Results and conclusions

Carbon isotope discrimination was higher in the crown-reduced trees by 1.2 ‰, compared to non-pruned and crown-thinned trees in 1999 (Figure 1.4). This was in agreement with the measured higher transpiration rates and stomatal conductance (Figures 1.2 and 1.3), and confirms the potential for greater water loss was from individual leaves within the canopies of crown-reduced trees. This contrasted with the soil moisture deficit data that indicated that the soil remained wetter under the crown-reduced trees (Figure 1.1). The difference in carbon isotope discrimination between treatments was lost in the second year following pruning (Figure 1.4), when a trend towards higher discrimination was then found in non-pruned and crown-thinned trees. For all trees, carbon isotope discrimination was some 2‰ higher in the second year, suggesting higher water use overall in 2000 as compared to the drier summer of 1999 (see objective 4).

- Carbon isotope discrimination implied that potential water use by individual leaves in the crown-reduced trees was higher than that from non-pruned or crown-thinned trees in the first year after pruning.
- Differential effects of pruning treatments on leaf water use were lost in the second year after pruning since similar values of carbon isotope discrimination were found for all treatments.
- Higher overall values of carbon isotope discrimination in the second year suggested that the trees in all treatments used more water in the second year after pruning.

Combining ^{13}C and ^{18}O analysis of leaves to elucidate a new way of determining water use patterns

Background

During the development of the proposal, the use of additional measurements of leaf water ^{18}O content became appropriate, to act as a proxy for evaporative enrichment in individual leaves (Farquhar et al 1998). Once water is transferred from twig to the leaves, the rate of evaporation leads to additional fractionation during transpiration, which, if dependent on humidity, is proportional to the extent of ^{18}O enrichment in leaf-water. As mentioned above, and subsequently found in many of the experiments, the leaf ^{13}C data could not be always directly related to overall tree water use, whether determined from soil moisture deficit or daily sap flux. Carbon isotope discrimination in organic material should provide a season-long integration of leaf water use. Therefore, in subsequent experiments we investigated the discrepancy between the high leaf conductance (Figure 1.2), water use and carbon isotope discrimination (Figure 1.3) in the leaves

of trees in the crown-reduced treatment, in contrast to the higher overall water use by non-pruned and crown-thinned trees (Figures 1.1, 1.5).

Evaluating and developing the means to measure daily whole tree water use using sap flow

Background

Sap flow can be determined by several methods including use of heat balance, heat pulse and thermal dissipation techniques (Smith and Allen, 1996). In this project, we used Granier-type Thermal Dissipation Probe (TDP) sets (TDP-30, Dynamax, Houston, USA) supplied by Delta-T Devices (Cambridge, UK) for our measurements except on Experiment 5 (see objective 3) where we also used heat pulse sensors (Greenspan Technology Pty Ltd, Queensland, Australia).

Each TDP set comprised of two metal probes containing fine thermocouples. The upper probe in each pair when installed contained a heater element. When inserted the heat from the heated probe is dissipated by moving sap. Thermal dissipation is more effective at higher rates of sap flow, and thus the difference in temperature between the upper and lower probes declines as the velocity of sap movement increases. Measured temperature differences between probes in a TDP set are related empirically to sap velocity, which when integrated over cross-sectional sapwood area (tissue conducting water) gives the flow rate (Granier, 1985; Smith and Allen, 1996). Installation in trunks below tree crowns enables measurement of whole tree transpiration (water use). TDP sets 30 mm length, 1.2 mm outside diameter were used in the field experiments.

Heat dissipation probe calibration

An initial experiment was conducted over 5 days to determine the accuracy of the TDP sets. A potted (25 l) cherry tree in leaf was placed on a balance in a glasshouse controlled environment cabinet and its water use was determined by automatically recording weight loss over 10 min intervals. A set of thermal dissipation probes (30 mm) were inserted into the trunk and the sap flow was determined over the same 10 min intervals as the balance. There was a strong linear relationship between the sap flux determined using the two methods. Thus sap flux (TDP) = 0.79 $\times$ sap flux (gravimentric), $r^2 = 0.91$. The underestimate of water loss relative to the balance was probably due in part to the fact that the tree continued to lose water during darkness and this did not make it possible to set up the probes with the required zero flow rate. This is unlikely to be a problem for trees in the field. Thus, the empirical calibration of Granier (1985) was used for the experiments in this project

Initially, the Dynamax TDP-30 sets were found to be unreliable for two reasons. Occasionally, anomalous data indicated sap flow before dawn which was unlikely. This was partly reduced by increasing the level of thermal insulation around the sensors and thus reducing thermal gradients. The second more serious problem was that the probes were very fragile and easily broken on removal from the trunk.

A new thermal dissipation probe was designed and built by the Technical Services Department, HRI, Wellesbourne. This was based on the original 30 mm long, 2 mm outside diameter probe design of Granier (1985), but the constantan heater element was substituted by five 10 Ω (surface mount) resistors connected in series close to each other. These sensors were very reliable and were more robust than the Dynamax probes. Since these probes followed closely to the design of Granier, no further calibration was necessary.

Measurement of sapwood area

In order to calculate daily water use (sap flux) the velocity of the sap (measured by the thermal dissipation or heat pulse probes) is multiplied by the area of sap conducting tissue (xylem) in the

trunk. In many species (including cherry), sap wood and heartwood can be fairly easily distinguished visually by colour.

The proportion of sap to heartwood was estimated from the cut ends of twenty logs of individual cherry trees of similar girth to the experimental trees. The logs varied in length from 1.67 – 3.67 m. The average diameter of sapwood and heartwood at the base and top of each log was recorded and related to the stem girth (circumference) at the same points. The following linear relationship was found; Sap wood area (cm^2) = trunk girth (cm) × 6.17 – 173 (r^2 = 0.94, n= 35). Subsequently, this relationship was used to convert trunk girth to sap wood area for all of the experiments using cherry trees.

Initial visual studies on London plane trees found it very difficult to determine the extent of heartwood on logs. An alternative method was therefore used. A 5 mm increment core was taken from the trunk of each experimental tree at the height that the thermal dissipation or heat pulse probes were inserted. The core was stored in a sealable plastic bag dipping in iodine-potassium iodide solution (2.5%). After a few seconds the starch in the sapwood (living tissue) stained black. The depth of the sapwood below the bark was measured and used to calculate sapwood area from the trunk diameter and bark thickness. Surprisingly, the apparent heartwood (non-living tissue) formed less than 1% of the conducting tissue in these trees, i.e. 99% of the tissue in the trunk below the bark was live and potentially capable of conducting water.

Sap flux (tree daily water use) in wild cherry trees pruned in April 1999

Methods

Twenty-four 30 mm TDP sets (Dynamax, Huston, USA) were inserted into 12 trees (i.e. two per tree and three tree per treatment). The probes were placed in a North and West aspect of the trunk approximately 140 cm above ground level. The probes were connected to two data loggers and measurements were collected every minute and integrated every ten minutes for each probe. Measurements were made during four periods, ranging from 3-23 days during July to September. Twenty-four TDP sets were used again in May and August 2000 to measure the residual effects of pruning from the previous year.

Results and conclusions

Daily water use (sap flux) averaged across all trees in all treatments was 28 l^{-1} d^{-1} tree^{-1} in July and August (Figure 1.5), but it dropped to 18 l^{-1} d^{-1} tree^{-1} in September and 12 l^{-1} d^{-1} tree^{-1} in October. Analysis of variance indicated that standard error of differences between the means were large compared to the means and differences between treatments were not significant (P>0.05). The lower daily sap fluxes later in the growing season were due to the cooler, wetter weather conditions.

In May 2000, the mean sap flux measured over three days varied between approximately 16 and 24 l^{-1} d^{-1} tree^{-1}. The control trees had the greatest flux and the reduced and thinned trees transpired less, but these differences were not statistically significant. In August, the mean sap flux measured over 26 days varied between 11 and 40 l^{-1} d^{-1} tree^{-1}, but again differences were not significant.

- The thermal dissipation probes were sensitive to differences in sap flux caused by changes in prevailing weather conditions.
- No apparent differences in daily sap flux caused by the pruning treatments were detected by thermal dissipation probes in the year of pruning or in the following year.

Developing a method to measure tree canopy leaf area

Background

Two different commercially available systems were investigated to measure total canopy leaf area and leaf configuration of trees: Analysis of digital hemispherical photographs, (HemiView 2.1, Delta-T Devices Ltd, UK), and the Accupar sun ceptometer (Decagon Devices Inc., Washington, USA).

The sun ceptometer measures the fraction of transmitted PAR wavelength that passes through the canopy and from this the gap fraction and the LAI can be calculated. Hemispherical photographs taken from the ground looking up at the canopy provide a record of the canopy. They are analysed to compute solar regime and canopy characteristics such as Leaf Area Index (LAI) by determining the gap fraction for different zenithal angles simultaneously, through a fish-eye lens. Canopy parameters are calculated according to Norman and Campbell (1989). The principles and history of hemispherical photography are described briefly in Rich *et al*. (1999). Ground-based measurements of leaf area index including commercial canopy analysers have been reviewed recently by Breda (2003), Jonnckheere *et al.* (2004) and Weiss (2004). For this project, Delta-T devices developed a new model that allowed analysis of single isolated trees and enabled trunk and major branches to be excluded from the analysis (J Wood, personal communication, 1999).

Calibration of HemiView

To establish the accuracy of the two indirect methods of measuring leaf area, it was necessary to determine canopy leaf area empirically by another independent method. This was achieved by relating branch total leaf area to branch basal girth of two non-experimental cherry trees adjacent to experiment 1. Twenty-two sample branches with a wide range of sizes were removed from the base to the top of the crowns and each branch basal girth was measured. The leaves from these branches were removed, counted and their area was determined. The following quadratic relationship was found; Leaf area $(m^2) = 0.0088\,x^2 + 0.1061x$, $r^2 = 0.95$, where x = branch girth (cm), r = correlation coefficient, n = 21.

The basal girth of every branch that intercepted the trunk was measured on each non-pruned tree in experiment 1 (see below). The total leaf area of each tree was calculated by converting the branch basal girth to a branch leaf area using the equation given above. The leaf areas for each branch were then added together to give a total for each tree. The relationship between the values of leaf areas based on branch sampling and the leaf areas of the non-pruned trees calculated by HemiView was linear, but the latter underestimated measured leaf area compared to the former (Figure 1.6).

Comparison of HemiView and Accupar

Values for drip line leaf area index (leaf area per unit area of ground covered by the crown) from Accupar were calculated from light interception data collected on 30 July 1999. The Accupar underestimated the leaf area compared to HemiView by a factor of about 2 (data not presented). Another study (Peper and McPherson, 1998) has reported a similar problem with sun ceptometer measurements of isolated tree canopies. Nevertheless, Accupar was capable of picking out the differences between the canopy manipulation treatments in terms of light interception, notably the low leaf area index of crown thinned trees. Because use of Accupar data does not remove the necessity of taking canopy volume measurements, it was evident that the HemiView technique provided the optimal compromise between speed of measurement, maintenance of records and accuracy, provided the pictures were taken under appropriate lighting conditions.

Canopy leaf area of wild cherry trees pruned in April 1995

Methods

Digital hemispherical images were collected under the canopy of every tree on 19 July and 20 August 1999, 11 May, 27 June and 26 July 2000 with a fisheye lens. The data were analysed with the HemiView 2.1 digital analysis system.

Two models were used to represent the 'volumetric shape' of the canopy. A half-ellipsoid model was used for the crown thinned and control trees, whereas a cylindrical model was used for the crown reduced trees. These models were chosen because they provided the best descriptions of the crowns. The volume of the canopy of the crown-reduced trees was 53% of that of the non-pruned trees, whereas that of the crown-thinned trees was 92%.

Results and conclusions

Crown thinned trees had significantly ($P<0.05$) lower total canopy leaf areas than the non-pruned trees in August (Figure 1.7). Although, the crown-reduced had lower leaf areas than the non-pruned trees the difference was not significant. The architecture of the canopy was also changed by the method of pruning. Leaf area density (leaf area per unit crown volume) and drip line leaf area index (leaf area per unit of ground drip line area) were significantly ($P<0.001$) different between the treatments. Both parameters were greatest in the crown-reduced trees, with non-pruned trees intermediate and crown-thinned trees the least. Thus, the crown-reduced trees had recovered by producing leaves in a more closely packed, denser canopy.

In the following year (2000), the non-pruned trees were close to full canopy in May and only a small further increase in total leaf area occurred by July. The pruned trees had larger proportional increases in leaf area between May and July, suggesting that each pruning treatment delayed the rate at which subsequent canopy leaf area development occurred. Fifteen months after application of the treatments (July 2000) the crown-thinned trees had significantly ($P<0.05$) less canopy leaf area than those that had received either of the other two treatments. The total canopy leaf area of the crown-reduced trees had recovered to levels similar to the non-pruned trees.

The architecture of the canopies remained different as the leaf area densities of crown-reduced and crown-thinned trees remained approximately double and half those of the non-pruned trees in July. These differences were significant ($P<0.001$). The greater leaf area density (and drip line leaf area index) of crown reduced trees between May and June indicated that, as the canopy developed during the growing season more self shading of leaves occurred which led to more compact canopies than those of the crown thinned or non-pruned trees. The crown-thinning did not influence shoot growing of the tree and therefore the canopies took longer to recover and remained more open.

- The modified hemispherical photographic image technique adapted for single trees use was shown to provide measurements of total canopy leaf area closely related to those determined using an allometric method based on summation of leaf areas on individual branches.
- Crown-reduced trees completely recovered their full canopy leaf areas within two years of pruning.
- Crown thinned trees took more than two years to recover their total canopy leaf area.
- Method of pruning had a large impact on the architecture of the canopy. Crown-reduction produced a compact, more dense canopy, whereas crown-thinning had the opposite effect.

Figure 1.1 The effect of crown-reduction and crown-thinning on soil moisture deficit and minimum soil moisture content in 1999 and 2000 of wild cherry trees pruned in April 1999 (Experiment 1). Vertical and horizontal bars are standard errors of differences between means. Monthly rainfall for the same period.
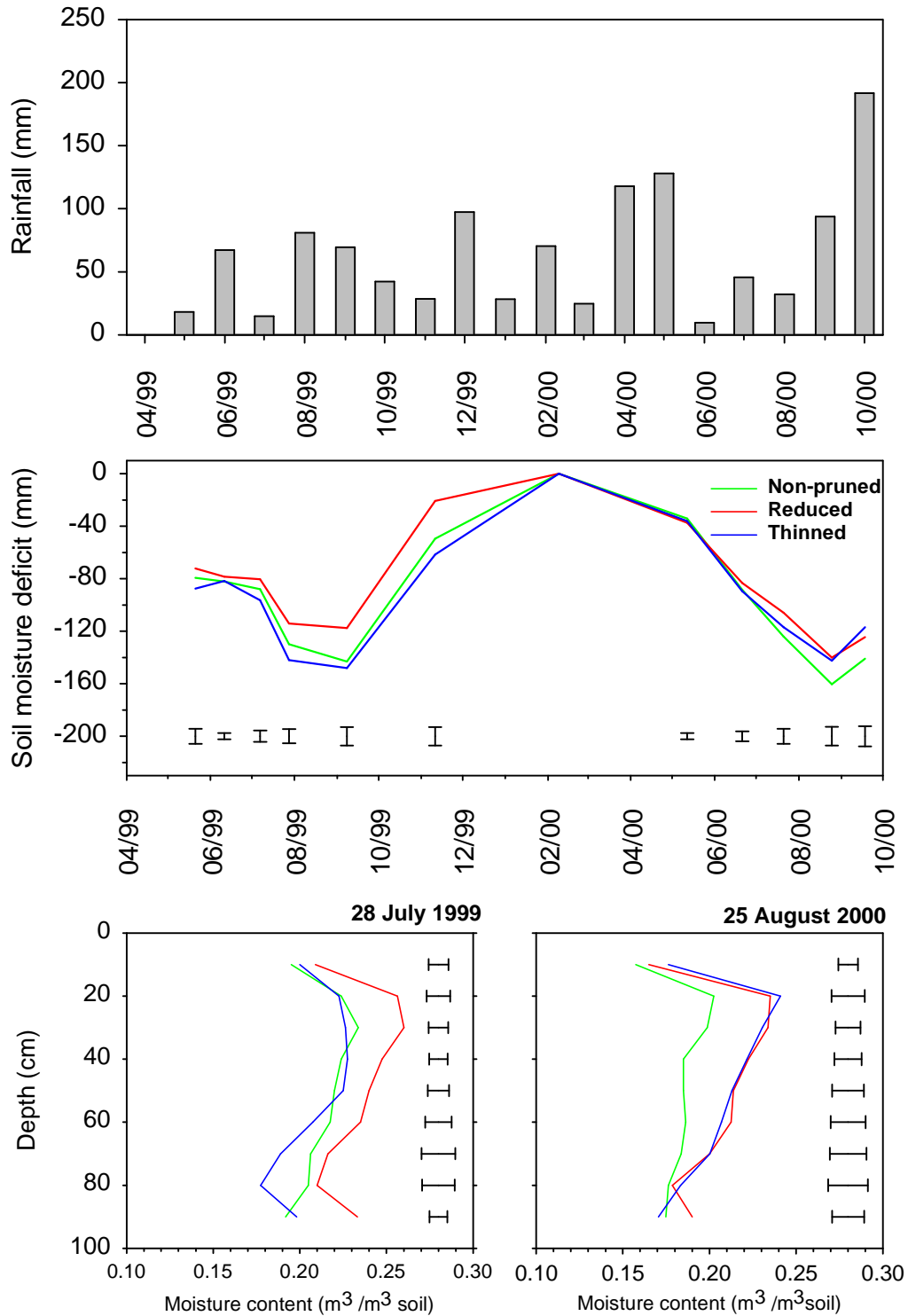
Figure 1.2: Diurnal cycle of leaf water potential (Ψ), leaf conductance (g_L) and light levels (photosynthetic active radiation, PAR) on wild cherry trees pruned in April 1999 (Experiment 1). Vertical bars are standard errors of differences between means
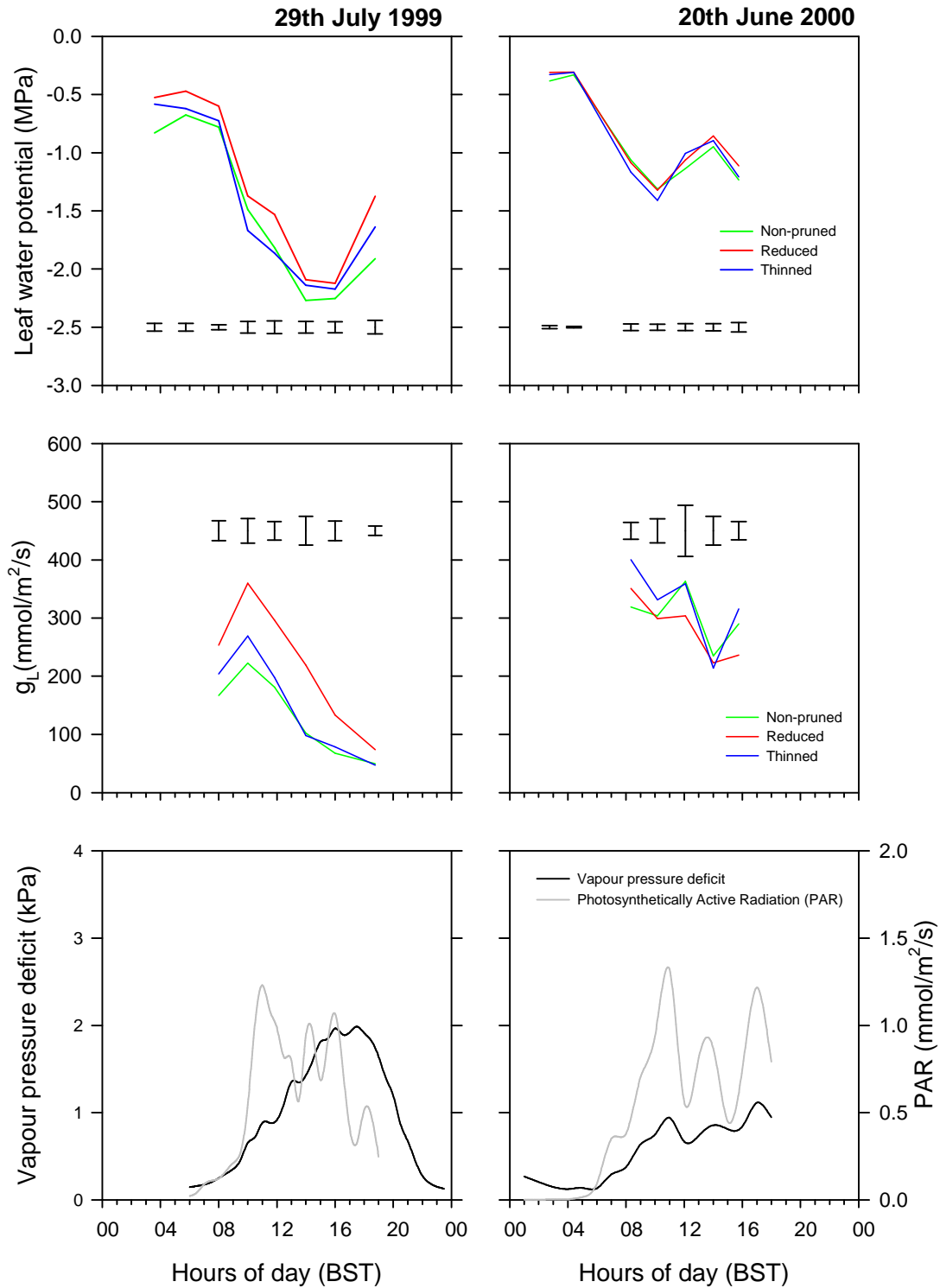
Figure 1.3: Assimilation rate, stomatal conductance and evaporation rate from instantaneous leaf gas exchange measurements made on wild cherry trees pruned in April 1999 (Experiment 1). Measurements made using ambient light and CO2 concentrations on the 29 July and 9 September. Vertical bars are the standard error of each mean, analysed individually
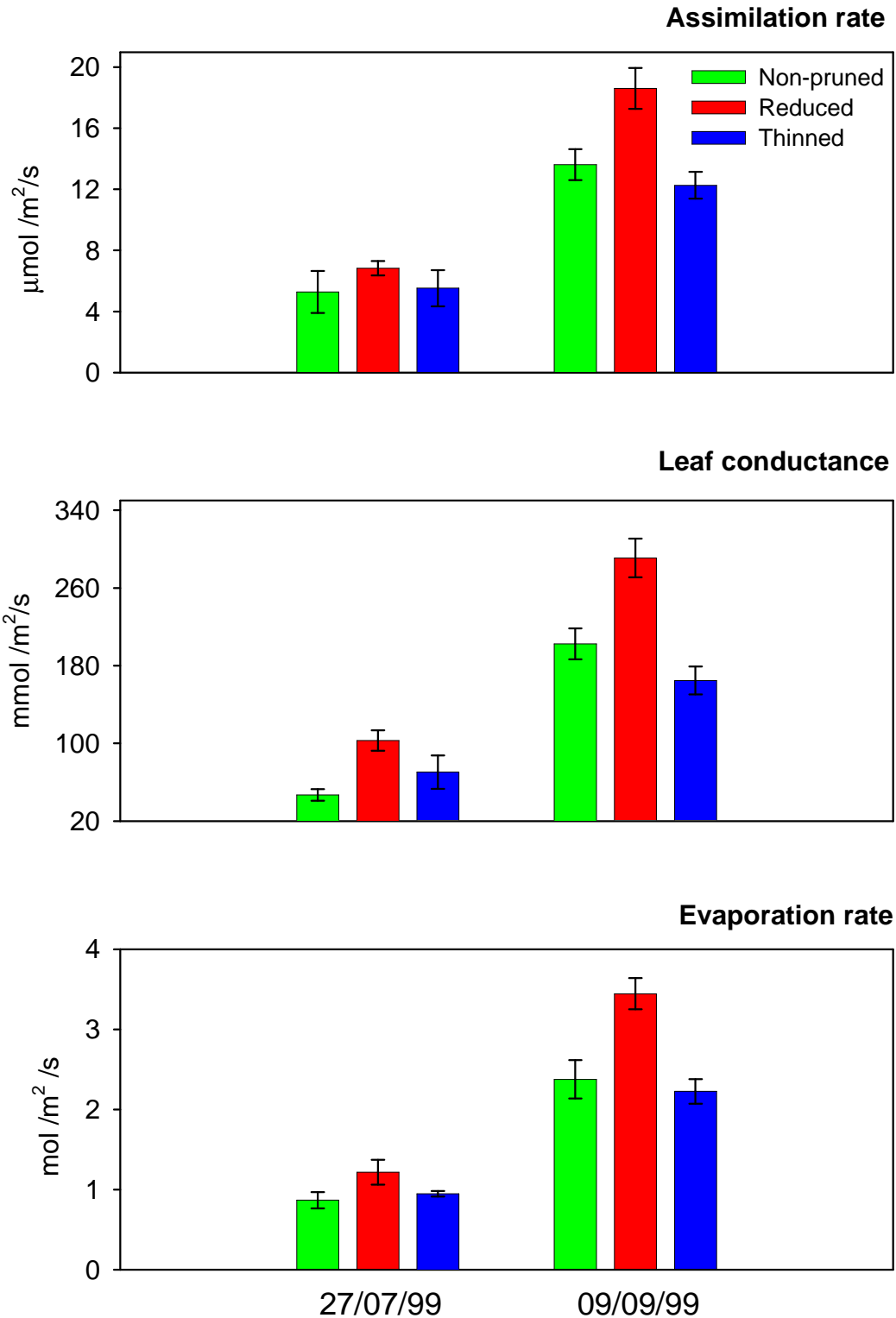
Figure 1.4: Carbon isotope discrimination (Δ ^{13}C) in bulk leaf matter from wild cherry trees pruned in April 1999 (Experiment 1). Samples were collected 21 August 1999 and 10 September 2000. Vertical bars are standard error of each mean, analysed individually
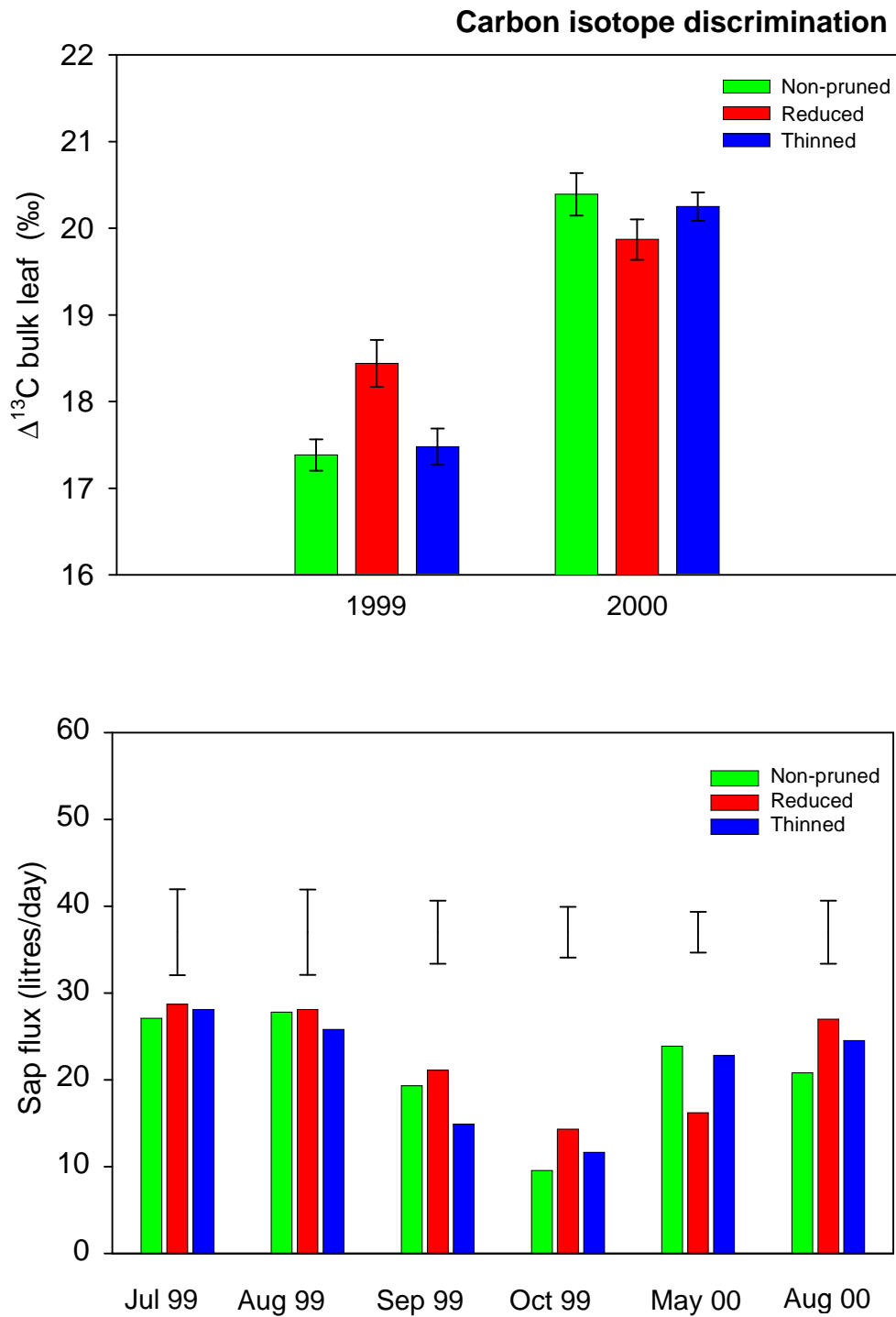
Figure 1.6: A comparison of hemispherical photographic and allometric determinations of canopy leaf area on non-pruned trees
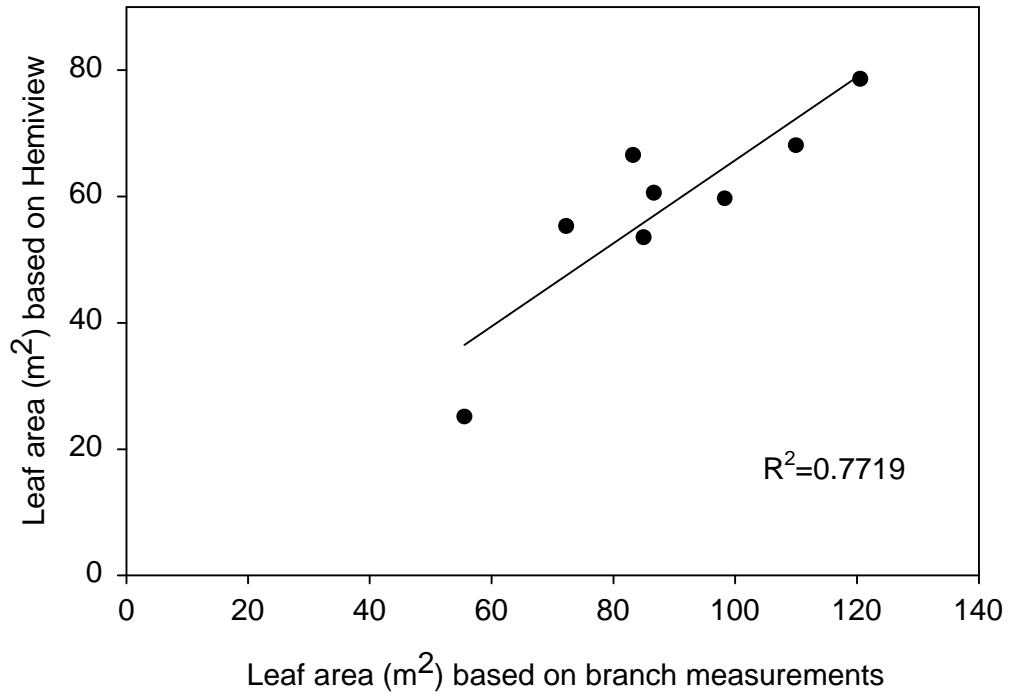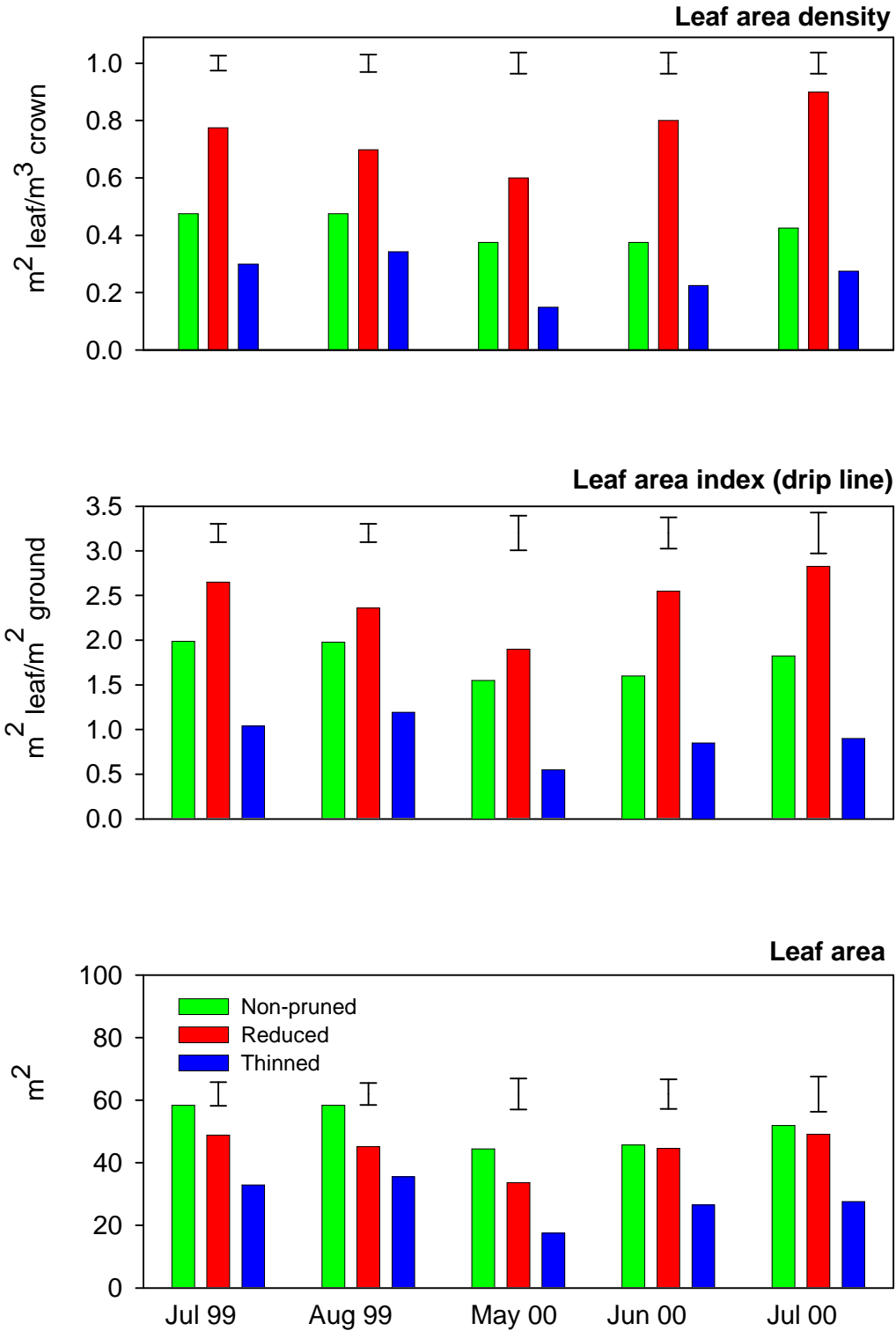
Figure 1.7: Leaf area density and total canopy leaf area for wild cherry trees pruned in April 1999 during 1999 and 2000 (Experiment 1). Vertical bars are standard error of differences between means

