Nutrient Disorders in Plantation Eucalypts

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Contents

Preface

The purpose of this manual is to illustrate the symptoms associated with essential nutrient deficiencies of those species of eucalypts which are now widely established in plantations. The manual focuses on three tropical/subtropical species *(Eucalyptus grandis, E. pellita and E. urophylla)* and one temperate species *(E. globulus)*. Other plantation species are included where illustrations were available. Techniques for identifying nutritional disorders are explained and deficiency symptoms are described in detail for twelve elements. These symptoms can be used to help determine deficiencies in nurseries or young plantations. However, symptoms are a guide to nutrient deficiencies and should be used with other diagnostic tools. For this reason, leaf analysis standards are included. Abnormal growth patterns in eucalypts may also result from nutrient toxicities and non-nutritional causes such as pathogens and environmental stresses. Some of these effects are illustrated in colour so that the reader is aware that these factors can produce symptoms that may be similar to those expressed in deficient trees.

The colour plates in this manual are of field specimens collected in People's Republic of China (P.R. China), the Philippines, Indonesia, Australia and Brazil; and of three- to six-month-old seedlings grown in glasshouses in Western Australia, predominantly at Murdoch University. The field specimens were obtained from three sources: deletion/addition fertiliser trials for single elements, fertiliser rates trials, and from the foliar analysis of healthy and unhealthy trees. In the glasshouse, nutrient deficiencies were induced in seedlings grown in virgin soils from south-western Australia and in solution culture. Four soils were used: a grey sand from Lancelin (for N, P, K, Cu, Zn), a white sand from Badgingarra (S, B), a white sand from Gnangara (Ca, Mg, Fe) and a red sandy loam from Bodallin (Mo). To obtain micronutrient deficiencies the macronutrient salts were purified to remove traces of heavy metals. Details of experimental procedures have been described by Dell and Robinson (1993).

This manual was researched with funding provided by the Australian Centre for International Agricultural Research, Project 90/44 'Increasing productivity of eucalypt plantations in China and the Philippines by inoculation with ectomycorrhizas and nutrient application' and Project 94/25 'Ectomycorrhizal fungi for eucalypt plantations in China'. We acknowledge the collaboration of the Chinese Academy of Forestry through its Research Institute of Tropical Forestry in Guangzhou, Academia Sinica through the Kunming Institute of Botany, and the Mycorrhiza Laboratory of the University of the Philippines at Los Baños. Joanne Robinson and Karen Deane provided most of the technical support in the glasshouses and analytical laboratory. Dr Mark Brundrett produced the computer graphics.

New to this edition

Information throughout the text has been updated or augmented and 107 new colour plates have been added to assist in the identification and correction of disorders in the field. Three new chapters (Chapters 1, 8 and 9) have been included, and Chapter 7 has been expanded to include new information on sampling trees for foliar analysis. Since the first edition was published in 1995, there has been a rapid expansion of eucalypt plantations in temperate Australia and elsewhere in the world. In the next decade, there will be increased plantings of eucalypts in sub-tropical parts of eastern Australia and elsewhere. Foliar analysis is now routinely being used by progressive plantation managers to monitor the nutrient status of eucalypts in plantations. Accordingly, the tables on foliar nutrient concentrations have been updated to include the most recent research findings.

The revision could not have been accomplished without the support or scientific input from: the ACIAR Fellowship Program (Daping Xu), Murdoch University, Chinese Academy of Forestry, Integrated Tree Cropping Ltd, Bukidnon Forest Incorporated, PT Indorayon, Liz Cornish, Catherine Chamberlain, Mike Calver and Amalia Sakya. Colour photographs were taken by B. Dell except where acknowledged in the captions.

We are grateful to the staff in the publishing unit of ACIAR for cheerfully guiding the manuscript to publication.

1. Introduction

Nutrient disorders have been recorded for eucalypts in nearly all the geographical regions where commercial plantations have been established (Table 1). The most often encountered disorders result from inadequate supplies of macronutrients, typically N, P or K, resulting in premature leaf drop and reduction in wood volume. As can be seen from Table 1, micronutrients, particularly B and Cu, may also limit productivity. Whilst micronutrient (B, Cu, Fe, Mn, Zn) disorders are often induced by the application of fertilisers containing only macronutrients (i.e. Ca, K, N, Mg, P, S), instances of primary B deficiency in China and Cu deficiency in Western Australia have been recently documented.