Objective 2:

Identify the main sources of water within the soil profile for tree water extraction and the influence of species and season

Using stable isotopes of oxygen to determine tree root water sources in wild cherry trees

Rationale

The rationale for this approach came from the success in identifying water sources for semi-arid shrubs, grasses and trees, (Ehleringer *et al.,* 1999), and the notable characterisation of hydraulic lift (Jackson *et al.,* 2000).

In regions where seasonality is pronounced, temperature effects during water evaporation and condensation lead to fractionation against ^{18}O (relative to ^{16}O), so that distinct signals occur in summer and winter rains or snowfall, which affect the recharge of groundwater. The soil water profile ^{18}O may reflect these inputs, and initially, it was proposed that only the ^{18}O signal in soil and stem water should be analysed, provided that a gradient in ^{18}O signal exists between upper and lower soil horizons. The method relies on the absence of fractionation during water uptake by roots. Therefore, twig water should indicate the source of water within the soil profile.

Methods

Samples of soil were collected at 20 cm depths from within the soil profile by placing 2-3 g of soil in 12 ml volume breath test vials ("Exetainers"^{TM}), with screw cap and viton rubber septa to prevent evaporation, for storage until subsequent extraction and analysis of ^{18}O composition. Samples of twigs were collected, 3-4 cm long and 3-5 mm in diameter and also stored in Exetainers, for extraction of unfractionated source water in the stem. An initial survey of soil samples from shallow (10-20 cm) and deep (60-70 cm) depths for the three crown pruning treatments was compared with twig water. Water was distilled under vacuum from each Exetainer, using liquid nitrogen, in a procedure taking 3-4 hours for each soil or twig sample. An aliquot of the water (usually 0.5 ml) was then equilibrated in a sealed glass vial for a minimum of 3 days with additional gaseous CO_2, allowing the ^{18}O signal of the water sample to be transferred to the CO_2 by isotopic equilibration. Each vial was then broken within a vacuum preparation line, with the sample of CO_2 then trapped cryogenically and purified prior to analysis with an isotope ratio mass spectrometer. During the course of the study, an investigation was undertaken to see whether direct equilibration CO_2 with soil, twig and leaf water could improve the rate of sample analysis, but absolute ^{18}O composition and precision was found to be compromised by this procedure, and so the lengthy sample extraction and analytical protocol was utilised throughout.

Results and conclusions

The results for the ^{18}O signal of soil profiles and associated twigs for the wild cherry trees in experiments 1 and 2 are illustrated with data collected in early September 1999 and 2000 (Figure 2.1), i.e. in bolt cases during the year after pruning. In 1999, the drier year, there was on average a 2 ‰ difference between soil water at 10-20 cm and 60-70 cm, with the surface water more enriched in ^{18}O ($\delta^{18}O$ less negative) because of evaporative enrichment and/or shifts in summer rainfall isotopic signature (Figure 2.1). However, despite limited replication, we may infer that the water used by all treatments is taken from deep in the soil profile. Thus, the ^{18}O signal extracted from the twigs is closer to that found deep in the soil profile in all three crown pruning treatments. In 2000, a cooler and wetter year than 1999, it was not possible to distinguish water sources within the soil profile (Figure 2.1) with data generally being more variable. This approach was not deemed to be worth pursuing in the light of subsequent developments of ^{18}O determination in leaf organic material and bulk water outlined below. Additional soil samples were collected in subsequent years in order to determine the variability in these signals, which probably reflect the

temperate climate of the UK where seasonal climatic differences do not generate consistent differences between groundwater and rainfall water isotope signals.

During subsequent experiments on cherry and London Plane we extended the sampling protocols to include the routine isotopic analyses of leaf water, as well as some determinations of ^{18}O in leaf organic material. These approaches were subsequently adopted by the consortium as an alternative to the soil profile component of the study.

- In a dry year, water at 60 cm soil depth is depleted in ^{18}O by 1 to 2 ‰ as compared to the evaporative enrichment seen at 15 cm depth.
- Twig water generally reflects the use of soil water from deeper within the profile.
- In a cooler, wetter year (2000 c.f. 1999) the isotopic profiles were smaller and capacity to distinguish water sources was less successful.

Long term impact of crown pruning and environment on tree water use (Experiment 2)

Crown-reduction and crown-thinning applied to wild cherry trees in 2000 and repeated in 2002

Soil drying by trees

Methods

Neutron probe access tubes were inserted to 1 m depth adjacent to one tree in each plot in Spring 2000. During August 2001, an additional access tube was placed 3 m from one tree in each plot. To allow determination of the effects of the second (2002) pruning treatments and allow comparisons with the trees pruned only once, more access tubes were inserted in April 2002. Measurements were taken at 10 cm intervals to a depth of 90 cm. All soil moisture deficits were referenced to field capacity on 16 December 2002.

Results and conclusions

In their year of application (2000) neither pruning treatment had a significant (P>0.34) effect on soil drying between June and September (Figure 2.2), although the soil under crown-reduced trees had less moisture deficit. In the following year (2001), there were no treatment differences in soil moisture. The soil was significantly (P<0.01) drier at 1 m than at 3 m from the trees, but there was no interaction between proximity to tree and pruning treatment, implying the (lack of) effect of pruning was consistent across a distance 3 m from the trunk.

In 2002, crown-reduction and crown-thinning applied in 2000 continued to have no effect on soil moisture at 1 m or 3 m, from the tree, i.e. differences between the treatments were not significant (P>0.05), confirming the results of the previous year. Surprisingly, the repeat crown-reduction and crown-thinning treatments applied in spring 2002 also had no effect on soil moisture content compared to the non-pruned trees (Figure 2.2). The soil moisture deficit developed during the growing season, at 1 m from the tree remained approximately double that found than at 3 m from the tree. Thus, the largest differences in soil moisture deficit again were related to proximity to the tree, rather than the pruning treatments applied in either 2000 or 2002.

The rainfall in 2003 was one third less than average and the soil moisture deficits were much greater for all treatments than in previous years. The soil moisture deficit increased more rapidly nearer to the tree and reached a maximum in late August (Figure 2.2). Thus, during June and July, the soil remained significantly (P<0.001) drier nearer the tree, but by August the difference in moisture deficit between the near and far positions from the tree had declined. This implies that as the season progressed, the water reserves near to the trees were exhausted and the trees roots accessed soil water further away from their trunk. When averaged across position from the tree,

none of the pruning treatments differed significantly (*P*>0.05) from the non-pruned (control) trees in their effects on soil drying.

- Irrespective of pruning treatment the greatest soil drying caused by the roots of the trees was localised at close proximity to the trunk.
- Crown-reduction and crown-thinning had negligible effects on soil drying compared to non-pruned trees up to 3 m distance from the trunk.
- Abstraction of water from the soil by tree roots becomes more distal from the trunk as the season progresses.

Impact of drought on response to crown pruning (Experiment 4)

Crown-reduction and crown-thinning applied to wild cherry trees in 2002 and soil drought applied in 2002 and 2003.

Soil drying by trees

Methods

Neutron probe access tubes were inserted at a distance of 1 m from every tree. Soil moisture was determined approximately monthly from May 2002. All soil moisture deficits were referenced to field capacity on 6 November 2003

Results and conclusions

The rain covers were very effective at excluding rainwater from close to the tree (Figure 2.3). The soil under the covers reached a maximum deficit of 115 mm in September, whereas the non-covered soil reached a maximum deficit of only 50 mm in July. In fact, the non-covered soil remained relatively moist i.e. near field capacity for much of the season. The covers caused the soil to be significantly (*P*<0.01) drier in the 10-90 cm horizon than the non-covered soil. These differences in soil moisture varied between 0.03 and 0.17 m^3 water/m^3 soil at individual depths and reached a maximum late summer. The severity of the drought also progressed further down the profile later in the growing season.

Until August, soil under the drought stressed crown-reduced and crown-thinned trees remained slightly wetter than under the control trees, resulting in soil moisture deficits that were approximately 10-20 mm less than for the control trees (Figure 2.3). However, these effects of pruning were not statistically significant (*P*>0.05).

In the following year (2003), the soil moisture deficit under all trees increased until September when it reached a maximum (Figure 2.3). The rain covers were very effective in inducing an early drought (c.f. the late drought in the previous year) and gradually increasing it throughout the season. In April, the soil moisture deficit under the droughted trees was approximately double that under the non-droughted trees and by May this had increased to approximately four times. Subsequently, due to the dry climatic conditions in this year, the absolute differences between the droughted and non-droughted soil declined until late September, when they became similar (Figure 2.3). The soil moisture deficit increased most rapidly during the period June to September for the non-drought stressed trees. The crown-reduction and crown-thinning treatments applied in 2002 had no effect (*P*>0.05) on soil drying.

Differences due to rain covers occurred mainly nearer the surface. Thus, in April and May the soil remained drier in the 10-60 cm depth zone than the under the non-droughted trees, but no differences occurred in the profile below. By July, the whole soil profile had become drier and the differences occurred only at 10-30 cm, subsequently they disappeared. No differences in soil

moisture at specific depths were found between the non-pruned trees and the trees either crown-reduced or crown-thinned (data not presented).

- The pruning treatments had only an insignificant effect on conserving soil moisture in the year of pruning or subsequently.
- Crown-reduction and crown-thinning did not influence the soil moisture conservation under droughted trees.

Assessing the impact of crown pruning on large amenity trees (Experiment 5)

Crown-reduction and crown thinning applied to London plane trees in 2000 and severe crown reduction applied in 2003

Soil drying by trees

Methods

Two access tubes per plot were inserted to 1 m depth in spring 2000. One tube was 2m from the tree and the other was mid-way between the two adjacent trees in an experimental plot i.e. 6 m from each tree. Extra access tubes were inserted at a distance of 2 m from each tree in spring 2002, such that the experiment contained a total of 54. The neutron probe was used to measure soil moisture at 1-2 month intervals. All soil moisture deficits were referenced to field capacity on 30 November 2000, except in 2003, which were referenced to 7 January 2004.

Results and conclusions

The soil moisture deficit increased rapidly in June and July 2000 reaching a maximum in August (Figure 2.4). The soil nearer the trees remained significantly ($P<0.001-<0.05$) drier than that further away until September. The pruning treatments had significant ($P = 0.002 – 0.055$) effects on soil moisture deficit during the period July to September. Soil under the pruned trees remained wetter than that that under the non-pruned trees. This effect was particularly marked at 6 m distance from the trees (Figure 2.4), where the difference between treatments was 40 mm, suggesting that the crown-reduction and crown-thinning treatments were most effective at decreasing soil drying further away from the tree. Either pruning method was equally effective at conserving soil moisture. The differences in soil moisture deficit between the pruned and the non-pruned trees were due to soil drying throughout the profile (Figure 2.5).

Following recharging with water from winter rainfall, the moisture content was uniform throughout the profile in May 2001 and the soil under all trees remained near field capacity (Figure 2.4). By July, water from the profile to 1 m depth was declining and the deficits increased to their maximum in August (Figures 2.4, 2.5). When the effects of pruning were averaged across distance from tree, crown-reduction and crown-thinning significantly ($P<0.001 – 0.05$) enhanced the soil moisture reducing the deficit by 20-30 mm compared to the non-pruned trees in August 2001. Subsequently, the soil moisture under all trees increased, although the soil under non-pruned trees remained drier and never recovered to field capacity during the winter. Low winter rainfall (Figure 2.4), coincided with the soil under the non-pruned trees not fully recharging with water below 60 cm depth. Crown-reduction was more effective in allowing soil recharge than crown-thinning at 6 m from the trunk, but nearer to the tree there was no difference between either pruning treatments.

In the third growing season after pruning (2002), differences in the soil moisture deficit (Figure 2.4) between the non-pruned, crown-reduced and crown-thinned trees were small, but significant ($P<0.05$) between August and November, i.e. during the period of maximum soil moisture deficit. The soil remained 10-20 mm wetter under the pruned trees than the non-pruned trees. The crown-reduced trees used less water at >70 cm depth than the non-pruned trees (Figure 2.5). Averaged

across distance from tree, soil under the crown-reduced trees was approximately 0.05-0.07 m^3 water/m^3 soil, wetter than for the non-pruned trees between June and October. Afterwards, the soil profile under the crown-reduced trees re-hydrated more quickly than for the control at > 60 cm depth, but by January, the soils under all of the treatments had reached field capacity and no differences occurred.

In 2003, three each of the previously non-pruned, crown-reduced trees and crown-thinned trees respectively were severely pruned (see Introduction). The pruning treatments caused significant ($P = 0.01 – 0.06$) differences in soil moisture deficit during the period June to September. Averaged across the 2 and 6 m positions, the largest differences in soil moisture deficit occurred between the trees that had been crown-reduced in 2003 and those that had not been pruned in that year (Figure 2.4). The decrease in soil drying due to the pruning in 2003 was first evident in June, when the soil under these crown-reduced trees remained wetter than for any of the other treatments. By August the soil profile under the trees crown-reduced in 2003 had a moisture deficit approximately one third less than under the non-pruned trees. These large differences in soil moisture deficit (approximately 50 mm) remained until autumn rainfall. Crown-reduction and crown-thinning treatments applied in 2000 had no residual effect on soil drying in 2003 (Figure 2.4).

For the trees pruned in 2003, the soil remained wetter below 40 cm depth at 2 m from the trees and at 6 m, the soil remained wetter throughout the profile. This indicates that the current seasons pruning reduced soil drying both lower in the profile near the trunk and throughout the profile further away (Figure 2.5). When the maximum deficit was achieved in September the soil was uniformly dry throughout the profile (10-80 cm) at 2 and 6 m (Figure 2.5) from the tree, for all the trees not pruned in 2003, irrespective of their previous pruning history. This suggests that these trees fully utilised all the available water within a 6 m radius. This confirms also last year's results that large effects of pruning in 2000 did not continue beyond two growing seasons.

- Canopy pruning by either crown-reduction or crown-thinning conserved soil moisture by 10-40 mm within a 1 m profile for three growing seasons after their application by the fourth season the effect had disappeared.
- Crown-reduction has a greater effect than crown-thinning in reducing soil drying towards the periphery of the root system.
- Crown-reduction allowed faster and more complete recharging of soil water during a drier than average autumn and winter.
- Very severe crown-reduction (90% volume decrease) applied in 2003 was substantially more effective in increasing soil moisture conservation than the 72% crown volume decrease applied in 2000.

Figure 2.1: Oxygen isotope composition (δ^{18}O) from water samples extracted from tree twigs or soil samples taken from shallow (10-20 cm) and deep (60-70 cm) depths for wild cherry trees pruned in April 1999 (top, Experiment 1) and March 2000 (bottom, Experiment 2)
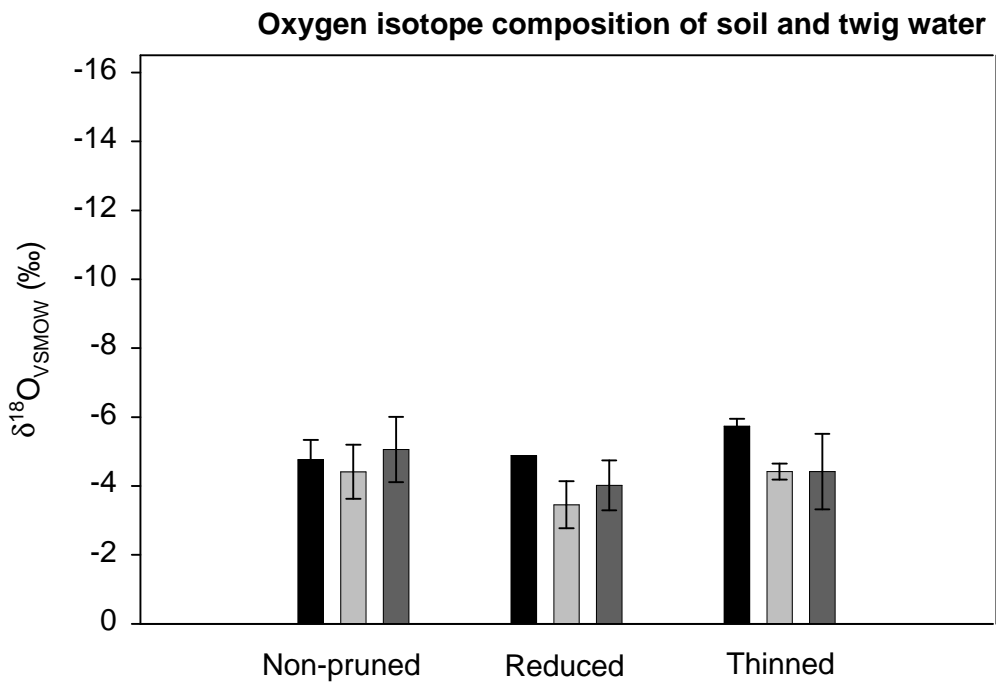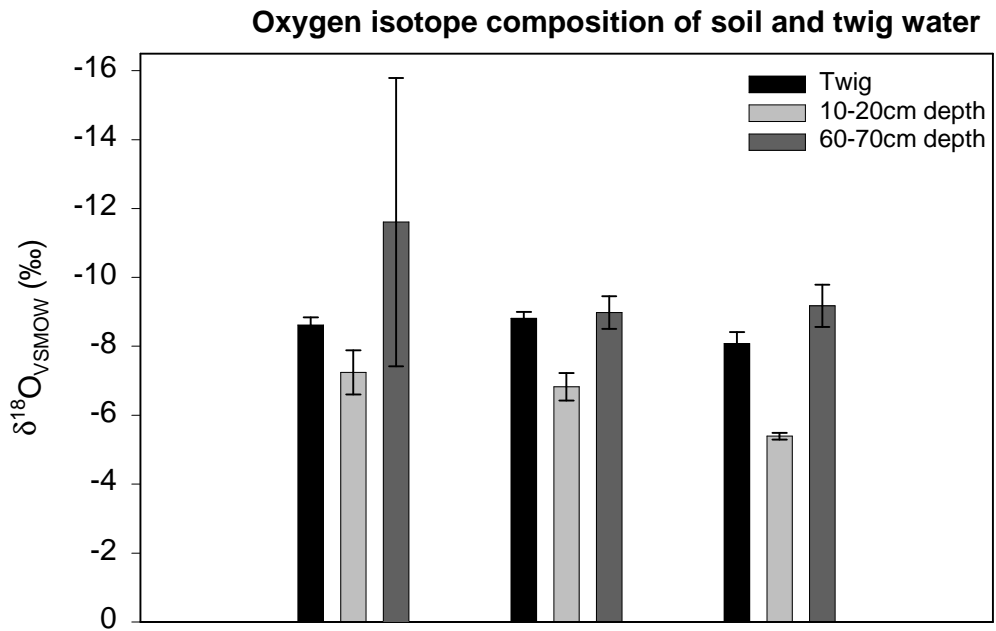
Figure 2.2: The effect of pruning treatments on soil moisture deficit (0-100 cm) at 1 and 3 m from the trunk under wild cherry trees pruned in March 2000 and re-pruned in April 2002 during the period 2000-2004 (Experiment 2). The reference date for field capacity was 16 December 2002. Vertical bars are the standard errors of differences between means. Monthly rainfall over the same period
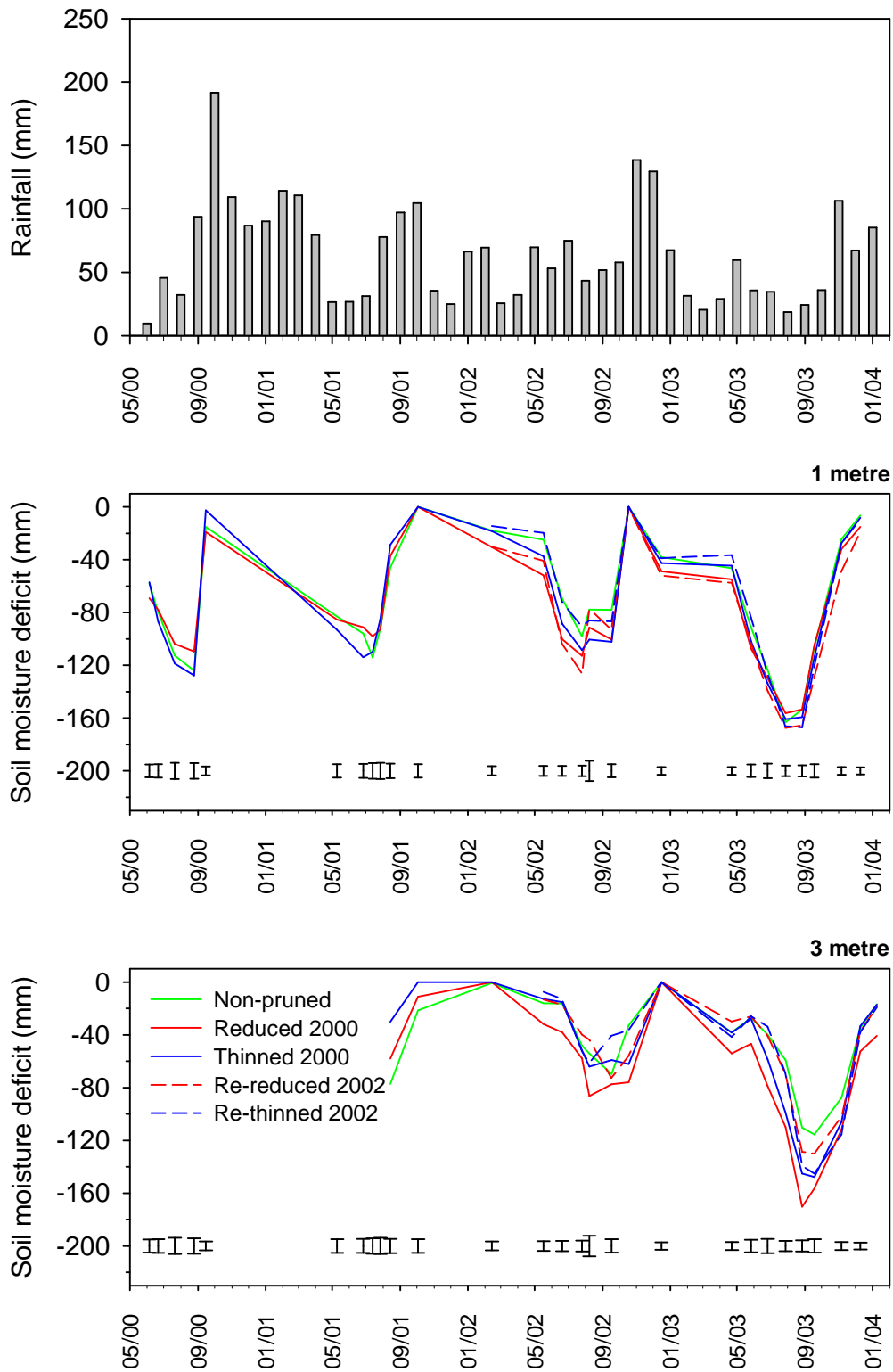
Figure 2.3: The effects of pruning treatments on soil moisture deficit (0-100 cm) under wild cherry trees pruned in April 2002 and soil droughted in 2002 and 2003 (Experiment 4). The reference date for field capacity was 16 December 2002. Vertical bars are the standard errors of differences between means. Monthly rainfall for the same period

Figure 2.4: The effect of pruning treatments on soil moisture deficit (0-100 cm) at 2 and 6 m from the trunk under London plane trees pruned in February 2000 and severely crown reduced in April 2003 during the period 2000-2004 (Experiment 5). Monthly rainfall for the same period. The reference date for field capacity was 30 November 2000, except data in 2003 which was 7 January 2004. Vertical bars are the standard errors of differences between means
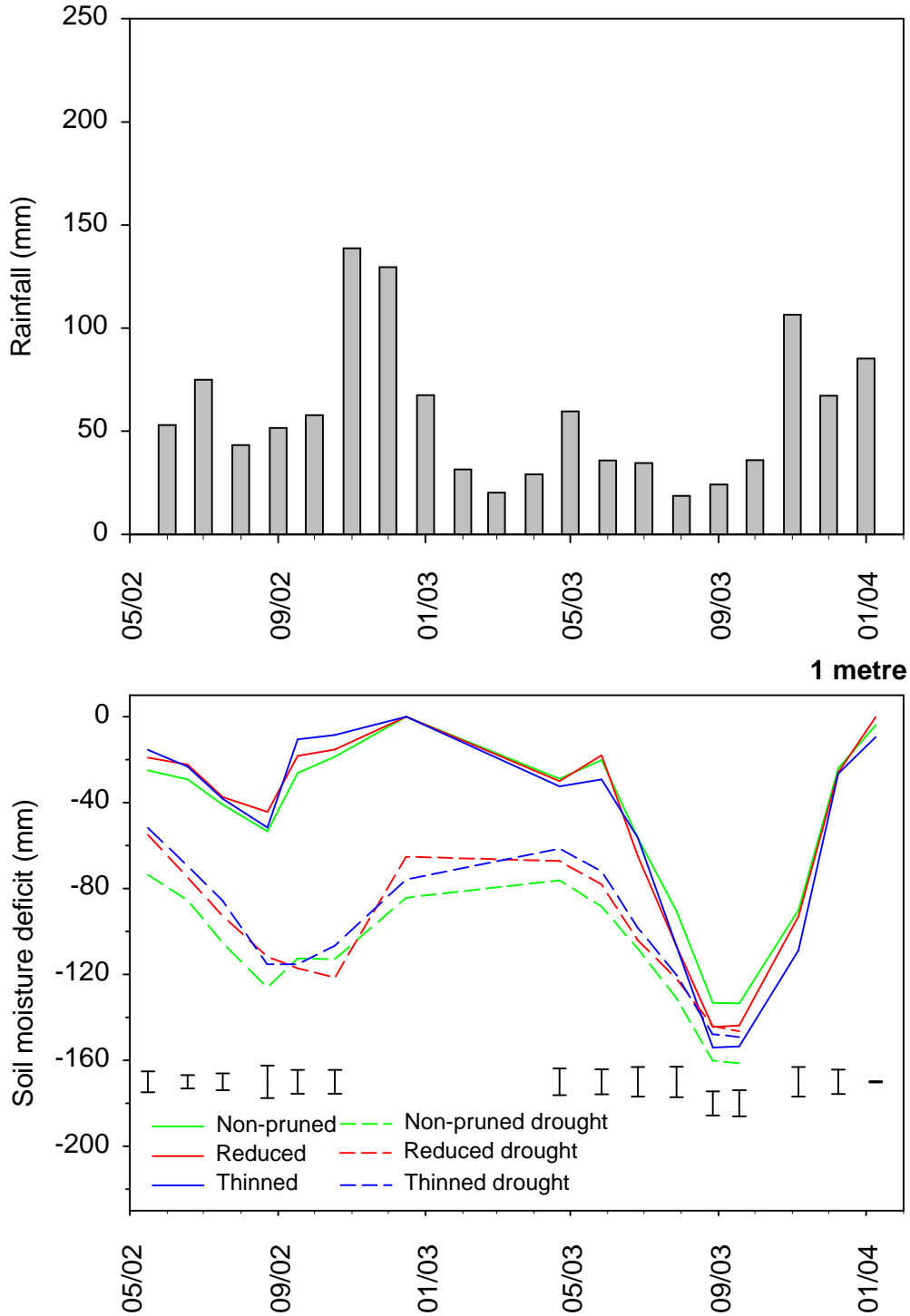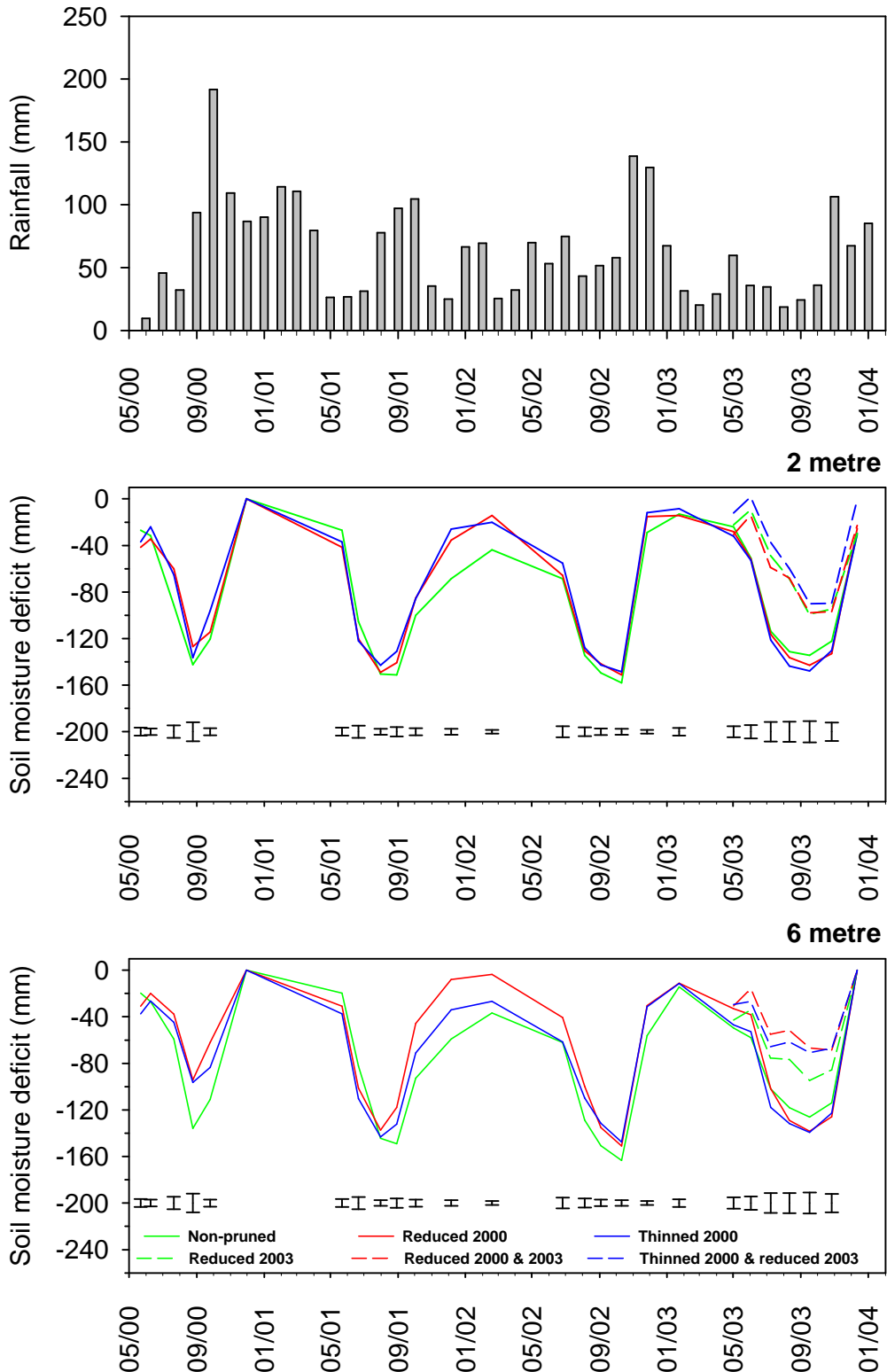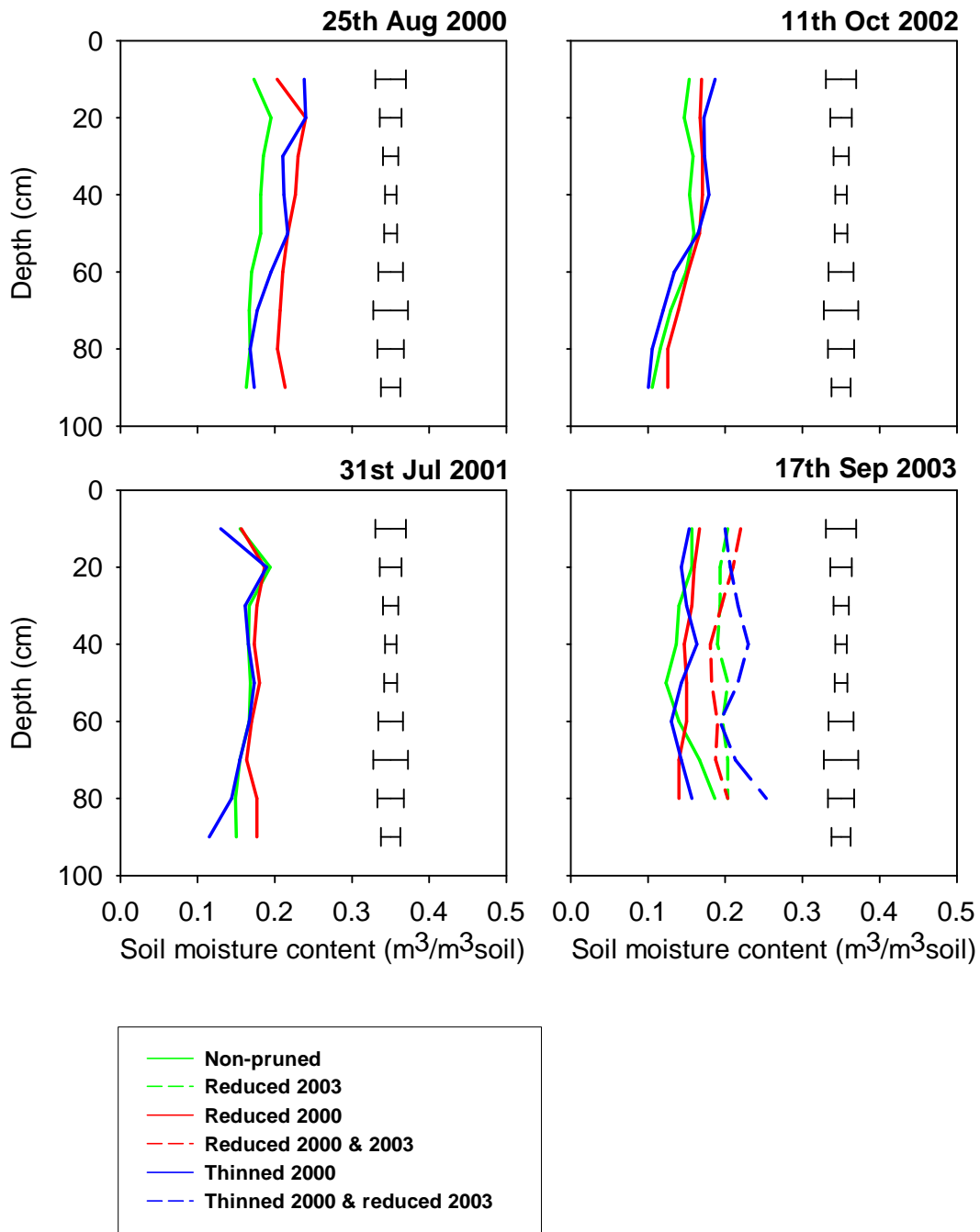
Figure 2.5: The effects of pruning treatments on minimum soil moisture content at 6 m from the trunk under London plane trees pruned in February 2000 and severely crown-reduced in April 2003 (Experiment 5). Horizontal bars are the standard errors of differences between the means

Objective 3:

Test the hypothesis that canopy manipulations by crown pruning and root restriction provide an effective way to control cherry and London plane trees growth and water use

Rationale

Results from experiments 1, 2, 3 and 5 have shown that crown-reduction increased soil moisture conservation by wild cherry and London plane trees, whereas crown-thinning had little effect (See objectives 1 and 2). To allow more accurate prediction of the impact of pruning and the development of improved practices, these effects must be linked to the recovery of leaf area within the canopy and tree water loss. The rate, form and shape of the re-growth of shoots and leaves within the canopy will impact on the extent and duration of soil drying following pruning. Differences may also occur between species due to differences in tree root mass, leaf size and re-growth capacity, the latter may be linked to assimilation rate and carbohydrate storage. The purpose of this objective was to determine the effects of pruning on the critical components of tree morphological structure, i.e. leaf size, shoot growth rate and shoot type and ultimately the impact on canopy leaf area recovery. Leaf nitrogen concentration was measured because it is a key factor known to determine and influence carbohydrate partitioning resulting from CO_2 assimilation and thus potential tree growth.

Restricting the volume of soil available for roots to exploit is an alternative method to pruning for controlling tree size and therefore water use (and root spread). This option is particularly suitable for newly planted trees. However, for success, determining the relationship between tree size and the volume of root restriction required is essential. Therefore, this formed another component of this objective.

Long term impact of crown pruning and environment on tree water use (Experiment 2)

Crown-reduction and crown-thinning applied to wild cherry trees in 2000 and repeated in 2002

Canopy re-growth

Methods

To determine canopy leaf area initially the volumetric shape of the crown was determined. A half-ellipsoid model was used for the crown-thinned and non-pruned trees, whereas a cylindrical model was used for the crown-reduced trees. The diameter of the ellipsoid or cylinder was defined as width to where the majority of the branch tips ended at the base of the canopy, extremities was excluded. Canopy depth was measured as the height from its base to the top, i.e. this excludes the height of the tree trunk. Digital hemispherical images were taken of each tree in the experiment on 20 March, 8 May, 28 June and 8 August 2000, 8 June, 19 July and 13 August 2001, 8 May and 6 August 2002 and 27 August 2003.

Results and conclusions

In the year of pruning (2000), the volume of the crown of the crown-reduced trees was decreased by 39% compared to the non-pruned trees, whereas that of the crown-thinned trees was decreased by 7%. The canopy of the crown-reduced and the crown-thinned trees recovered 94% and 50% respectively of their total leaf area compared to the non-pruned trees within the first year after pruning (Figure 3.1). Differences between the treatments in total canopy leaf area ($P<0.001$), leaf area density ($P<0.001$) and drip line leaf area ($P<0.001$) were significant. The greater leaf areas on the crown-reduced trees were achieved by the leaves being packed more close together at a higher density. The decrease of canopy leaf area caused by crown-thinning was due to a more open canopy reducing crown leaf area density to less than in the non-pruned trees.

This was due to increased extension from the epicormic shoots rather than the current season's terminal shoots. By August 2001, the crown-reduced trees had much greater (approximately 60%) total leaf areas than either the non-pruned or crown-thinned trees (Figure 3.1). The canopies of the crown-reduced trees remained more compact either of the other two treatments, the density of leaves within the canopy was much greater. The crown-thinned trees recovered to their original canopy leaf areas.

During the third growing season after pruning (2002), the differences caused by pruning in 2000 in total leaf canopy area, leaf area density and drip line leaf area index between treatments were still apparent. The crown-reduced trees had 37% greater total canopy leaf areas ($P<0.05$) than the non-pruned trees and those crown-thinned (Figures 3.1). The leaf area density was also 59% greater for the crown-reduced trees. However, the leaf area density of the crown-reduced trees, was approximately half that found in the previous year (2001). The canopy heights were approximately 1 m greater in 2002 than in the previous year, but the canopy radii were similar. Thus, the reduction in leaf area density was probably due to a combination of the leaves being spread more evenly within a larger volume of canopy and slower growth.

The re-reduction (30% crown-volume) applied in 2002 stimulated shoot growth to such an extent that the canopy had fully recovered its full leaf area by August 2002 (Figure 3.1). Again this was associated with a substantial increase in the leaf area density compared to the non-pruned trees ($P<0.001$). This was not the case for the crown re-thinned trees, since their canopies were more open (less dense) and by August contained 40% less leaf area than the non-pruned trees.

By the end of the 2003 growing season, the effect of re-pruning in 2002 on canopy leaf area had also disappeared as differences were no longer significant ($P=0.192$), but the leaf area density of the crown-reduced trees remained significantly greater ($P<0.001$) than for the other treatments. Thus, for crown reduction applied in 2000 and 2002, the largest effect on leaf area density occurred in the second year after the application of pruning.

- Crown-reduced trees recovered their canopy leaf areas within one growing season of pruning
- Crown-thinned trees required two growing seasons to recover their canopy leaf area.
- Crown-reduction stimulated shoot growth and caused substantial changes in the canopy leaf architecture by increasing leaf area density within a smaller crown volume.
- Crown-thinning opened the canopy and reduced canopy leaf area density, without invigorating growth.

Shoot growth

Methods

Measurements were made on ten randomly selected terminal extension shoots per tree from four trees per treatment after cessation of growth in 2000-2001. On 30 July 2002, mean shoot length, total shoot leaf area and number of leaves per shoot were measured on the same four shoots, per shoot type, growing on the north side of the canopy.

Results and conclusions

Crown-reduction substantially invigorated tree growth by increasing terminal shoot extension in the first season after pruning (Figure 3.2). Shoot lengths of the crown-reduced trees were increased five fold compared to the non-pruned trees. In the second year after pruning this effect continued, but to a lesser extent and was restricted to terminal shoots. Crown-thinning also caused greater shoot extension, but the effect was much smaller than that caused by crown-reduction. The pruning treatments applied in 2000, continued to influence terminal shoot length, but had no effect on lateral shoots in the third season after pruning.

The effect of the crown re-reduction (applied 2002) was much larger. Mean terminal shoot length increased almost four fold and lateral shoot length doubled compared to the non-pruned trees (3.2). Crown re-thinning (applied 2002) had no effect on terminal or lateral shoot lengths.

The original crown-reduction and crown-thinning treatments (applied 2000) persisted in 2002, increasing the total shoot leaf area by a small amount, but, again the effects of the 2002 re-reduction treatment were much larger. Terminal shoot leaf area of crown re-reduced trees was almost double that of the non-pruned trees (Figure 3.2). The average size of individual leaves on terminal shoots was increased also by 56% as a result of crown re-reduction. Crown re-reduction increased the leaf area of lateral shoots by 60% compared to the shoots on non-pruned trees. Crown re-thinning had no effect on leaf area of either terminal or lateral shoots.

Crown re-reduction, crown-thinning and crown re-thinning increased the mean number of leaves per terminal shoot compared to the non-pruned trees (Figure 3.2). Crown re-reduction had the greatest effect, increasing the average number of leaves per shoot by four compared to the shoots of non-pruned trees.

- Crown-reduction applied singly and/or reapplied after two years substantially increased terminal shoot extension growth for three growing seasons after pruning.
- Crown-thinning had a much smaller effect on increasing terminal shoot growth than crown-reduction.
- Increases in shoot length, leaf area, number of leaves per shoot and the average size of individual leaves on lateral and terminal extension shoots caused by crown-reduction determine why canopy leaf area and leaf area density increased rapidly over the course of a growing season in the crown re-reduced trees (Figure 3.1).

Leaf growth and nutrient content

Methods

Measurements of leaf area: weight relationships and C and N content were determined on leaf samples sent for carbon isotope analysis. These samples were collected on 3 October 2000, 20 September 2001 and 13 August 2002. Three leaves were collected from the north side of the canopy of three trees per treatment on each date.

Results and conclusions

The areas of individual leaves on trees subjected to crown-reduction were greater during the three growing seasons after pruning (Figure 3.3). Although there was a slight reduction in successive years, mean leaf areas nearly doubled following the second pruning treatment in 2002. The mean leaf area of crown-thinned trees was unaffected by pruning. Although leaves from crown-reduced and crown-thinned trees had lower specific leaf weights (SLW, a measure of how efficiently the leaves produce carbohydrate to capture light) in the first year after pruning, in general terms SLW was constant across all treatments in subsequent years, including the re-pruning treatments in 2002 (Figure 3.3). Following the first pruning treatment, leaf N content was increased in crown-reduced trees, but differences disappeared subsequently and leaves were unaffected when the pruning treatments were repeated in 2002 (Figure 3.3).

- Trees produced larger leaves for three growing season after crown-reduction, the largest effect occurred during the year of pruning.
- Crown-thinning did not influence leaf size or leaf N content.
- Crown-reduction caused increases in leaf N content for two years following first pruning.
- Repeated pruning did not influence leaf N content.

Tree water use (sap flux)

Methods

Sap flux per tree was summarised by averaging the mean sap flux measured for 12 days in September 2000; 7, 12 and 9 consecutive days in June, August and September 2001 respectively and 26 consecutive days during July and August 2002. Two TDP sets per tree were used on three tree per treatment in 2000 and three TDP sets per tree were used in 2001 and 2002. The probes were placed in the trunk approximately 1.4 m above ground level.

Results and conclusions

The variability of sap flux both within and between trees was large and this led to large standard errors associated with these measurements and thus helped to mask true differences between treatments on sap flux (Figure 3.4). During September 2000 (year of pruning), the average sap flux (daily water use) of the crown-reduced and crown-thinned were not significantly different from the non-pruned trees (P=0.617). In the next year (2001), the crown-reduced trees had the greatest average sap flux in each month and the crown-thinned trees had the least earlier in the season (June and August). By September the crown-thinned and crown-reduced trees had similar sap fluxes. However, differences between treatments were not significant (P=0.095, June; P=0.0152 August; P=0.561 September).

During 2002, when the trees were pruned a second time, a consistent, but non-significant (P>0.05) trend of less water use by the re-thinned and re-reduced trees was indicated over 26 days in July and August (Figure 3.4). The re-reduced and re-thinned trees had 16% and 8% respectively less sap flux than the non-pruned trees.

- Thermal dissipation probes did not consistently detect differences in sap flux between trees.

Effects of crown pruning on the phenology of re-growth (Experiment 3)

Crown-reduction and crown-thinning applied to wild cherry trees in 2001

Seasonal growth patterns

Pruning has been shown to have a significant impact on canopy development and architecture (Genard, *et al*., 1998, Lakso, 1984, Porter and Llewelyn, 1984). Therefore detailed observations were made of shoot and leaf growth to provide a more detailed understanding of canopy responses to crown-reduction and crown-thinning.

Methods

Observations of shoot and leaf development were made by visual inspection of shoots growing on the south side of the tree throughout the season on all of the trees. The seasonal growth was divided into five growth stages ('new leaf production', 'shoot extension', 'full canopy', 'leaf senescence' and 'leaf fall').

Results and conclusions

Crown-reduction prolonged the production of new leaves by approximately 6 weeks and shoot growth by 8 weeks compared to the control trees (Figure 3.5). This increase in the growth period induced by crown-reduction was largely due to shoots that developed from epicormic and lateral buds. This confirms that crown-reduced trees have greater potential for re-growth than crown-thinned trees. The extended initial growth period of crown-reduced and crown-thinned trees delayed the onset of full canopy compared to the non-pruned trees. Crown-reduction prolonged the duration of leaf senescence and leaf fall, delaying the onset of dormancy by approximately 2 weeks relative to the non-pruned and crown-thinned trees.

·   Crown thinning does not influence seasonal phenology.
·   Crown reduction extends canopy development allowing for greater rates of re-growth.

Leaf and shoot development

Previous studies on fruit trees have shown that pruning has the potential to significantly invigorate lateral and terminal shoot growth (Lakso, 1984, Porter and Llewelyn, 1984). Thus shoots included in this study were sub-divided according to their position on the branch. Leading shoots were classified as 'terminal' whilst those behind where classified as 'lateral'. In addition to these, epicormic shoots emergent specifically on larger branches in the crown-reduced trees were also included.

Methods

Measurements of leaf area were made on individual leaves to determine the effect of crown-reduction and crown-thinning on leaf size. Thirty-five fully expanded leaves were taken from each shoot type (terminal, lateral and epicormic) from each tree.

Terminal shoot lengths were measured on 30 April, 14 May, 4, 18 June, 6 August and 1 October in 2001 to determine the impact of the two canopy pruning treatments on shoot extension. On each occasion measurements were made on the same four shoots on the south side of each tree.

Results and conclusions

Individual leaves taken from epicormic and lateral shoots on crown-reduced trees were significantly ($P<0.01$) larger than leaves taken from either the non-pruned or crown-thinned trees (Figure 3.6). The greater leaf area of individual leaves produced by epicormic and lateral shoots in response to crown-reduction is consistent with results from experiments 1 and 2.

Both crown-reduction and crown-thinning significantly ($P<0.01$) influenced shoot growth during the season after pruning (Figure 3.6). Between 4 and 18 June 2001 the rate of shoot growth in the crown-reduced trees increased more rapidly than in the non-pruned and crown-thinned trees, and ceased later. By August the shoots in the crown-reduced trees were significantly longer than those of the crown-thinned trees and non-pruned trees. This data clearly illustrates that crown reduction stimulates significantly greater rates of growth allowing rapid regrowth in these canopies.

·   Crown-reduction both invigorates and prolongs terminal and lateral shoot growth allowing for rapid re-growth of the canopy during the first season following the application of pruning.
·   Crown-thinning does not invigorate or prolong tree growth compared to non-pruned trees.

Impact of drought on response to crown pruning (Experiment 4)

Crown-reduction and crown-thinning applied to wild cherry trees in 2002 and soil drought applied in 2002 and 2003

Canopy re-growth

Methods

Digital hemispherical images were taken of each tree in the experiment on 8 May and 6 August 2002 and 19 May and 27 August 2003. A half-ellipsoid model was used to represent the 'volumetric shape' of the canopies of all the trees. Analyses of the images of the canopies of each tree were used to determine the canopy leaf area, drip line leaf area index and leaf density.

Results and conclusions

The canopy leaf areas of the crown-reduced and crown-thinned trees were approximately half that of the non-pruned trees one month after pruning. These differences were significant ($P<0.001$). However, within another 3 months (August 2002) the canopies of the crown-reduced trees had recovered, whereas those of the crown-thinned trees remained approximately 40% less than the non-pruned trees (Figure 3.7). The stimulation of shoot growth of the crown-reduced trees was clearly indicated by the substantial increase in canopy leaf area density. Therefore, the canopy contained a similar leaf area to the non-pruned trees, made up of larger leaves packed more closely together into a smaller volume. The drought treatment had no effect on the canopy growth of the trees and both of the pruning treatments produced similar effects in the either the droughted or non-droughted trees.

In the second growing season (2003) after pruning, the canopy leaf areas (Figure 3.7) of the crown-reduced and crown-thinned trees were again approximately half of those of the non-pruned trees in May ($P<0.001$). By August, the differences between all the treatments in canopy leaf area had disappeared ($P=0.871$) although the crown-reduced trees maintained a higher and the crown-thinned trees maintained a lower leaf area density than the non-pruned trees ($P=0.06$). Therefore, the pruning treatments delayed the onset of growth early in the season, but later the pruned trees compensated by increasing their growth relative to the non-pruned trees. The soil drought did not influence canopy leaf area development in the second growing season

- Crown-reduced trees recovered their canopy leaf area within one growing season.
- Crown-thinned trees recovered their canopy leaf area within two growing seasons
- Crown-reduction invigorated trees growth and led to substantial increase in canopy leaf area density.
- Crown-reduction and crown-thinning delayed the onset of growth in the second year after pruning.

Shoot growth and development

Methods

Shoot length and leaf area was measured on four terminal and four lateral shoots per tree on the north side of the canopy on 18 August 2002. Shoots were sampled on three trees per treatment. The measurements on shoot length and number of leaves per shoot were repeated in 2003.

Results and conclusions

The length of terminal shoots of crown-reduced non-droughted and droughted trees was more than double those of their respective non-pruned controls (Figure 3.8) in the year following pruning.

Lateral shoot lengths were also increased following crown-reduction. This effect persisted but to a lesser extent in the second year after pruning. Drought alone had no effect on the extension growth of shoots.

Crown-reduction also approximately doubled the total leaf area per terminal shoot compared to the non-pruned trees in the non-droughted and droughted soils respectively in the year following pruning (Figure 3.8). Lateral shoot leaf area was similarly increased. Crown-thinning had a negligible effect on terminal shoot leaf area, but increased lateral shoot leaf area during the two seasons after pruning compared to non-pruned trees. Crown-reduction increased leaf size on extension and lateral shoots compared non-pruned trees (data not presented). Drought alone had no effect on the total leaf area per shoot.

Crown-reduction and crown-thinning increased the mean number of leaves on terminal and lateral shoots in the two years following pruning (Figure 3.8c). The effect of crown-reduction was largest in soil-droughted trees in the first year after pruning.

- Crown-reduction increased terminal and lateral shoots extension and leaf areas compared to non-pruned trees.
- Crown-thinning did not influence terminal shoot extension.
- Crown-reduction and crown-thinning increased the number of leaves per shoot.
- The effect relative to the non-pruned trees of crown-thinning on numbers of leaves per shoot and shoot length was greatest on lateral shoots.
- Soil drought did not influence growth.

Leaf growth characteristics

Methods

Leaf samples were collected on 21 July 2002 and 21 August 2003. On each date four leaves were collected from the north side of the canopy of three trees per treatment.

Results and conclusions

Relative to the non-pruned trees crown-reduction, increased individual leaf size of trees in droughted and non-droughted soils during in the year of pruning (2002) and the following year (Figure 3.9). Crown-thinning had a similar but smaller effect. Specific leaf weight tended to be lower in crown-reduced trees in the first year after pruning (Figure 3.9). In contrast to Experiment 2, nitrogen content tended to be highest in the leaves of crown-thinned trees in the year following pruning, but values were similar in the second year, irrespective of the drought regime (Figure 3.9).

- During the two years following pruning, crown-reduced trees produced larger leaves than non-pruned trees.
- In the first year after pruning specific leaf weights of crown reduced trees were less than non-pruned and crown-thinned trees.
- Crown-thinning caused a small increase in leaf size compared to non-pruned trees.

Tree water use

Methods

Three TDP sets were used in three trees per treatment in all of the six drought and pruning treatment combinations. Thus a total of 54 heat dissipation probes were used on 18 trees. Good traces were obtained for 12 days in June, 14 days in July and 10 days in August and 24 days in September 2002. This was repeated in for 17 days in June, 20 days in July and 18 Days in August 2003.

Results and conclusions

The greatest water use occurred in June (overall mean of all treatments 50 l^{-1} d^{-1} $tree^{-1}$) and subsequently the overall monthly averages were similar until September 2002 (Figure 3.10). During June and July the water use of the crown-reduced trees averaged across drought treatments was 29-39 % less than the non-pruned trees. Over the same period, the crown-thinned trees had 21-29 % less water use than the non-pruned trees (Figure 3.10). These differences were significant ($P<0.01$) and were consistent irrespective of whether the trees were droughted or not. During August and September the effects continued, but were smaller and not significant ($P>0.05$).

During the second growing season after pruning, crown-reduction and crown-thinning did not influence the daily sap flux (Figure 3.10). Differences between treatments were not significant (*P*>0.33). This concurs with the soil moisture measurements that also indicated that the both of the pruning treatments no longer influenced soil drying.

Surprisingly, the drought treatments had no effect on the water use of the trees in either year (data not presented). Since the soil was known to be droughted near the trees, the trees must have accessed water from beyond the droughted zone or sufficient water remained available for growth.

- Crown-reduction and crown-thinning deceased daily sap flux in the year of pruning, and the effects were greatest early in the growing season.
- Crown-reduction deceased sap flux more than crown-thinning.
- Pruning did not significantly influence sap flux during the second season after pruning.
- The soil droughting treatment had no effect on sap flux in either year.

Assessing the impact of crown pruning on large amenity trees (Experiment 5)

Crown-reduction and crown-thinning applied to mature London plane trees in 2000 and severe reduction applied in 2003

Canopy re-growth

Methods

Digital hemispherical images were captured for each tree in the experiment on 20 March, 9 June, 21 July, and 25 August 2000, 10 July and 30 August 2001 and 27 August 2003. A half-ellipsoid model was used to represent the 'volumetric shape' for all the treatments applied in 2000 as the tops of the trees were not so severely effected (flattened) as was the case with the wild cherry tree experiments. A cylinder model was used for the severe pruning treatment applied in 2003 because this gave a better representation of the modified canopy shape. Analyses of the images of the canopies of each tree were used to determine the canopy leaf area, drip line leaf area index and leaf density.

Results and conclusions

The London plane trees were much larger than the cherry trees in experiments 1,2 and 4 and their final total canopy leaf area was approximately 10 times greater (Figure 3.11). In May, the canopy had developed only 30% of its leaf area (c.f. wild cherry trees 75%). Their canopies tended to develop later in the growing season between June and August (data not presented).

The crown-reduction and crown-thinning decreased the volumes of the canopies of trees by 72 and 6% respectively. Four months after pruning (August 2000) the total leaf areas of the crown-reduced and crown-thinned trees were approximately one third and two thirds respectively of the non-pruned trees (Figure 3.11). These differences were significant ($P<0.001$). The leaf area density of crown-reduced trees was much greater than the non-pruned trees, whereas for the crown-thinned trees the reverse was true. This suggests that substantial re-growth of leaves and shoots occurred within the more compact canopy of the crown-reduced trees, but did occur in the crown-thinned trees. The drip line leaf area indexes of crown-reduced and crown-thinned trees were similar to each other and less than the non-pruned trees, indicating that the trees apportioned their re-growth to the ground area covered.

In August 2001 (i.e. 16 months after pruning) the canopies of trees receiving either pruning treatment had recovered a large proportion of their leaf areas. The total leaf areas of the of crown-reduced and crown-thinned trees were approximately 70 and 85% respectively of those the non-pruned trees (Figure 3.11). Thus a considerable amount of extra growth that had occurred within the crown-reduced canopy during this second year after pruning compared to the other two treatments. In fact, the leaf area density was more than double that found in the previous year. Furthermore, the average size of individual leaves on whole branch samples (see also Figure 3.13) in the crown-reduced trees (274 cm^2) was approximately double that found in the non-pruned (138 cm^2) and crown thinned trees (106 cm^2). However, the crown-reduced trees still had significantly ($P=0.01$) less leaf area than the non-pruned trees, but the difference between the crown-thinned and non-pruned trees was not significant ($P>0.05$). The leaf area density of the crown-thinned and non-pruned trees were similar, suggesting that the crown thinning treatment produces a balance in the re-growth which is more akin to the non-pruned trees.

By September 2002 (29 months after pruning), the crown-reduced and crown-thinned trees had fully recovered their canopy leaf areas compared to the non-pruned trees as there were no differences between the treatments (Figure 3.11). The canopy of the crown-reduced trees remained significantly denser ($P<0.05$) than for either of the other treatments. The average size of individual leaves was similar (Figure 3.12). This contrasts sharply with the last year, therefore, the invigoration caused by the pruning treatments applied in 2000 had declined. The crown-reduced trees were maintaining similar leaf areas by packing their slightly larger leaves into smaller crown volumes.

The 90% crown-reduction applied in 2003 greatly reduced subsequent canopy leaf area development. The pruned trees had recovered only one quarter of their canopy leaf area c.f. those of the non-pruned trees four months after pruning (Figure 3.11). These differences in canopy leaf area were statistically significant ($P<0.001$). Irrespective of their previous history, the newly and re-pruned trees almost trebled their leaf area density in response to the pruning treatments, confirming that the new growth following pruning produced larger leaves more closely packed together.

The total leaf area of the canopy of trees crown-reduced or crown-thinned in 2000 was similar to those of the non-pruned trees, confirming the recovery observed last year. Since the canopy volume of the crown-reduced trees remained smaller than for either of the other two treatments, the recovery of leaf area was due again to a greater density (approximately double that of the control) of leaves within the crown.

- Crown-thinned trees recovered their canopy leaf areas within two growing seasons.
- Crown-reduced trees recovered their canopy leaf areas within three growing seasons.
- Crown-reduction produced trees with more compact canopies with larger individual leaves, more closely packed together resulting in greater leaf area densities.
- Crown-thinning had no impact on leaf size and the trees maintained more open canopies.

Leaf growth characteristics

Leaf samples were collected on 9 September 2000, 30 August 2001, 3 September 2002 and 11 September 2003. On each date, three leaves were collected from the north side of the canopy of three trees per treatment.

Results and conclusions

Crown-reduction increased leaf size in the two years of pruning following crown-reduction in 2000 (Figure 3.12). Following the severe crown-reduction in 2003, leaf size was substantially increased and specific leaf weight reduced (Figure 3.12). Nitrogen content was greatest in the leaves of the crown-reduced trees in the year following pruning. Subsequently this effect disappeared, except in 2003 when the severe reduction treatment increased leaf N content (Figure 3.12).

- Crown-reduction substantially increased leaf size in the two years following pruning. (This response was similar to that found for wild cherry trees in Experiments 1, 2 and 4.)
- The larger leaves on the crown-reduced trees had greater leaf N contents in the year of pruning.

Tree water use

Methods

Heat pulse sensors (Greenspan Technology Pty Ltd, Queensland, Australia) were used to measure sap flux in the London plane trees in 2002. Two probes were used per tree and each probe contains two sensor pairs. Thus, sap flux was measured at four positions within each tree on three trees per treatment. In order, to obtain replication of measurements, the probes were moved every six days between three blocks throughout the season. Thermal dissipation probes were used to measure sap flux in 2001 and 2003. Three probe sets were used on three trees per treatment and data were averaged over 5 days in July, 9 days in August and 7 days in September 2001. In 2003, nine TDP sets were used per tree on three pairs of severely crown-reduced and non-pruned trees. Data were averaged over 12 days in September.

Results and conclusions

A similar problem of variability in the measurements between and within trees was found to that of the cherry trees. In 2001 no significant differences between treatments in average sap flux per tree occurred in July and August, but in September the crown-thinned trees had the lowest daily sap flux, however, this difference was not significant (P=0.662). In the third growing season after pruning (2002), the greatest sap flux occurred in late summer (August and September). The mean daily sap flux of the non-pruned trees was slightly greater than the other treatments in July and August, otherwise, the differences between the treatments were smaller. The variability between and within trees was large and the standard errors of the differences between the means also were large. Therefore, none of the differences between the treatments were statistically significant (P>0.05). This suggests that carry over effects of the pruning treatments applied in 2000 did not persist in 2002. This is in agreement with the measurements of soil moisture deficit. No

differences in sap flux were detected in 2003 between the non-pruned and the severely pruned trees.

- Heat pulse and thermal dissipation based technique were not sensitive enough to detect treatment differences in daily sap flux in large trees.

An alternative approach to controlling growth in newly planted amenity trees (Experiment 6)

Norway maple and lime trees planted in spring 2000 into tree pits of volume varying between 250-2000 l, lined with a geotextile material impermeable to roots

Tree growth

Methods

During the dormant season, the shoot extension of these trees was measured by sampling every branch on every tree. Initially the number of shoots was estimated and a division factor (n) used to allow approximately 40 shoots to be measured. The division factor (n) varied between 1 and 10, e.g. one shoot being measured to every tenth shoot being counted. The actual number of shoots measured per tree was recorded. The total shoot extension was the sum of the measured shoot lengths multiplied by the division factor. The growth data was highly variable between trees and therefore the data was $\log_e$ transformed to stabilise variance.

Results and conclusions

In the first year after planting none of the root restriction treatments influenced the growth of the trees of either species (Figure 3.14). By the end of the next year (2001), the lime trees had approximately 15 times greater shoot extension than the Norway maple trees (Figure 3.14). For both species the trees growing in the smallest volume of soil (250 litre) had less growth compared to the unrestricted trees and those within the largest volume (2000 litres) had the greatest. This was probably due to the soil disturbance caused by creating the planting pits improving the physical and nutritional conditions for root growth compared to the unrestricted trees. However, neither of these effects effect were significant (P=0.15 and P=0.44 for lime and Norway maple respectively).

By the end of the third growth season (2002), the trees growing in the smallest volume of soil had substantially less shoot extension (P=0.008 and P<0.001 for lime and Norway maple respectively) compared to their respective unrestricted trees (Figure 3.14). For both species, restricting the roots within 2000 litres continued to have no effect on growth. The effects on cumulative growth assessed by measuring girth also indicated large and significant effects caused by the root restriction treatments ≤ 1000 litres (Figure 3.14). These effects continued in the fourth growing season so that the trees growing in the smallest volume of soil (250 litre) had substantially (75-90 %) reduced shoot extension (P<0.01) compared to their respective unrestricted controls. The shoot extensions of Norway maple and lime trees were decreased by 28% and 64% respectively for those trees growing in the 1000 litre pits. Thus, lime appeared to be more sensitive than Norway maple to this method of growth control. This confirms the results found in the previous year, that after three growing seasons 1000 litres is not a large enough volume of soil to support the full growth of either species. The trees within the restricted volumes (≤1000 litres) remained healthy, indicating that this technique may be used successfully to control vigour without detriment to the tree. For both species, restricting the roots within 2000 litres had no effect on growth compared to the non-restricted treatment, therefore between 1000 and 2000 litres is ample to maintain full growth for up to four years.

- Root restriction may be used successfully to control tree vigour without detriment to tree health.

- The shoot extension growth was closely related volume of root restriction for soil volumes ≤ 1000 l.
- A lined planting pit of 2000 l is ample to support full growth for four growing seasons.

Controlling water use of trees to alleviate subsidence risk

Figure 3.1: Leaf area density, drip line leaf area index and total canopy leaf area determined by digital analysis of hemispherical images for wild cherry trees pruned in March 2000 and re-pruned in April 2002 for the period 2000-2003 (Experiment 2). Vertical bars are standard error of differences between means
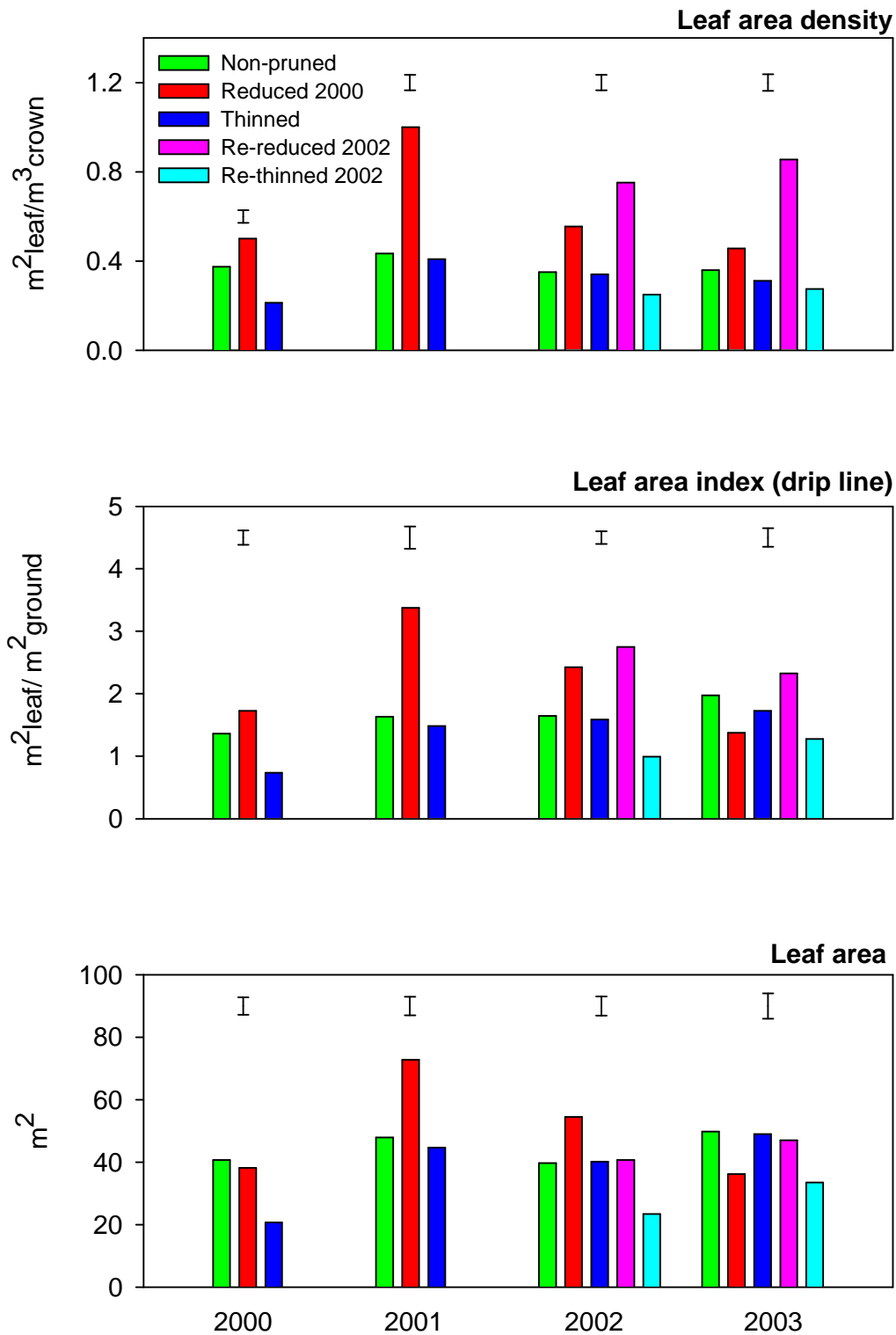
Figure 3.2: Shoot growth of wild cherry trees pruned in March 2000 and re-pruned in April 2002 (Experiment 2). Terminal shoot length was measured after cessation of growth in 2000 and 2001. Leaf area and number of leaves per shoot measurements were made on 30 July 2002. Vertical bars are standard error of differences between means

Controlling water use of trees to alleviate subsidence risk
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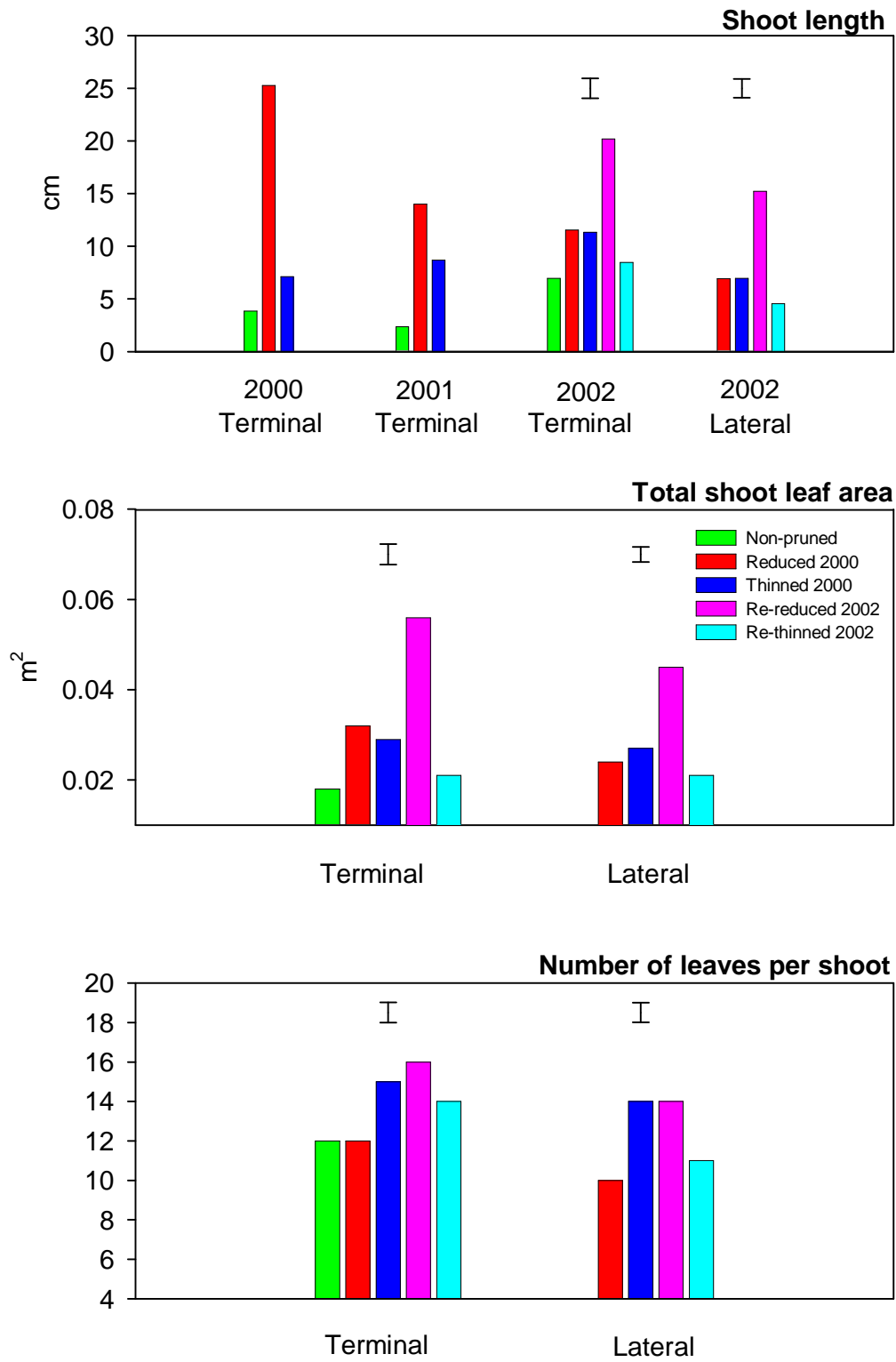
Figure 3.3: Leaf size, specific weight and nitrogen content of wild cherry trees pruned in March 2000 and re-pruned in April 2002 (Experiment 2). Vertical bars are the standard error of each mean, analysed individually
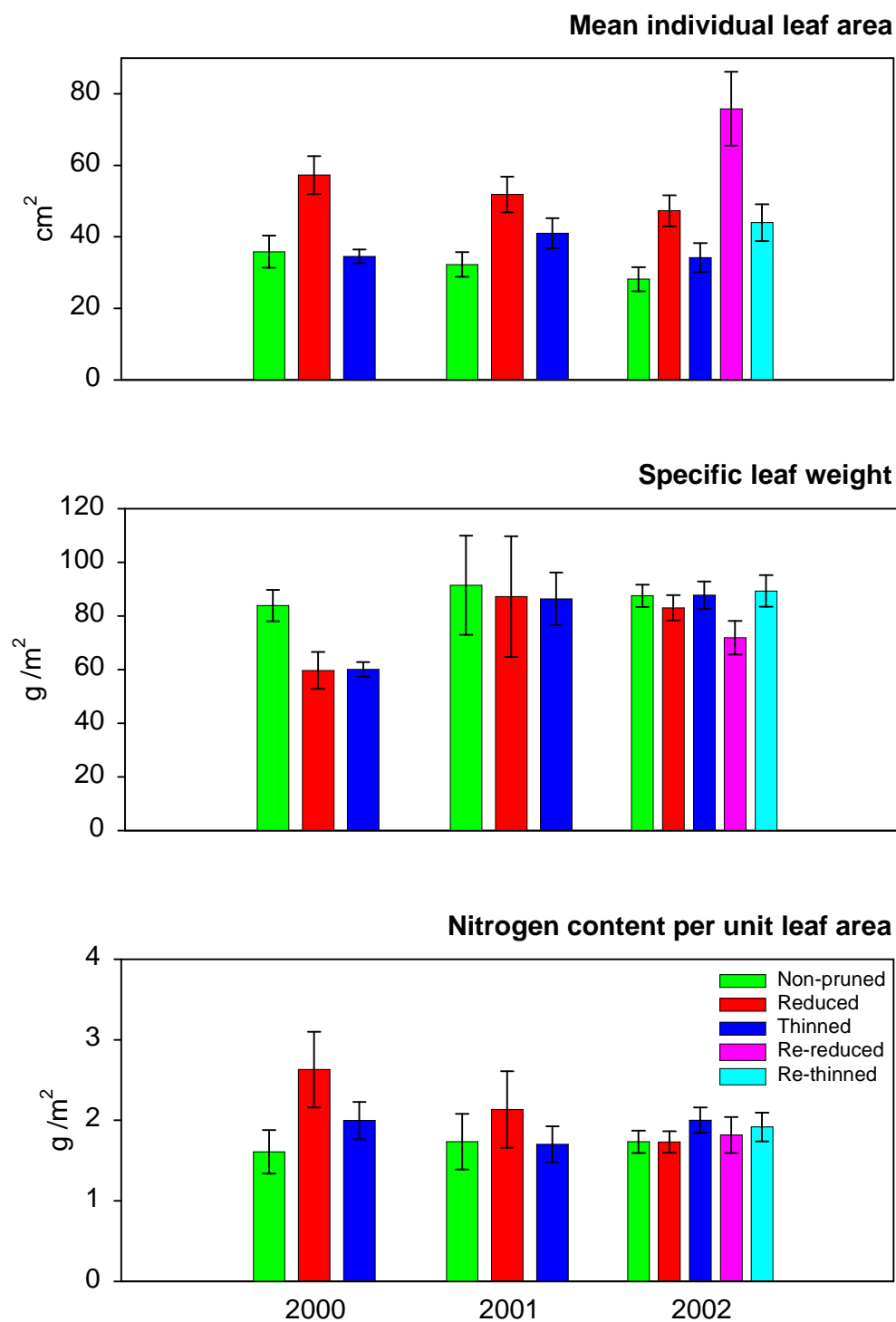
Figure 3.4: The average daily sap flux (water use) for wild cherry trees pruned in March 2000 and re-pruned in April 2002 (Experiment 2). Vertical bars are the standard error of differences between means
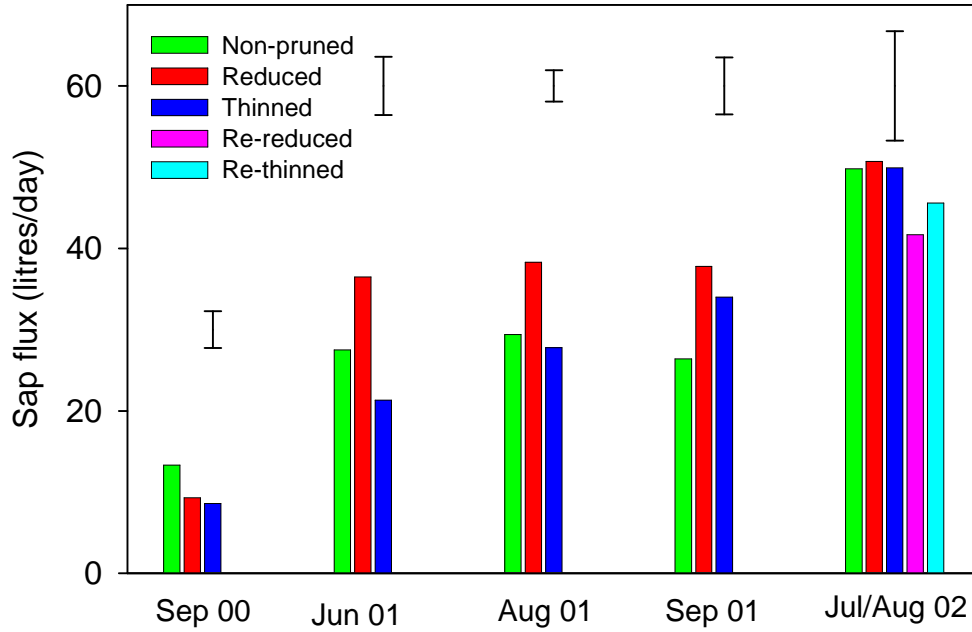
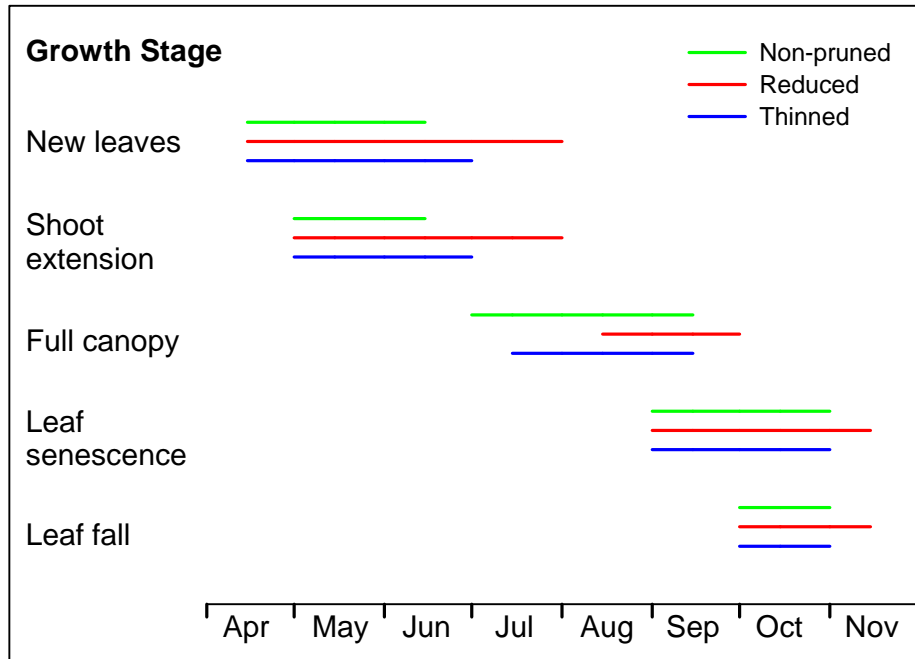Figure 3.5: Growth and development of wild cherry trees pruned in March 2001 (Experiment 3)

Figure 3.6: Mean leaf size on terminal, lateral and epicormic extension shoots, and the growth of terminal extension shoots of wild cherry trees pruned in March 2001 (Experiment 3). Vertical bars are standard errors of differences between the means
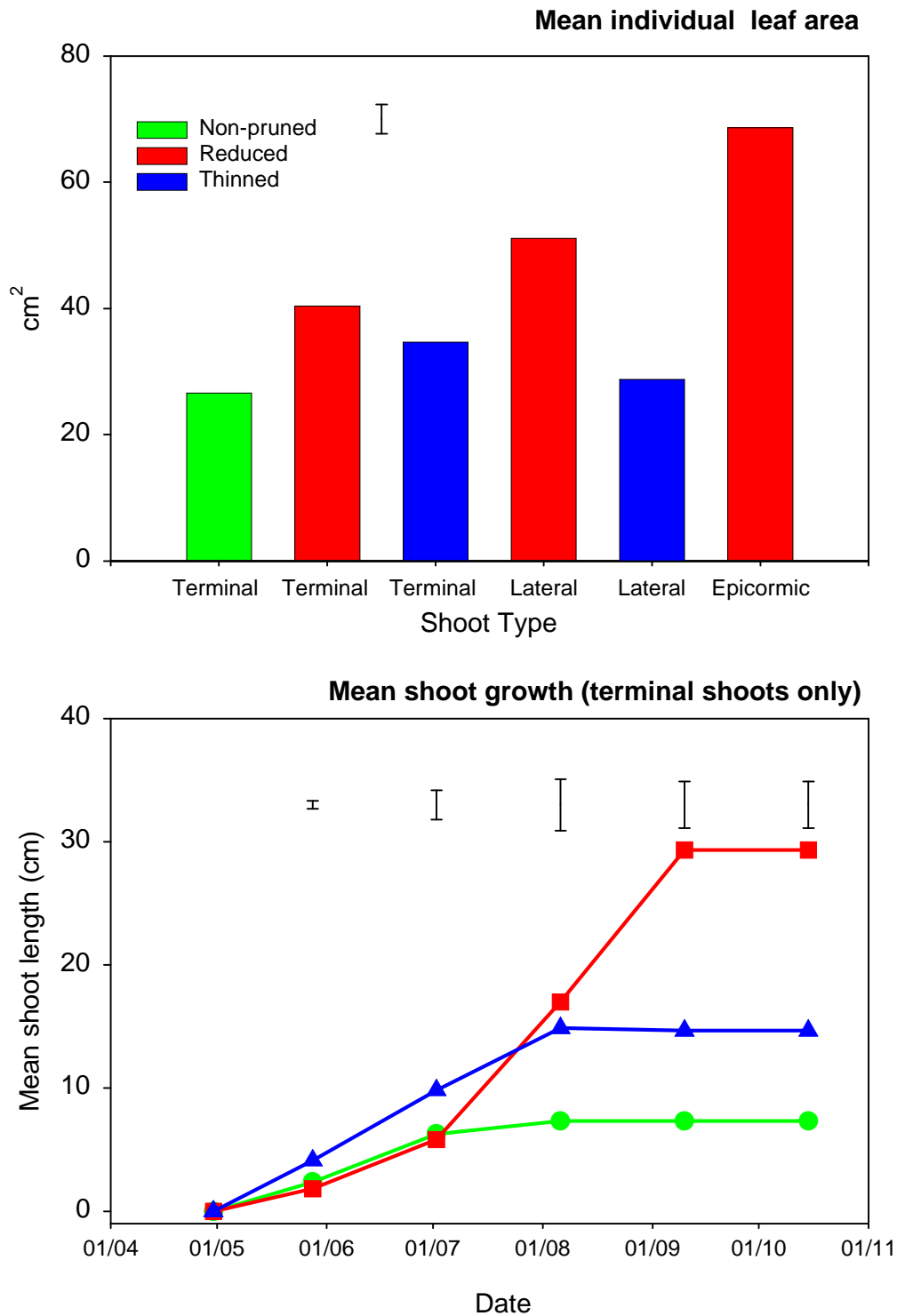
Figure 3.7: Leaf area density, drip line leaf area index and total canopy leaf area determined by digital analysis of hemispherical images for wild cherry trees pruned in April 2002 and soil droughted in 2002 and 2003 for the period 2002-2003 (Experiment 4). Vertical bars are standard error of differences between means

Figure 3.8: Final mean shoot length, total shoot leaf area and number of leaves per shoot of wild cherry trees pruned in April 2002 and soil droughted in 2002 and 2003 (Experiment 4). Vertical bars are standard error of differences between the means of pruning treatment for comparison within droughted and non-droughted treatments respectively

Controlling water use of trees to alleviate subsidence risk
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Figure 3.9: Mean size, specific weight, and nitrogen content of leaves of wild cherry trees pruned in April 2002 and soil droughted in 2002 and 2003 (Experiment 4). Vertical bars are the standard error of each mean, analysed individually

Figure 3.10: The average daily sap flux (water use) meaned across drought treatments for wild cherry trees pruned in April 2002 and soil droughted in 2002 and 2003 for the period 2002-2003 (Experiment 4). Vertical bars are the standard error of differences between means
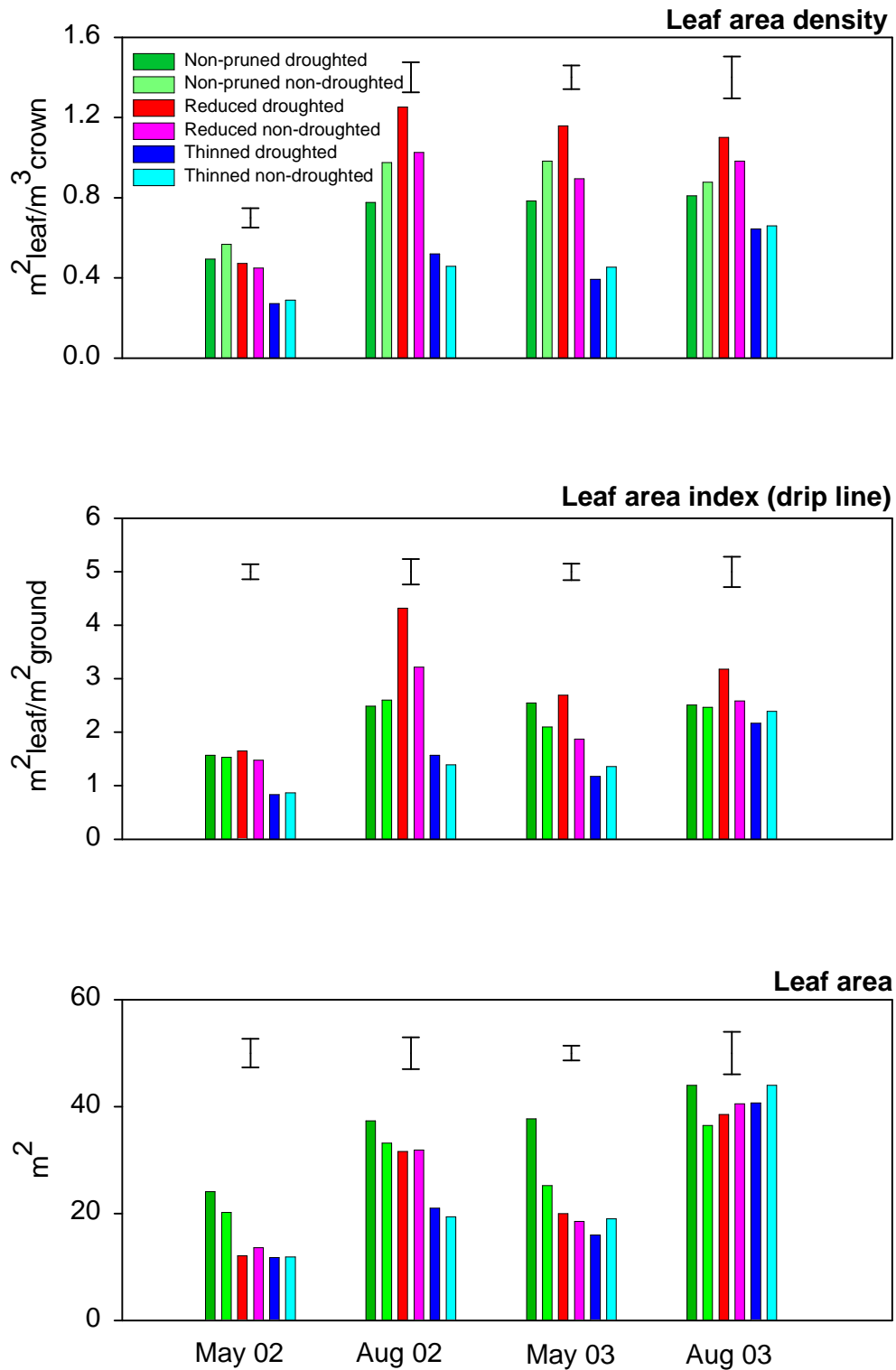
Figure 3.11: Leaf area density, drip line leaf area index and total canopy leaf area determined by digital analysis of hemispherical images for London plane trees pruned in February 2000 and severely pruned in April 2003 for the period 2000-2003 (Experiment 5). Vertical bars are standard error of differences between means

Figure 3.12: Leaf size, specific weight and nitrogen content of leaves of London plane trees pruned in February 2000 and severely pruned in April 2003 (Experiment 5). Vertical bars are the standard error of each mean, analysed individually
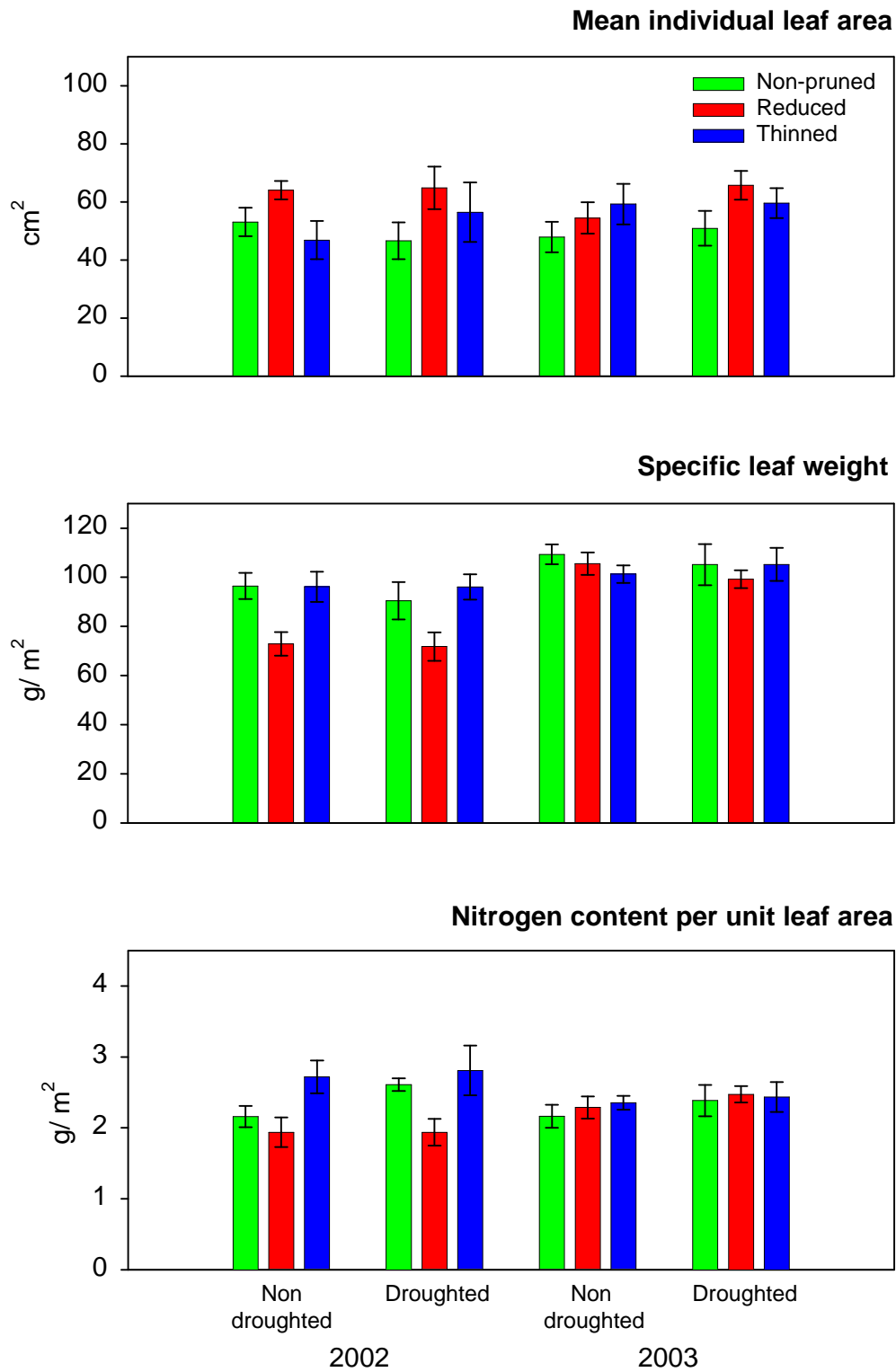
Mean individual leaf area

Specific leaf weight

Legend:
- Non-pruned
- Reduced 2000
- Thinned 2000
- Reduced 2000 & 2003

Nitrogen content per unit leaf area

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Figure 3.13: The average daily sap flux (water use) London plane trees pruned in February 2000 and severely pruned in April 2003 for the period 2001-2003 (Experiment 5). Vertical bars are the standard error of differences between means
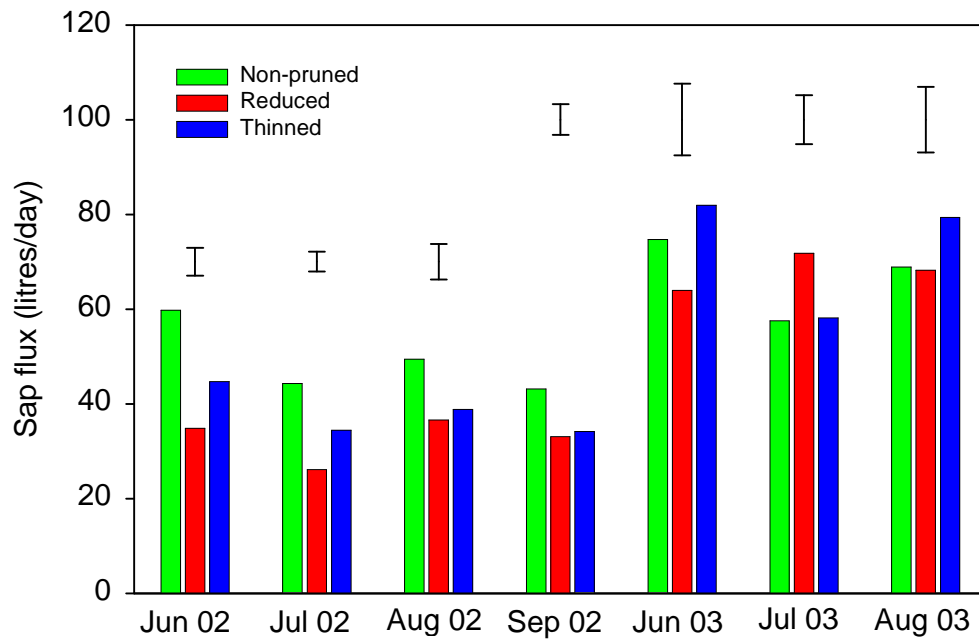
Figure 3.14: The effect of root restriction by different volumes of soil within geotextile lined pits on the growth of Norway maple and Lime trees over four growing season after planting in spring 2000. To stabilise variance the data were $\log_e$ transformed. Vertical bars are the standard error of differences between means

Objective 4:

Determine the influence that changes in the environment have in the control of water use in isolated trees, and develop an understanding that enables responses to canopy manipulation of different species to be predicted

Integration of climate, pruning and tree water use (leaf water relations)

<u>Long term impact of crown pruning and environment on tree water use (Experiment 2)</u>

Crown-reduction and crown-thinning applied to wild cherry trees in 2000 and repeated in 2002

Leaf water relations

Methods

Gas exchange measurements were made using a CIRAS-1 differential CO_2/H_2O infrared gas analyser with a broadleaf automatic Pa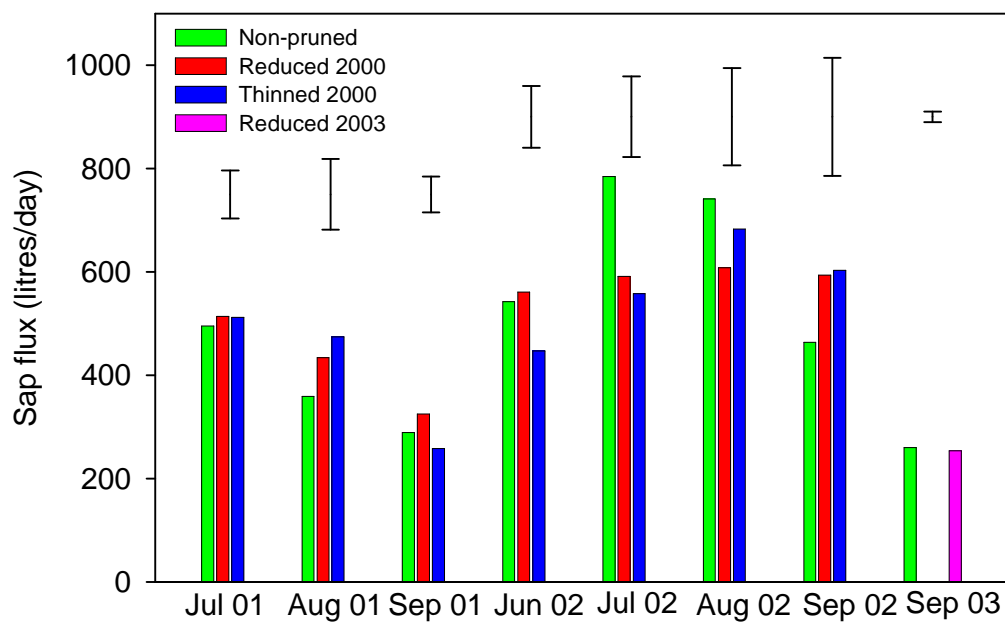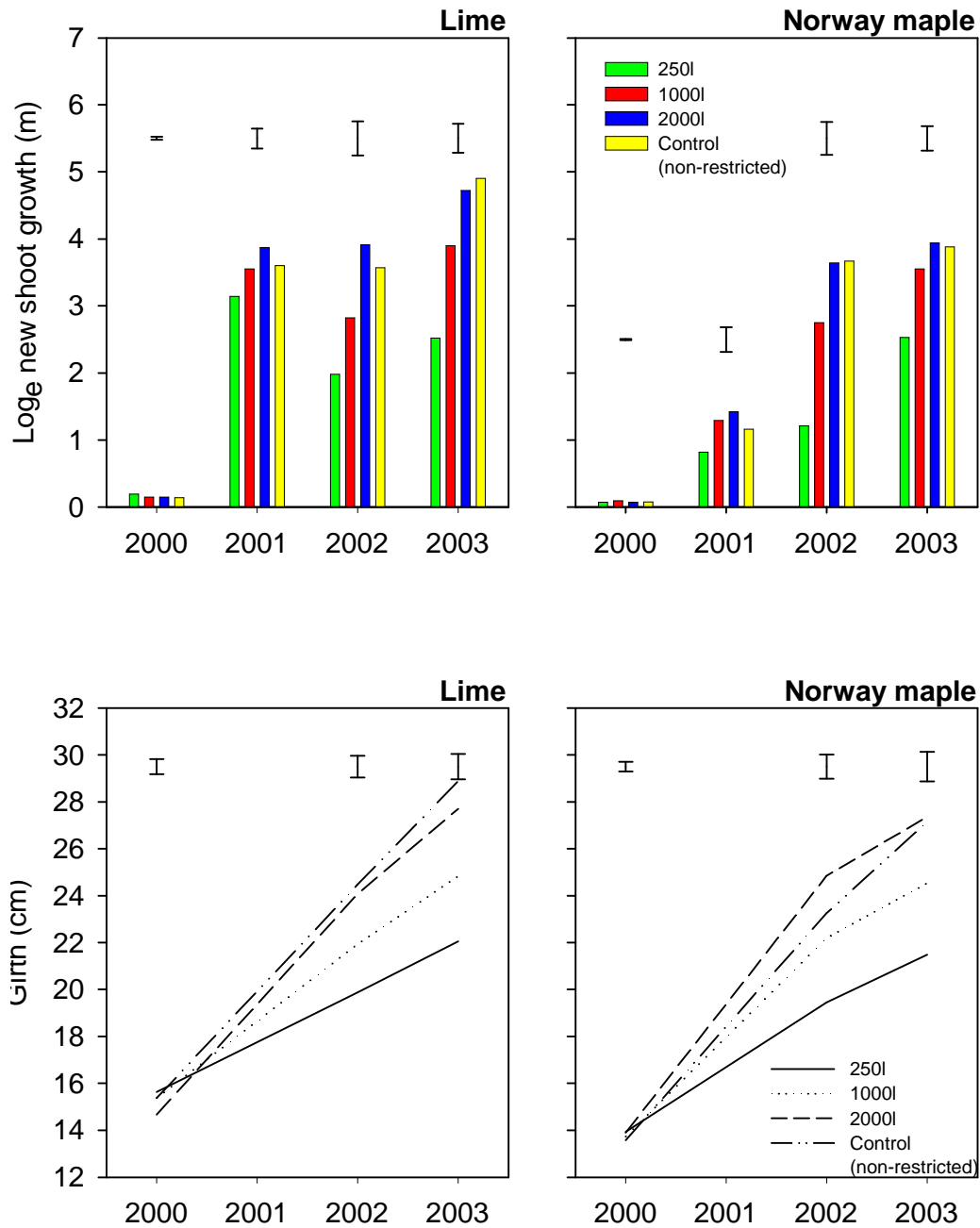rkinson leaf cuvette (2.5 cm^{-2}) (PP Systems Ltd, Hertfordshire, UK). On 15 August 2000 gas exchange was measured on three leaves per tree from three trees per treatment, using a broad leaf cuvette under ambient light and ambient CO_2 concentrations. On 28 August 2002 leaf gas exchange was measured using saturating light (with an artificial light source to illuminate the leaf cuvette at an intensity known to induce maximum photosynthesis, i.e. 750 μmol m^{-2} s^{-1}) and at ambient CO_2 concentrations, on three leaves per tree from three trees per treatment.

A pressure Scholander type chamber (Chas W. Cook & Sons Ltd, Birmingham, UK, or Skye Instruments, Llandrindod Wells, Powys, Wales) was used for measuring leaf water potential (Ψ) and a steady-state, continuous flow porometer (PP Systems Ltd, Hertfordshire, UK) was used to determine leaf conductance to water vapour. Leaf water potential and conductances were measured over diurnals time courses on sunny days (19 June and 25 July 2000). Leaf conductance was measured on three fully exposed leaves per tree from 4 trees per treatment. All leaves were completely expanded and fourth or fifth from the growing shoot tip. Following removal from the tree, each leaf was immediately placed in a humid bag and its leaf water potential measured within 30 s.

Results and conclusions

When gas exchange was measured under ambient light, there were consistently higher rates of assimilation (net uptake of CO_2) and leaf conductance to water vapour in the leaves of the crown-reduced trees (Figure 4.1), but little difference in instantaneous water use efficiencies between the treatments. By the time of re-pruning in 2002, when measured under saturating light intensity, assimilation rates and stomatal conductances were similar for leaves on trees in all three treatments. The second pruning treatment again had the effect of increasing assimilation rate in the crown-reduced trees, although stomatal conductances were greater in leaves of both the crown-reduced and crown-thinned trees following re-pruning (Figure 4.1).

Leaf water potentials were generally unaffected by pruning treatment except at mid-day in July 2000 after the first pruning treatment, when the crown-reduced trees had higher leaf water potentials, (implying less drought stress) than either of the other two treatments. A typical diurnal time course of water relations is illustrated for June 2000 (Figure 4.2). Leaf conductance was greater throughout the day in the leaves of the crown-reduced trees.

- Both instantaneous and diurnal gas exchange measurements indicate potential for greater assimilation rates (i.e. photosynthesis and growth) and leaf conductance (potential to lose water) in the crown-reduced trees in the year following pruning.

- Crown-thinning had little impact on assimilation rates and decreased water use efficiency.

Boundary layer conductance

Methods

On 28 August 2000, four artificial leaf sensors were placed the canopy of one tree per treatment. The artificial leaf sensors were located at 50 cm intervals, along a two meter sensor boom at a height of 4 m above ground level. The sensor boom was orientated so as to form a transect of the canopy, running from the fringe towards the trunk at the centre. All artificial leaf sensors were connected to data loggers to measure and compare temperature differences between the evaporating surface and the surrounding air. Intermittently, the artificial leaf sensors were disconnected, removed from the canopy and weighed to give an estimate of the evaporation rate.

Results and conclusions

Light penetration (radiation) in the canopy was least for the crown-reduced trees, and most for crown-thinned trees, with the non-pruned tree intermediate between the other two treatments (Figure 4.3). Under such conditions, evaporation rate determined directly from water loss by the sensor was greatest in the crown-thinned trees, and least in the crown-reduced trees (Figure 4.3). The data from each artificial leaf sensor showed that boundary layer conductances was least in the crown-reduced trees and greatest in the crown-thinned trees.

- Crown-reduced trees are not well coupled to conditions in the bulk atmosphere external to the canopy.
- Evaporative demand within the canopy of crown-reduced trees was decreased due to less vapour pressure deficit due to less radiation penetration.
- Leaves of crown-reduced trees are therefore restricted in their water loss to factors outside the influence of the leaf (i.e. stomata).
- Leaves of crown-thinned trees, are well coupled to the conditions in the atmosphere external to the canopy (i.e. boundary layer conductance is greater than for non-pruned trees) causing higher rates of water loss.

Stable isotopes integrating ^{13}C and ^{18}O signals

Background

From the preliminary results, presented under Objective 1, it was shown that carbon isotope discrimination was greater in the crown-reduced trees. The gas exchange of leaves from crown-reduced trees, when measured in a well-stirred cuvette, also supported the notion of higher assimilation and transpiration (evaporation) rates. However, it seems *in situ* that the greater leaf boundary layers, found in the dense re-growth of the crown-reduced trees negates the anticipated effects of increased whole tree transpiration (daily sap flux) and soil drying. First, we shall explain ^{13}C isotope data in terms of gas exchange. Second we shall show how the ^{18}O signal in leaf water and leaf organic material may indicate higher transpiration rates in crown-thinned trees of wild cherry and London plane trees.

Methods

Leaves for carbon isotope analysis were collected on 3 October 2000, 20 September 2001 and 13 August 2002. On each date, three leaves were collected from the north side of the canopy of three trees per treatment. Leaf samples used for oxygen isotope analysis of leaf water were collected on 26 July 2000, 28 August 2001 and 13 August 2002. On each date four leaves were collected from the north side of the canopy of three trees per treatment. Leaves for determination of oxygen isotope composition of leaf organic matter were collected on 10 October 2000. Three leaves were collected from the north of the canopy of three trees per treatment. All leaves were fully expanded and fourth or fifth from the growing shoot tip.

Results and conclusions

The pattern of increased carbon isotope discrimination in crown-reduced trees compared to non-pruned and crown-thinned trees was repeated for the first two years after pruning in 2000 (Figure 4.4). This implies that during the life of the leaf the stomatal apertures remained wider for the leaves of the crown-reduced trees than for the leaves of trees in the other treatments. Following the re-pruning of the trees in 2002, carbon isotope discrimination in leaves of crown-reduced trees was again higher than for the re-thinned trees (Figure 4.4). Thus, higher ^{13}C discrimination was not associated with higher rates of water use, but the opening of stomata to enhance CO_2 uptake when gas exchange was limited by the boundary layer. This is similar to shaded leaves growing deep within a forest canopy. However, in the first growing season after pruning, evaporative enrichment of ^{18}O, seen in both leaf water and organic material (Figure 4.4) were greater in the leaves of crown-thinned trees than the crown-reduced trees. Following re-pruning, there was again a tendency for greater evaporative enrichment to be associated with the leaves of the re-thinned trees.

- The high carbon isotope discrimination seen in leaves of crown-reduced trees was related to low leaf boundary layer conductance. Stomata opened wide to maximise CO_2 uptake and allow ^{13}C discrimination to be expressed, but water loss was largely uncoupled from this process due to a small vapour pressure deficit than for non-pruned or crown-thinned trees.
- Evaporative enrichment of ^{18}O in leaf water was a more consistent estimate for overall leaf transpiration rate for crown-thinned trees.

Impact of drought on response to crown pruning (Experiment 4)

Crown-reduction and crown-thinning applied to wild cherry trees in 2002 and soil drought applied in 2002 and 2003

Leaf water relations

Methods

Leaf water potential was measured on four days during the summer 2002 to provide an estimate of tree water status. Leaf water potential was measured mid-day (i.e. when the trees experience maximum drought stress) on five occasions between June and August 2003.

Results and conclusions

In the year of pruning, on 22 July and 30 August 2002 pre-dawn leaf water potential of the non-pruned trees were lower than those of trees in either of the pruning treatments averaged across drought treatments (Figure 4.5). These differences were significant (P=0.108 and P=0.008 for each date respectively). This suggests that the pruned trees were less drought stressed and more soil water was available for their roots.

On all of the measurement days the leaf water potentials at midday for the crown-reduced and crown-thinned trees were higher (i.e. less negative) than for non-pruned trees. However, the leaf conductances (Figure 4.5) of the trees averaged across drought treatments were similar for all pruned and non-pruned trees. Therefore, factors other than leaf conductance were influencing water loss. A significant interaction ($P<0.01$) occurred between drought stress and pruning treatments on 19 June. The leaf conductances of crown-reduced trees in the droughted soil were greatest, whereas for those in the non-droughted soil the opposite was true. Surprisingly, the drought itself had no effect on the leaf conductance or leaf water potential (when averaged across pruning treatments).

In the second year (2003), after pruning, the pruning treatments generally had no effect on mid-day or pre-dawn leaf water potentials. No evidence for residual effects of the previous year's pruning treatments was found for leaf conductances (data not presented). The drought stress treatment had no effect ($P>0.05$) on leaf water potential of any trees on any occasion compared to the non-drought stressed trees (data not presented). Since the dry weather during July and August caused rapid drying of the soil (see Figure 2.3), it is not surprising that these trees expressed similar levels of drought stress. The pre-dawn leaf water potentials (which are estimates for soil water availability to roots) measured in July and August confirmed that soil moisture conditions were equally limiting for the drought-stressed and non-drought stressed trees.

- In the year of pruning, leaves of the non-pruned trees showed lower leaf water potentials than those of either pruning treatments indicating they were more drought-stressed.
- Soil drought applied in this experiment had no effect on the leaf conductance or leaf water potential.
- The effects of crown-reduction and crown-thinning on leaf water relations had disappeared by the second year after pruning.

Boundary layer conductance

Methods

In 2002 measurements of boundary layer conductance were made using a revised version of the artificial leaf sensor described in section 1. These sensors included a modified artificial leaf based on realistic leaf shape and size taken from wild cherry trees. The new leaf component had a much greater surface area than that used in 2000, giving a more sensitive estimate of boundary layer conductance. Measurements were made on 26 June, 18, 19 July and 13 September. On each date four artificial leaves were arranged at 50 cm intervals along a 2 m sensor boom running through the northern fringe of the canopy, at a height of 4 m. All measurements were made simultaneously on one tree per pruning treatment.

The same procedure was repeated on 5, 12 and 16 June, 8, 9 and 14 July 2003. From 5 August the artificial leaf sensors were rearranged so that the sensor boom was orientated so as to form a transept of the canopy, running from the fringe towards the trunk at the centre. Measurements were made on 5, 6, 8, 14, 20 and 21 August 2003. The artificial leaf sensors were connected to data loggers to record temperature differences between the evaporating surface and the surrounding air. Periodically, the artificial leaf sensors were disconnected, removed from the canopy and weighed to give an estimate of the evaporation rate throughout the day.

Results and conclusions

Pruning led to significantly higher leaf boundary layer conductance in crown-thinned trees on three of the four measurement days. Leaf boundary layer conductance decreased in the crown-reduced trees relative to non-pruned trees (Figure 4.6). These findings confirmed the results found from the other wild cherry experiments.

Rates of evaporation from the artificial leaf component on the evaposensor illustrate the effect that leaf and canopy boundary layers have on canopy water loss (Figure 4.6). The fastest rates of evaporation occurred within the canopy of crown-thinned trees and the slowest within the canopy of crown-reduced trees. In the second year after pruning, there was no effect of crown-reduction or crown thinning on leaf boundary layer conductance or evaporation rates (data not presented).

- The fastest rate of evaporation occurred from leaves within the canopy of crown-thinned trees and the slowest within the canopy of the crown-reduced trees.
- The decrease in leaf boundary layer conductance in the canopy of crown-reduced trees meant that for any stomatal aperture, water loss was less than that of leaves in the canopies of crown-thinned trees.
- The effects of crown-reduction and crown-thinning on boundary layer conductance were limited to the year after pruning.

Gas exchange

Methods

Leaf gas exchange was measured using saturating light and ambient CO_2 concentrations. Four leaves per tree from four trees per pruning treatment were measured on 2 August and 19 September 2002, 14 June and 14 August 2003.

Results and conclusions

The pruning treatments did not influence assimilation rates and water use efficiency of leaves in the canopies of trees in either of the two years after pruning (data not presented).

Stable isotopes integrating ^{13}C and ^{18}O signals

Methods

Leaves for carbon isotope analysis were collected on 21 July 2002 and 21 August 2003. On each date three leaves were collected from the north side of the canopy of three trees per treatment. Leaves for oxygen isotope analysis of leaf water were collected on 13 September 2002, 12 June 2002 and 9 July 2003. On each date four leaves were collected from the north side of the canopy of three trees per pruning treatment. All leaves were fully expanded and were the 4th or 5th leaf back from the growing tip.

Results and conclusions

Carbon isotope discrimination followed the expected pattern for the first year after pruning in 2002, as both non-droughted and droughted crown-reduced wild cherry trees had higher discrimination (Figure 4.7). However, in the second year after pruning, leaves of both non-pruned and crown-thinned trees had higher ^{13}C discrimination than in the previous year, suggesting that stomatal aperture was better coupled to overall evaporative demand.

Crown-thinned trees had greater evaporative enrichment of ^{18}O in leaf water, in September 2002 and June and July 2003 (Figure 4.7) suggesting greater water loss (transpiration) from the leaves; this concurred with measurements of water loss from artificial leaves. However, other factors may also influence leaf water isotope enrichment. If the differences in humidity (leaf to air) are similar within the contrasting canopies (and the boundary layer effect is less well marked) then evaporative enrichment may be lower in highly transpiring leaves since evaporative cooling will reduce the fractionation expressed (Barbour *et al.,* 2000, 2002). For this reason, the ^{18}O enrichment data may vary with the extent of coupling within the canopies with similar relative humidities.

- Integrating water use determined over the life of the leaf suggests that leaf conductance was greater for leaves from the canopy of crown-reduced trees than crown-thinned or non-pruned trees.
- Crown-thinned trees had greater evaporative enrichment of ^{18}O in leaf water than non-pruned and crown-reduced trees, suggesting greater transpiration rates.

Assessing the impact of crown pruning on large amenity trees (Experiment 5)

Crown-reduction and crown thinning applied to mature London plane trees in 2000 and severe reduction applied in 2003

Leaf water relations

Methods

Leaf water relations were measured on 6 trees in three separate blocks during a diurnal time course on the 18 September 2003. Measurements were made on two or three fully expanded and exposed leaves per tree.

Results and conclusions

On 18 September 2003 (i.e. when the soil was at its driest), only a small non-significant (*P*>0.05) reduction in pre-dawn water potential of the trees crown reduced in 2003 was detected (Figure 4.8). This suggests that a similar amount of soil water was available irrespective of treatment, but is not in agreement with the soil drying patterns for this experiment (see Figure 2.4). The pre-dawn water potential values (approximately – 0.2 MPa) showed trees to be subject to little drought stress. Therefore, although the drying patterns of the crown-reduced and non-trees were quite different, the latter were able to access sufficient water to minimise drought stress.

The leaf water potentials at mid-day for the trees crown-reduced in 2003 were similar to those of the non-pruned trees. The trees crown-reduced in 2003, did not conserve their water by closing their stomata (Figure 4.8). This is clearly indicated by the large values (approximately 300 mmol/m^2/s indicates open stomata) in leaf conductance, whereas the non-pruned trees reduced their stomatal aperture (indicated by low values < 100 mmol/m^2/s). These differences between the treatments were significant (*P*<0.05). This is consistent with initial post-pruning effects found in previous years for the other experiments on cherry trees.

- Leaf conductance of leaves from crown-reduced trees was greater than those from crown-thinned and non-pruned trees.
- Crown-reduced trees did not conserve water by reducing leaf conductance mid-day, whereas the non-pruned trees did.
- Leaf water potentials (Ψ) of the trees were unaffected by pruning treatment.

Stable isotopes integrating ^{13}C and ^{18}O signals

Methods

Leaves for carbon isotope analysis were collected on 9 September 2000, 30 August 2001, 3 September 2002 and 11 September 2003. On each date three leaves were collected from the north side of the canopy of three trees per treatment. Leaf samples used for oxygen isotope analysis of leaf water were collected on the same dates. Three leaves were collected from the north of the canopy of three trees per treatment to determine oxygen isotope composition of bulk leaf matter on 9 September 2000. All leaves were fully expanded and fourth or fifth from the growing shoot tips.

Results and conclusions

The London plane trees provided less clear ^{13}C discrimination signals than the wild cherry trees. The ^{13}C signal of crown-reduced trees was similar to that of the non-pruned trees (i.e. both were higher that the crown-thinned trees) in the first year after the 2002 pruning (Figure 4.9). It was not until the severe re-pruning in 2003 that a large shift in carbon isotope discrimination occurs, i.e. a 3 ‰ difference between non-pruned and re-reduced leaf signals.

The results for [18]O enrichment were consistent, as bulk leaf water more evaporative enrichment occurred in all years in the crown-thinned trees than for the crown-reduced trees. The organic signal [18]O data were also consistent with evaporative enrichment in crown-thinned trees when sampled in 2000.

- Interactions between carbon isotope discrimination, and oxygen isotope composition of leaf water and leaf organic material can be used to distinguish between the effects of crown pruning treatments on evaporation from leaves and leaf conductance.
- Carbon isotope discrimination was uncoupled from stomatal limitation of photosynthesis in crown-reduced trees. Leaf conductance increased to enhance the diffusive supply of CO_2 in the shaded environment, of the dense canopy.
- Oxygen isotopes in leaf water or organic material provide a way to reveal the extent of evaporation at the leaf and canopy level for leaves exposed to more radiation, particularly in the crown-thinned trees.
- The use of ^{13}C and [18]O to distinguish carbon limitation and water loss is applicable to both wild cherry and London plane trees.

Weather at East Malling 1999-2003

Weather conditions are summarised in Figure 4.10

1999

The fifty-year average rainfall for this site for the period between April and September is 302 mm; in 1999 the rainfall for the same period was 284 mm, thus 18 mm below average. June and August were wetter than average and July was much drier. The theoretical soil moisture deficit can be calculated by subtracting actual rainfall from the Penman's theoretical calculated evaporation (i.e. moisture loss from the ground vegetation). During 1999, the accumulated deficit declined rapidly between May and July to reach a minimum around 250 mm in August.

2000

Weather conditions in 2000 were exceptional. Rainfall in April and May was more than double the fifty-year average. Total solar radiation generally was lower in 2000 than in 1999, particularly in July.

The calculated soil moisture deficit did not start to accumulate until early June and reached a minimum of only 200 mm in August. For a large part of the growing season adequate water supplies were available for the trees and the climatic conditions that drive water loss from the leaves (transpiration) into the surrounding atmosphere were less severe than in the previous year.

2001

Although rainfall was exceptionally high during the winter between April and September the rainfall was only 36 mm greater than the fifty-year average. Solar radiation levels were similar to those of 1999 and greater than 2000, monthly rainfall was also similar to 1999 and less than 2000. The accumulated deficit followed a similar pattern to that in 1999.

2002

Rainfall was 22 mm greater than the fifty-year average. for the April-September period. Solar radiation levels were similar to those of 2000 and less than 1999 and 2001, monthly rainfall was also similar to 2001. The accumulated soil moisture deficit followed a similar pattern to that found in 2000, but reached a minimum of only 179 mm. The deficit was less severe than in previous years due to a combination of near average rainfall and a high proportion of dull days that reduced the potential for high evaporation.

2003

The weather during the growing season was much drier than average. Rainfall was only two-thirds of the fifty-year average. The driest months were July, August and September. Solar radiation levels were generally as great, or greater than, those of previous years and September was particularly sunny. Average monthly temperatures were 1.5-3.1°C greater than the 40 year average during June, July and August (data not presented).

In 2003 the calculated deficit followed a similar pattern to the 1999 and 2001 until the end of July, but it continued to decline during August and September to a minimum of 368 mm. Thus, during the course of this project, 2003 was the year in which the trees were subjected to the most drought stressed conditions.

Figure 4.1: Maximum assimilation rate, leaf conductance and water use efficiency for wild cherry trees pruned in March 2000 and re-pruned in April 2002 (Experiment 2). Vertical bars are standard error of each mean, analysed individually

Figure 4.2: Diurnal measurements of leaf water potential, leaf conductance, light levels (PAR) and vapour pressure deficit (VPD) on 20 June 2000 for wild cherry trees pruned in March 2000 (Experiment 2). Vertical bars are standard error of each mean, analysed individually

Figure 4.3: Photosynthetically active radiation (PAR), rate of evaporation and boundary layer conductance determined on 28 August 2000 within the canopy for wild cherry trees pruned in March 2000 (Experiment 2). Artificial leaves (E1…E4) where arranged along a 2m sensor boon which was orientated to form a transept, running from the canopy fringe (E4) towards the trunk (E1)

Figure 4.4: Carbon isotope discrimination in bulk leaf matter, oxygen isotope composition of leaf water and oxygen isotope composition of leaf organic matter for wild cherry trees pruned in March 2000 and re-pruned in April 2002 (Experiment 2). Oxygen isotope in leaf organic matter was measured in 2000 only. Vertical bars are standard error of each mean, analysed individually

Carbon isotope discrimination

Oxygen isotope composition of bulk leaf water
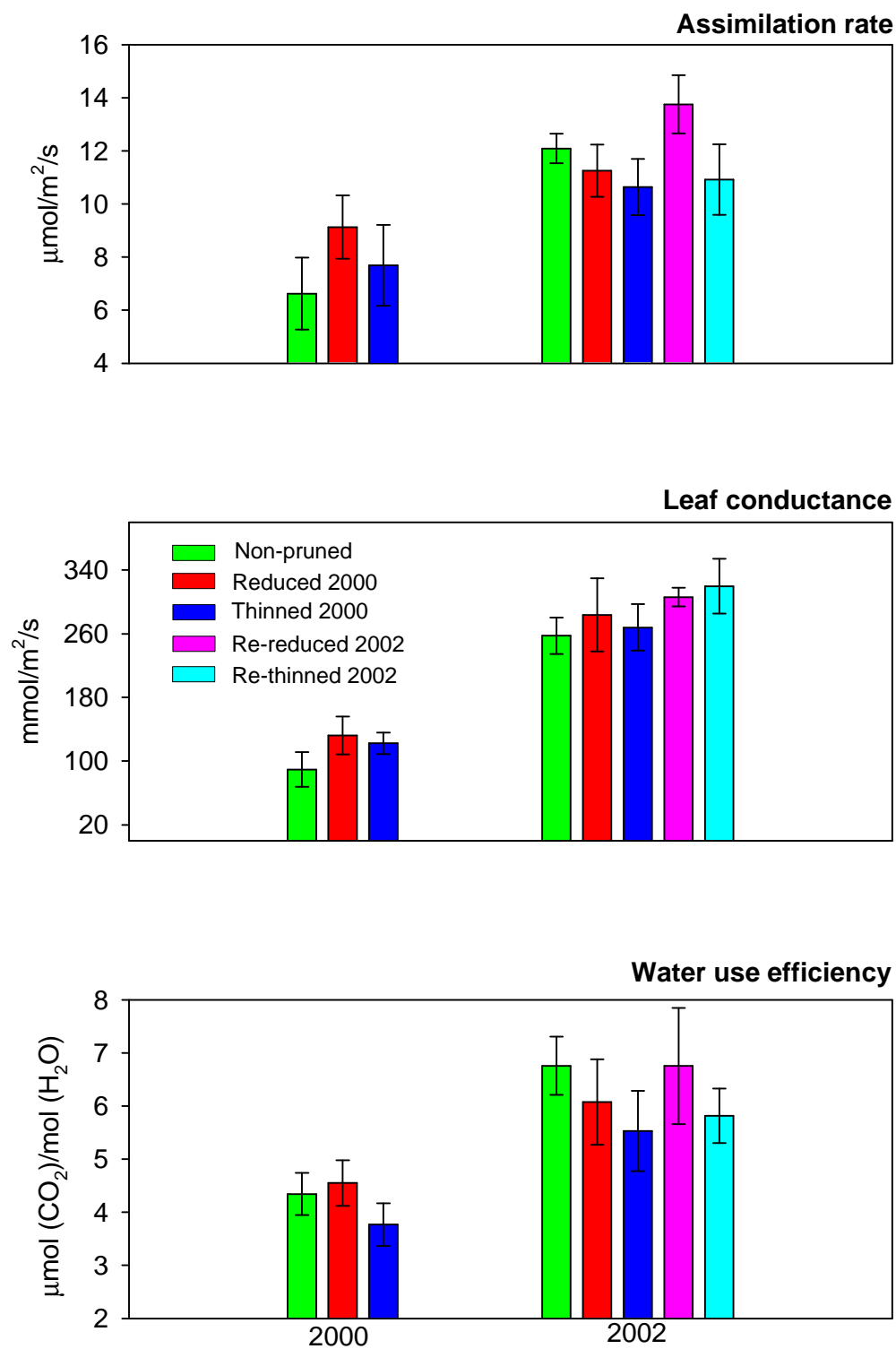
Oxygen isotope composition of leaf organic materal

Figure 4.5: Diurnal measurements of leaf water potential leaf conductance, light levels (PAR) and vapour pressure deficit (VPD) on wild cherry trees pruned in April 2002 and soil droughted in 2002 (Experiment 4). Vertical bars are standard errors of differences between the means

Figure 4.6: Boundary layer conductance and evaporation rate for wild cherry trees pruned in April 2002 (Experiment 4). Vertical bars are standard error of each mean, analysed individually
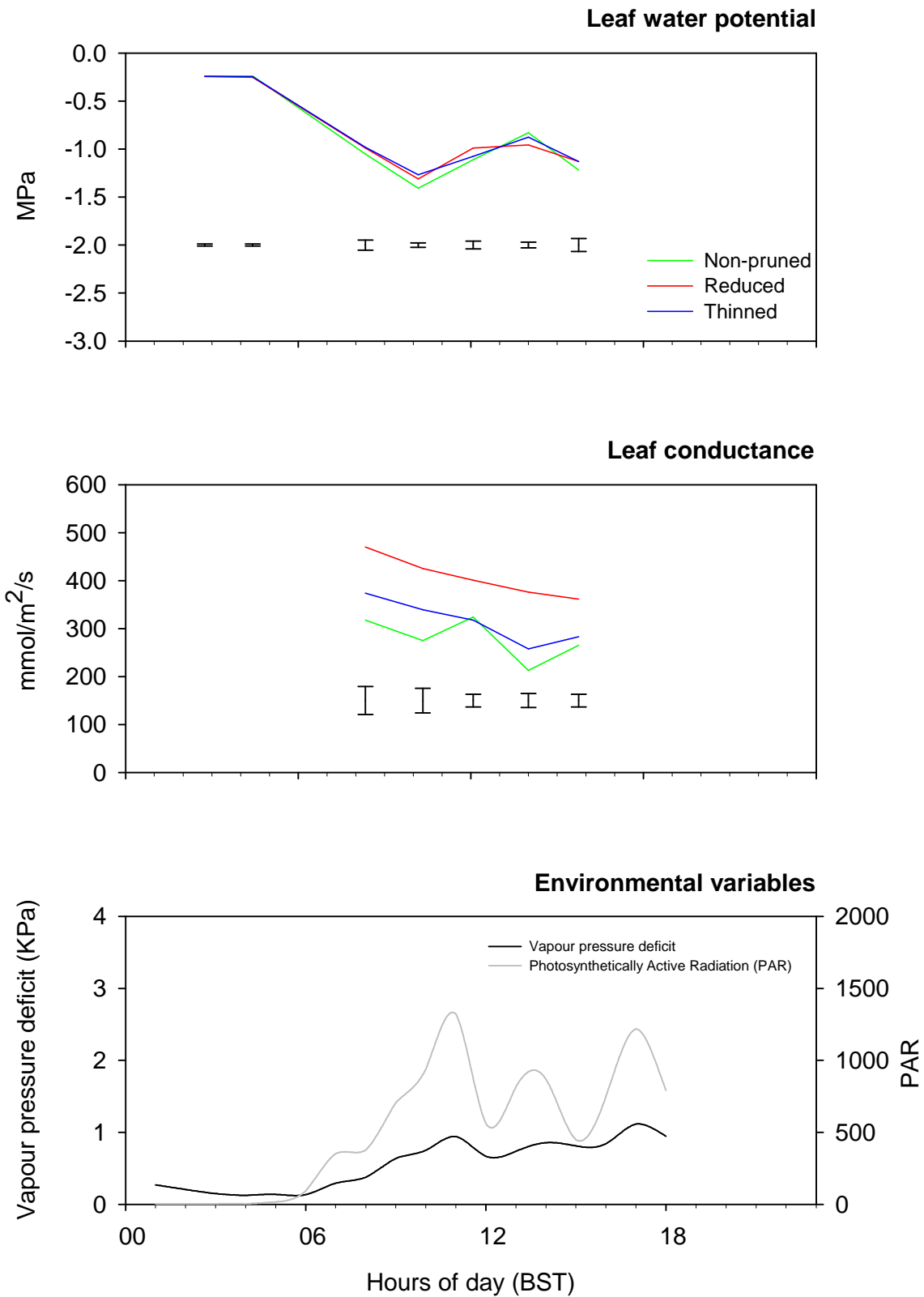
Figure 4.7: Carbon isotope discrimination in bulk leaf matter and oxygen isotope composition of bulk leaf water for cherry trees pruned in April 2002 (Experiment 4). Vertical bars are standard error of each mean, analysed individually

Figure 4.8: Diurnal measurements of leaf water potential, leaf conductance and light levels (PAR) and vapour pressure deficit (VPD) on London plane trees severely crown-reduced in April 2003 (Experiment 5). Vertical bars are standard errors of differences between means
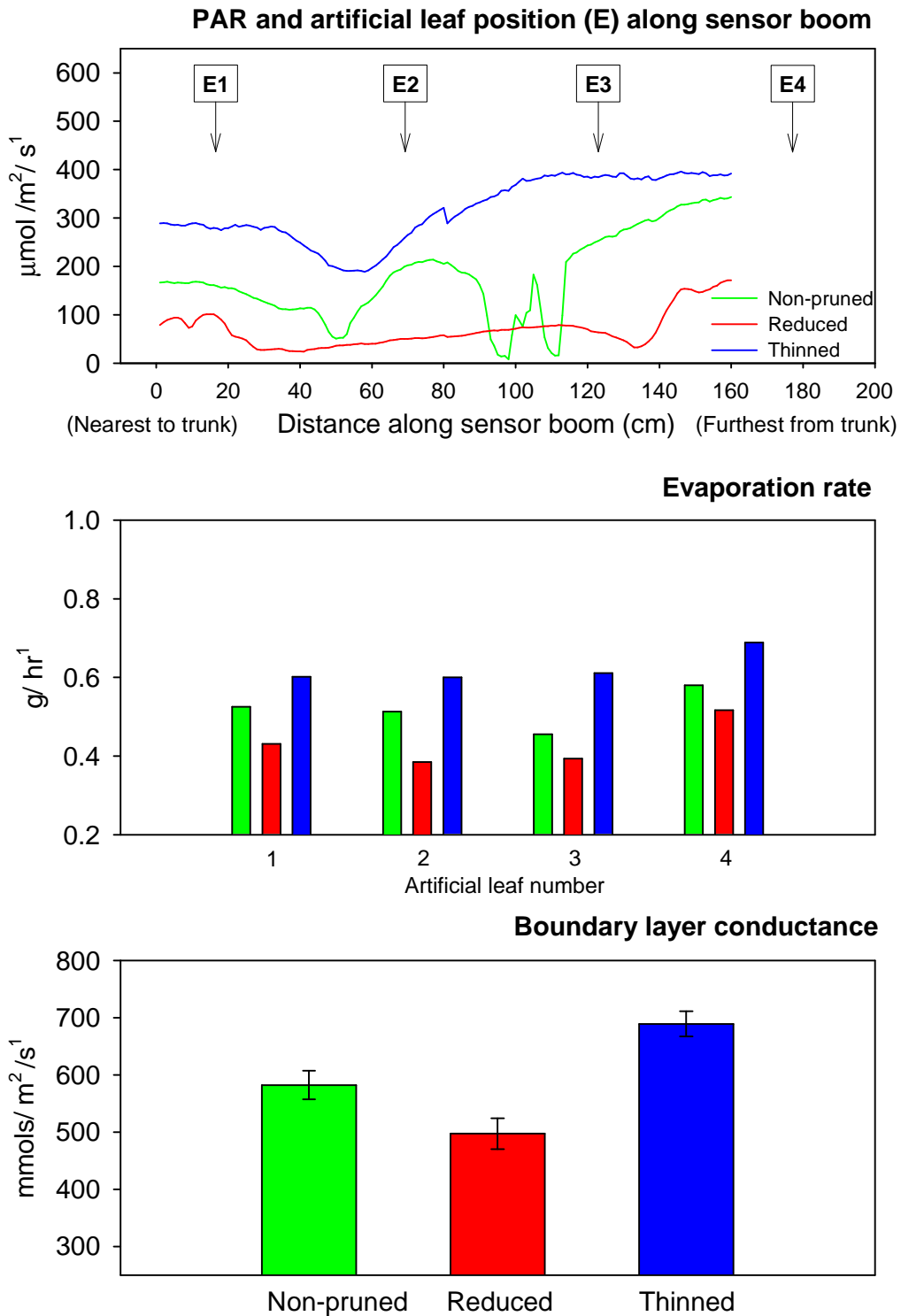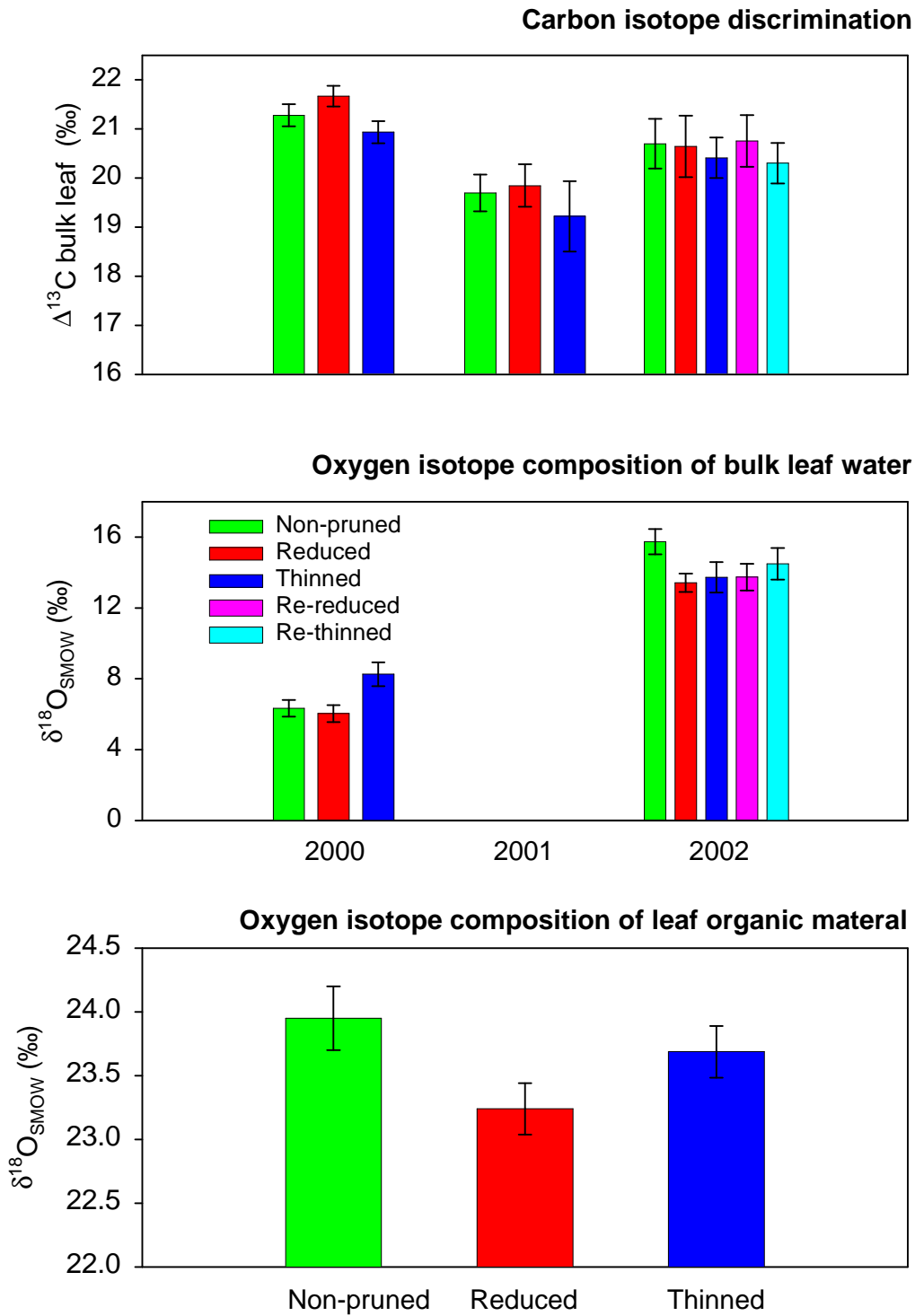
Figure 4.9: Carbon isotope discrimination in bulk leaf matter, oxygen isotope composition of bulk leaf water and oxygen isotope composition of bulk leaf matter for London plane trees pruned in February 2000 and re-pruned in April 2003 (Experiment 5). Oxygen isotope composition of bulk leaf matter was measured in 2000 only. Vertical bars are standard error of each mean, analysed individually

Figure 4.10: Weather conditions at East Malling during the period 1999-2003

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Summary of outcome against milestones

Primary milestones are in bold

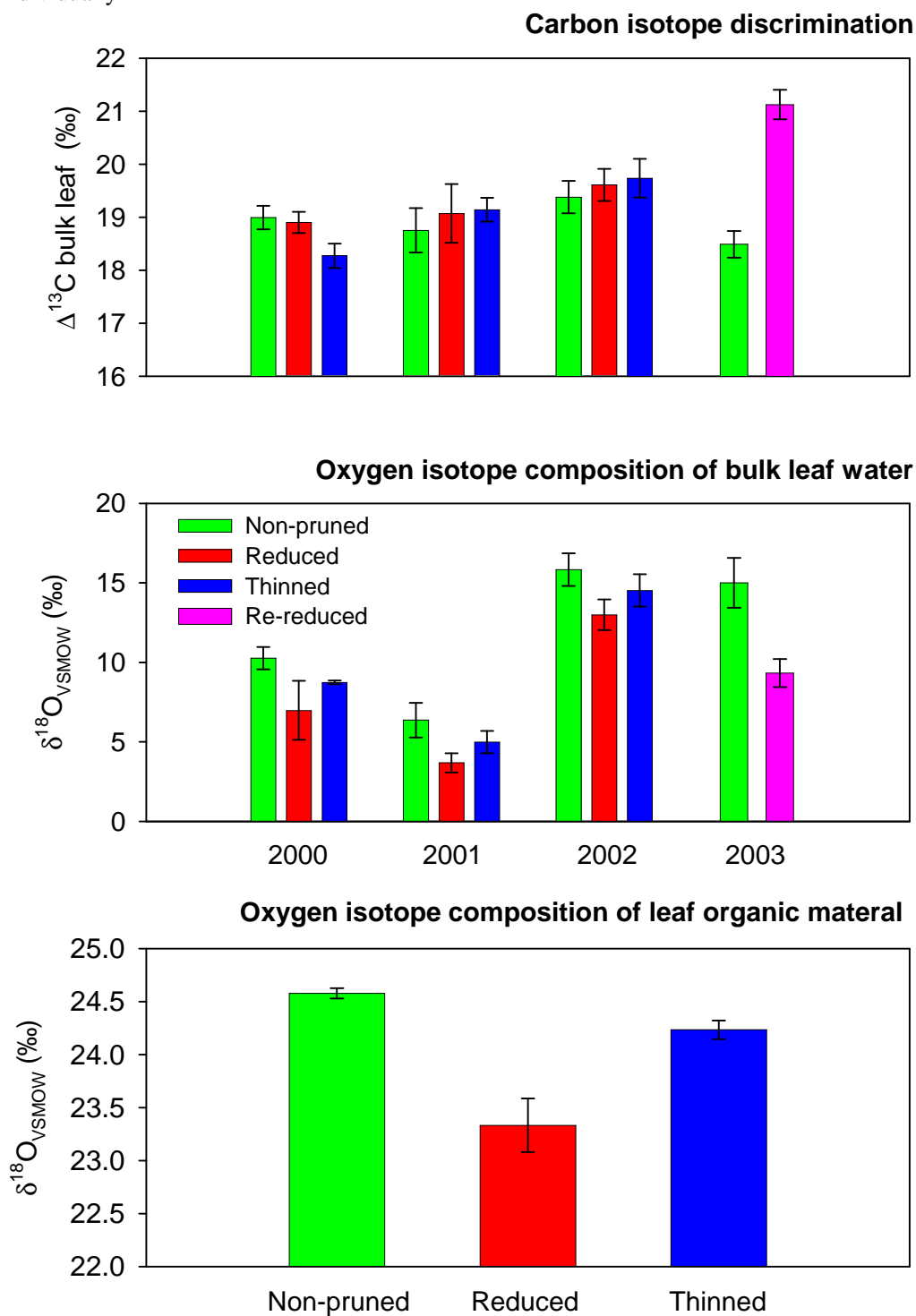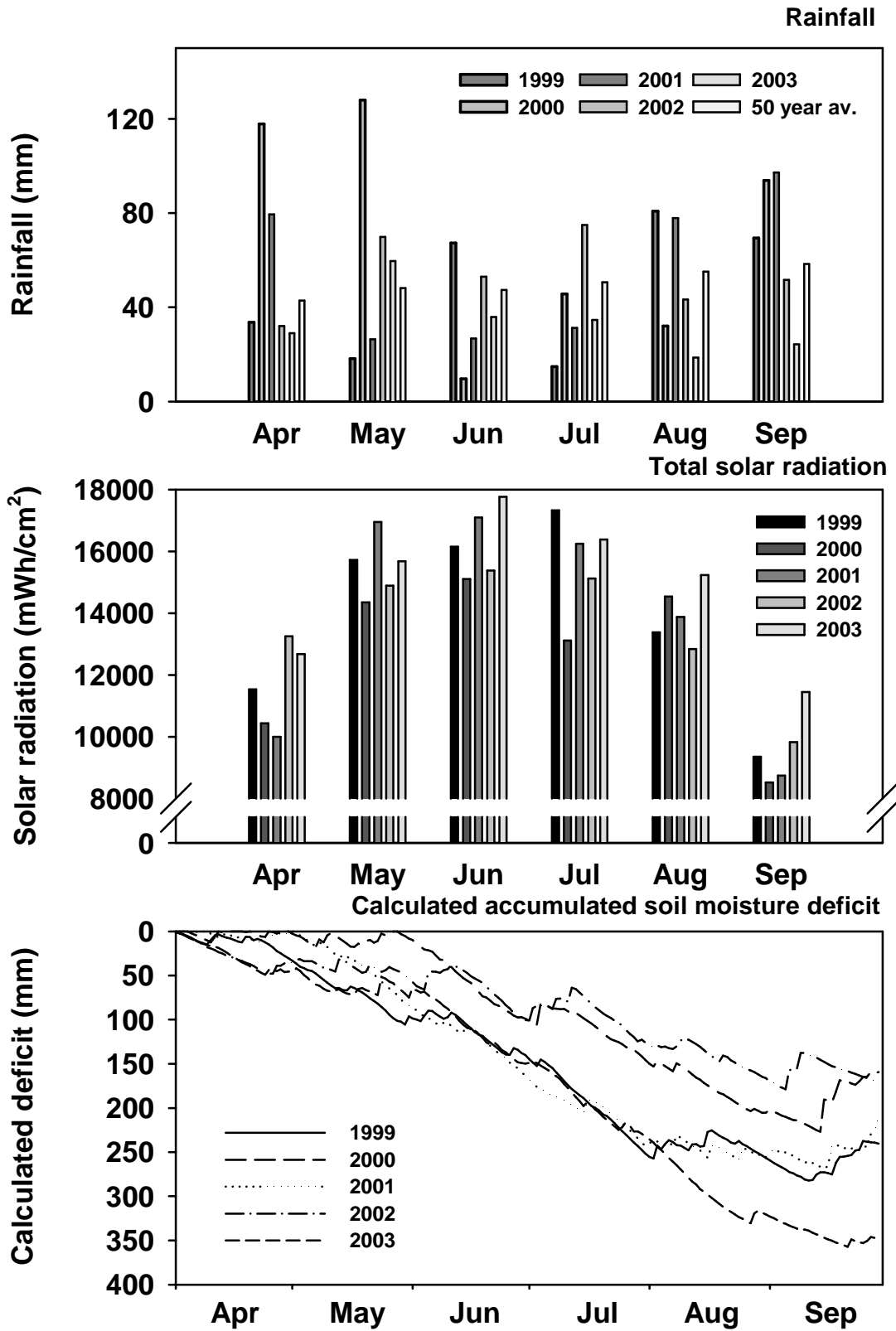| Task	Milestone	Date due	Status	Comment
	Objective 1			
1.1	Assess procedures for measuring leaf water use (cherry trees)	08/12/99	Achieved	Experiment 1
1.2	Assess procedures for measuring tree water use (cherry trees)	08/12/99	Achieved	
1.3	Assess methods to determine canopy size (cherry trees)	08/12/99	Achieved	
1.4	Summarise results from tree canopy area determinations	19/01/00	Achieved	
1.5	**Deliver results report for tasks 1.1 -1.4**	**19/01/00**	Achieved	Experiment 1
1.6	Develop protocols to scale from leaf to canopy	11/02/00	Achieved	Experiment 1
1.7	**Appropriate methodology identified**	**27/03/00**	Achieved	Experiment 1
	Objective 2			
2.1	Determine protocols for isotopic analysis (cherry and London plane trees)	28/04/00	Achieved	Experiments 1, 2, 5
2.2	Collect samples for ^{18}O isotope analysis (cherry and London plane trees)	15/09/00	Achieved	^{13}C also measured in leaf samples
2.3	Complete soil drying and ^{13}C and ^{18}O stable isotope ratios measurements (cherry and London plane trees)	08/12/00	Achieved	Experiments 1, 2, 5
2.4	Interpret tree soil water use patterns using isotopic data and soil moisture data	19/01/01	Achieved	Experiments 1, 2, 5
2.5	**Determine suitability of natural isotopes to measure water use**	**19/01/01**	Achieved	Natutal isotpes found to be unsuitable for determining location of water in profile (Experiments 1, 2)
2.6	Apply labelled isotopic water to soil (cherry and London plane trees)	27/04/01	Not achieved	Procedure for measuring evaporative enrichment in leaf organic material and bulk water adopted instead (Experiment 2, 4)
2.7	Collect samples for ^{18}O isotope analysis (cherry and London plane trees)	14/09/01	Achieved	Bulk leaf water and leaf organic material analysed (Experiments 2, 4)
2.8	Measure soil drying and ^{18}O isotopic ratios (cherry and London plane trees)	04/01/02	Achieved	^{13}C also measured in leaf samples (Experiments 2, 5)
2.9	Interpret tree soil water use patterns (cherry and London plane trees)	15/02/02	Achieved	Experiments 2, 5
2.10	Summarise results on soil water use experiments (cherry and London plane trees)	12/03/02	Achieved	Experiments 2, 5
2.11	**Deliver results report water use (cherry and London plane trees)**	**01/04/02**	Achieved	Experiments 2, 5
2.12	Collect samples for isotopic analysis (cherry x drought stress and London plane trees)	09/10/02	Achieved	Milestone revised, Experiments 4, 5
2.13	Summarise results of isotopic analysis (cherry x drought stress and London plane trees)	20/11/02		Experiments 4, 5

2.14	**Deliver results report water use (cherry x drought stress and London plane trees)**	**01/02/03**	Achieved	Revised milestone, results from experiment 2 supplied in addition
2.15	Collect samples for isotopic analysis (cherry x drought stress and London plane trees)	11/11/03	Achieved	Revised milestone (Experiments 4, 5
2.16	Summarize results soil water use patterns (cherry x drought stress and London plane trees)	09/12/03	Achieved	Revised milestone (Experiments 4, 5
2.17	Deliver results report (cherry x drought stress and London plane trees)	19/01/04	Achieved	Revised milestone (Experiments 4, 5
	Objective 3	30/12/03		
3.1	Develop and agree experimental plan with industry partners	21/04/99	Achieved	Experiments 1, 4
3.2	Apply crown pruning treatments (cherry trees)	30/06/99	Achieved	Experiment 1
3.3	Measure canopy growth (cherry trees)	29/02/00	Achieved	Experiment 1
3.4	Summarise growth responses (cherry trees)	11/04/00	Achieved	Experiment 1
3.5	**Produce analysis of growth results (cherry trees)**	**01/03/00**	Achieved	Experiment 1
3.6	Apply canopy reduction treatments (cherry trees)	08/03/00	Achieved	New cherry tree experiment set up (Experiment 2)
3.7	Apply canopy reduction treatments (London plane trees)	20/04/00	Achieved	Experiment 5
3.8	Measure canopy growth (cherry, London plane trees)	13/12/00	Achieved	Experiments 1, 2, 5
3.9	**Summarise canopy growth results data (cherry and London plane trees)**	**01/11/00**	Achieved	Experiments 1, 2, 5
3.10	Measure canopy growth response (cherry and London plane trees)	31/10/01	Achieved	Experiments 2, 5
3.11	**Summarise canopy growth results data (cherry and London plane trees)**	**03/09/01**	Achieved	Experiments 2, 3, 5
3.12	Measure crown pruning re-growth response (cherry and London plane trees)	18/10/02	Achieved	Experiments 2, 4, 5
3.13	Summarise canopy growth results (cherry and London plane trees)	04/11/02	Achieved	Experiments 2, 4, 5
3.14	Analyses crown growth (cherry and London plane trees)	29/11/02	Achieved	Experiments 2, 4, 5
3.15	**Deliver canopy growth results report (cherry and London plane trees)**	**14/11/02**	Achieved	Experiments 2, 4, 5
3.16	Locate site and plant oak, lime and Norway maple trees for pruning and root restriction experiments	24/03/00	Part achieved	Root restriction experiment set up, suitable clay soil site not found for pruning experiment, trees planted at East Malling (Experiment 6)
3.17	Finish initial measurements to determine tree size and shoot growth lime and Norway maple trees)	05/05/00	Achieved	Milestone revised (Experiment 6)
3.18	Determine annual growth (lime and Norway maple root restriction experiment)	20/02/01	Achieved	Experiment 6
3.19	Determine annual growth (lime and Norway maple root restriction experiment)	20/02/02	Achieved	Experiment 6
3.20	Apply crown pruning treatments (Oak and lime trees). Determine interaction between drought and crown pruning on cherry trees	16/04/02	Part achieved	See appended comment re 3.20 (Experiment 4)

Controlling water use of trees to alleviate subsidence risk

3.21	Measure canopy growth (Cherry drought experiment, lime and Norway maple root restriction)	01/10/02	Achieved	Measurements continued on London plane trees and cherry tree experiments in excess of milestone requirement (Experiments 2, 4, 5, 6)
3.22	Summarise growth results data (cherry x drought stress, lime and Norway maple root restriction)	10/12/02	Achieved	See comment 3.21
3.23	**Deliver canopy growth results report (cherry x drought stress, lime and Norway maple root restriction)**	**01/01/03**	Achieved	See comment 3.21
3.24	Measure crown reduction responses (cherry x drought stress, lime and Norway maple root restriction)	30/09/03	Achieved	See comment 3.21
3.25	Summarise canopy growth results data (cherry x drought stress, lime and Norway maple root restriction)	08/12/03	Achieved	See comment 3.21
3.26	**Deliver results report for canopy growth (cherry x drought stress, lime and Norway maple root restriction)**	**08/12/03**	Achieved	See comment 3.21
3.27	Measure water use in crown pruned trees (cherry and London plane)	10/11/00	Achieved	Experiments 1,2, 5
3.28	**Summarise water use results data (cherry and London plane trees)**	**10/11/00**	Achieved	Measurement period extended until December 2003 (Experiment 2, 5)
3.29	Measure leaf and canopy water use (cherry and London plane trees)	09/11/01	Achieved	Experiments 2, 5
3.30	**Summarise results data leaf and canopy water use (cherry and London plane trees)**	**09/11/01**	Achieved	Experiments 2, 5
3.31	Measure leaf and canopy water use (cherry x drought stress, London plane trees)	08/11/02	Achieved	Experiments 2, 4, 5
3.32	Summarise leaf and water use data	08/11/02	Achieved	Experiments 2, 4, 5
3.33	Measure leaf and canopy water use in (cherry x drought stress)	02/10/03	Achieved	Experiment 4, data provided from experiments 2 and 5 in addition
3.34	Summarise leaf and canopy water use results (cherry drought)	30/12/03	Achieved	Experiments 2, 4, 5
3.35	**Complete crown pruning experiments and data summary**	**30/12/03**	Achieved	Experiments 1,2, 3, 4, 5, 6
3.36	Initiate root restriction experiment (oak and lime)	23/02/00	Achieved	Norway maple substituted for industry considered it a more relevant species (Experiment 6)
3.37	Measure growth responses to root restriction (Norway maple and lime trees experiment)	23/07/03	Achieved	Experiment 6
3.38	Summarise growth responses to root restriction (Norway maple and lime trees experiment)	17/09/03	Achieved	Experiment 6
3.39	Deliver results report on impact and effectiveness of root restriction	28/11/03	Achieved	Experiment 6
	Objective 4			
4.1	Collect environmental data from East Malling through out project	14/05/03	Achieved	
4.2	Complete preliminary analysis of effects of crown pruning, on growth and water use	26/09/03	Achieved	Experiments 1, 2, 3, 4, 5
4.3	Establish the relationships between leaf and canopy water use and environmental factors for different species	21/11/03	Achieved	Integrated with results from objective 2 in this report

4.4	Complete integration of crown pruning effects on growth, water use and environmental data	20/01/04	Achieved	
4.5	**Prototype predictive approach developed relating canopy water use to climate**	**13/02/04**	Part achieved	Variability of sap flux measurements, but successful use of 'artificial leaves'
	Objective 5			
5.1	Integrate experimental results	08/12/03	Achieved	
5.2	Conduct a cost benefit analysis of crown control methods	24/10/03	Achieved	Data provided by Giles Biddle
5.3	Interact with industry to develop prevention protocols	16/03/04	On going	
5.4	Develop initial prevention and risk management guidelines	26/03/04	Achieved	
5.5	**Delivery preliminary guidelines for industry refinement**	**30/03/04**	Achieved	

Originally, milestones 3.21-3.26 related to the planting and utilisation of an experiment using oak and lime trees. Although the trees were planted on schedule, the growth of the trees was not large enough for experimental use. During the early phase of the project the weather was wetter than average. Therefore, with the agreement of the consortium a new experiment (No 4) was substituted for that planned for the oak and lime trees that used mature wild cherry trees and determined the interaction of soil drought and crown pruning techniques. The measurements carried out in this experiment were the same as those originally planned for the oak and lime tree experiment. In addition, the measurement period for the wild cherry and London plane experiments initiated in 2000 were extended to cover the full life of the project.

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Glossary of terms

Artificial leaf: An evaporative surface that may be placed in the canopy which is used to measure boundary layer conductance (g_b) and potential water loss.

Assimilation (A): See photosynthetic fixation.

Boundary layer: An unstirred layer of air at the leaf surface or within an isolated canopy which does not readily mix with the bulk air around the tree.

Boundary layer conductance (g_b): The homogeneity of the gaseous and thermal microenvironment around a leaf determines the diffusive limitation imposed by the boundary layer. The rate of transfer of water vapour between the leaf and the bulk atmosphere can be determined using a artificial leaf.

C:N This is ratio of carbon to nitrogen within plant organic matter.

^{12}C: The most abundant stable isotope of carbon comprising 98.9% of carbon.

^{13}C: The heavy stable isotope of carbon, comprising 1.1 of carbon.

C_a: External, ambient air (a) carbon dioxide (CO_2) concentration.

Canopy architecture: The structural arrangement of branches and shoots.

C_i: Intercellular (i) carbon dioxide concentration.

C_i/C_a: The ratio of internal (i) to external (a) CO_2 concentration, which represents the internal drawdown of CO_2 within a leaf and a control point for indicating water loss against CO_2 uptake.

Climatic factors (or variables): Meteorological variables such as temperature, humidity, wind speed and sunshine hours which influence growth rate and water use of plants.

Crown reduction: A pruning method that reduces crown volume, but allows the trees natural natural shape to be preserved. This involves an overall reduction of both height and spread by removing the outer portions of all major branches. The normal industry standard is to aim to reduce the canopy leaf area by 30% (BS3998: 1989, Lonsdale, 1999).

Crown thinning: A pruning method that reduces the number of side branches coming off all of the major branches not affecting the original volume of the tree crown. The normal industry standard is to aim to reduce the canopy leaf area by 30% (BS3998: 1989, Lonsdale, 1999).

Diffusion: The movement of gases. When diffusion occurs gas molecules, such as water vapour, move from areas of high concentration, such as inside the leaf, to areas of lower concentration, such as the air outside the leaf.

Diurnal: Measurements made during night and day.

Discrimination (D): See isotopic discrimination.

Field capacity: The water content found when a thoroughly wetted soil has drained for about two days. It is determined in the field during the winter period and is used a reference point against which to calculate the soil moisture deficit. It represents the maximum moisture content normally found in a particular soil.

Gas exchange: The rate of exchange of water vapour and carbon dioxide between the leaf and its surrounding atmosphere.

Growing season: The months in which trees actively grow (April to October).

Instantaneous water use efficiency (WUE$_i$): Determined as the instantaneous ratio of Assimilation / Evaporation (A/E), or from gas exchange measurements.

Isotope: The atomic number is determined by the number of protons in a nucleus, and for a given element different isotopes occur when the atomic mass varies because of additional neutrons. The majority (98.9%) of carbon atoms will have a mass of 12 (6 neutrons + 6 protons,) but a small number (1.1%) will have a mass of 13 (^{13}C) due to an additional neutron and are stable, as compared to the radioactive decay undertaken by the much smaller proportion of ^{14}C.

Isotopic ^{13}C analysis: Mass spectrometric analysis of CO_2 usually prepared from combusted leaf organic material.

Isotopic composition (d): The isotopic composition (δ) is the ratio of heavy to light stable isotopes within a substance relative to a standard. For carbon this would be a ratio of $^{13}C/^{12}C$ expressed relative to a standard with a known absolute isotopic composition which is arbitrarily set at 0. Ambient CO_2 (δ_p) is depleted in ^{13}C by a few part per thousand (‰) compared to the reference carbonate, and therefore have negative $^{13}\delta$ C values.

Isotopic discrimination (D): Because the heavy isotope is slower to diffuse and react, discrimination arises during diffusion and fixation by Ribose-bis-phosphate-caboxylase-oxygenase (Rubisco), and so biological material is depleted in ^{13}C compared to source CO_2. High discrimination leads to plant material more depleted in ^{13}C (more negative $^{13}\delta C$), particularly when stomata are open with a high conductance, Ci/Ca ratio and water use. Low discrimination (plant material less depleted in ^{13}C) might normally be found in leaves with low stomatal conductances and low water use. Carbon isotope discrimination (Δ) may be calculated as follows:

$$\Delta = (\delta_a - \delta_p)/(1 + \delta_p)$$

Where: Δ = discrimination, δ_a = isotopic composition of atmospheric CO_2, δ_p = isotopic composition of the plant sample.

Isotope fractionation: The change in isotopic composition associated with a specific biochemical reaction or diffusion process. Kinetic fractionations occur during the diffusion and fixation of CO_2, leading to biological discrimination. An example of a physical (equilibrium) fractionation occurs during the evaporation of water, when water molecules (H_2O) containing the more common light oxygen isotope (^{16}O), evaporate more readily than those with the heavy isotope (^{18}O). In a leaf, water becomes enriched in ^{18}O during evaporation.

Isotopic signal profile: Water sources vary in isotopic composition, allowing the location of source water uptake by plants to be determined provided that sufficient shift in isotopic composition occurs between surface layers and deeper groundwater.

Leaf area: The total surface area of one side of a leaf or leaves.

Leaf area density: The total area of leaves per unit volume of the canopy.

Leaf area index (drip line): Canopy leaf area per unit ground area covered to the outer edge of the canopy.

Leaf water potential (Y): A measure of the 'effective concentration' of water relative to that of pure water at the same temperature and pressure. It is used in this report to relate to the driving forces for water movement. A completely non-drought stressed leaf has a theoretical water potential of zero, as a plant becomes more drought stressed the water potential decreases, (i.e. becomes more negative). A plant always equilibrates overnight with soil water potential, and so pre-dawn measurements on leaves arte closely related to soil water potential.

^{16}O: The most abundant stable isotope of oxygen.

^{18}O: A rare heavy isotope of oxygen.

Photosynthesis: The capture of the sun's energy and the fixation of free carbon dioxide to produce simple carbohydrates that may then be used for growth.

Photosynthetic capacity (A_{max}): The maximum rate at which carbon dioxide can be fixed during photosynthesis.

Photosynthetic enzymes: Enzymes which are specifically involved in the chemical reactions of photosynthesis. The most important of these is Ribulose-bis-phosphate carboxylase (Rubisco).

Photosynthetic fixation: Uptake of CO_2 and assimilation resulting in simple carbohydrates catalysed by Rubisco.

Photosynthetic rate: The rate at which CO_2 is fixed.

Relative humidity (rh): A measure of the water content of the atmosphere (air) relative to its total capacity.

Sap flow: The rate at which sap is transported up the trunk of the tree.

Sap flux: The total amount of sap that flow through the trunk for a given period (usually 24 h).

Season shoot development: Growth characteristics of trees over the growing season (e.g. time and rate of leaf emergence, total number of leaves per shoot, rate of shoot elongation etc.)

Soil moisture deficit (SMD): Soil moisture deficit is calculated by subtracting the total profile moisture content at an individual date from the total profile moisture content at field capacity. It is equivalent to the amount of water required to return the soil moisture content to field capacity.

Specific leaf mass (SLM): A measure of the thickness of a leaf (leaf dry weight per unit leaf area).

Stomata: Pores that occur on the underside of leaves that regulate the loss of water vapour to the atmosphere and the rate of carbon dioxide uptake from the atmosphere.

Stomatal/leaf conductance: Stomatal conductance is a measure of the capacity of leaves to lose water via transpiration (g_s) calculated from the gradient in vapour pressure between leaf interior and ambient air and the rate of evaporation, which can be corrected to describe carbon dioxide uptake (g_s). Thus, higher stomatal conductance enables a greater transpiration rate to be achieved and a more rapid exchange of carbon dioxide (i.e. CO_2 for photosynthesis) across the leaf surface, with a higher C_i/C_a ratio also related to water use and carbon isotope composition for air well stirred above leaves. Leaf conductance is the same measure as stomatal conductance except that it includes the epidermis (the contribution of the epidermis is very small relative to the stomata).

Transpiration (E): A measure of the actual water loss per unit area of leaf via the evaporation of leaf water to form water vapour which may then diffuse out of the leaf, a function of leaf and canopy boundary layers and environmental conditions.

Tree water use: Integrated flux of water removed from the soil and lost from the tree via transpiration, i.e. the total amount of water that passes from the roots, through the trunk and goes to the atmosphere from the leaves.

Vapour pressure deficit: The difference between the water vapour pressure within the intercellular spaces of the leaf (assumed to be saturated at the leaf temperature) and that in the bulk atmosphere outside the leaf.

Water use efficiency (WUE): Seasonal integration of assimilation and evaporation, or the amount of water lost relative to carbon gain (growth), whether measured instantaneously via gas exchange or integrated over longer periods of growth, for which carbon isotopes provide a proxy in leaves in well stirred air.

Water sources: Water within the soil profile may have different isotopic compositions reflecting summer or winter rains, or direct evaporation (see isotopic signal profile).