Table 1: Occurrence of nutrient deficiencies in eucalypt plantations

	B	Ca	Cu	Fe	K	Mg	Mn	N	P	S	Zn
Australia	•		•	•	•		•	•	•	•	•
Brazil	•	•			•	•		•	•	•	•
Chile	•		•		•			•	•		
China	•			•	•			•	•	•	•
India					•			•	•		
Indonesia	•		•		•	•		•	•		•
New Zealand	•			•				•	•		
Philippines	•			•	•	•		•	•		•
Portugal	•	•						•	•		
South Africa		•	•		•	•		•	•		•
Thailand	•				•			•	•		
Zambia	•										

Increasing records of micronutrient disorders in plantation eucalypts suggests that the capacity of micronutrients to limit productivity has not been adequately recognised in the past (Xu and Dell 1998). As a consequence, many operational fertiliser trials have not included micronutrients in their basal fertiliser mix. Information in this manual should help to rectify this in the future.

The productivity of many plantations in India and China (Xu et al. 2000) is less than one third of the world average for eucalypt plantations. In part, the low productivity can be attributed to soil constraints (Table 2) and inadequate fertiliser prescriptions (Xu et al. 2000). In parts of Guangdong Province, for example, the routine application of 100 kg of NPK fertiliser per hectare at planting is insufficient to maintain tree growth longer than three years. Thereafter, the canopy density declines rapidly (Plate 1), depressing cambial activity. In some regions, soil fertility has declined as a result of the removal of organic nutrient reserves and soil erosion (Plate 2). Eucalypt leaves and branches are collected for fuel (Plate 3) and remaining litter is carried by people down-slope to be burnt on arable land. The regular harvesting of foliage for distillation of oil (Plate 4) is sustainable only if nutrients harvested are replenished at the same rate.

Table 2: Some soils resulting in nutrient deficiencies in eucalypts

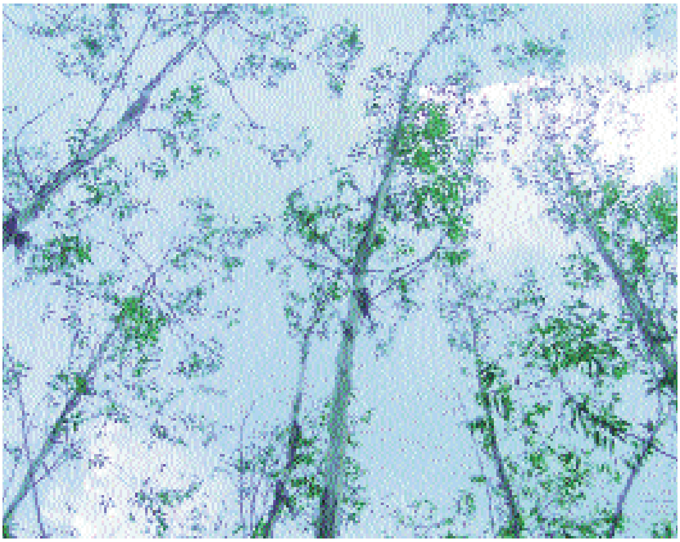
B	Sandy soils, especially those derived from granite and sandstones; quaternary deposits; peaty soils; some soils of high iron content; some serpentine soils
Ca	Acidic sandy soils
Cu	Sands; sandy gravels; calcareous soils; peaty soils; soils rich in soluble N
Fe	Sands; peaty soils; soils over limestone; alkaline soils; some serpentine soils
K	Sandy soils in high rainfall areas; soils of low cation-exchange-capacity; some volcanic soils; peaty sands; many duplex soils
Mg	Sands; calcareous soils
Mn	Sands; sandy gravels; calcareous soils
N	Sandy soils; soils low in organic matter
P	Highly weathered soils of coarse texture; high P-fixing soils (e.g. ferrosols, laterites, red and yellow earths); peaty soils; some volcanic soils
S	Sandy soils; soils low in organic matter
Zn	Acid and calcareous sands; lateritic soils; black earths; some volcanic soils

Many foresters in China, and in other countries, are still using soil analysis to identify nutrient constraints to productivity of eucalypt plantations. Unlike agricultural soil, most of the forest lands available for plantation eucalypts are quite variable. Hence data obtained from soil samples rarely represent the fertility of all sites in a plantation. Moreover, some soil data do not correlate with tree growth. For example, in Hainan, the Bray P in a sandy soil from a quaternary deposit is much higher than that in a clay loam from basalt, but tree growth on the latter soil is much better than that on the sandy soil.

Nutrient diagnosis and prognosis by chemical analysis of foliar samples will, over time, replace much of the soil analysis being undertaken at the present time. However, soil analysis will continue to be important for the monitoring of soil K in many parts of the world, and to a lesser extent for N and P. Soil analysis will also be useful in setting some sustainability indices and for identifying non-nutritional soil constraints (e.g. sodic soil, acid soil, ultramafic soil) to tree growth. For example, in the Philippines nickel toxicity was diagnosed as the causal factor of poor growth in eucalypts and yellowing in *Acacia mangium* (Dell 1997) on the basis of soil analysis. This was confirmed by foliar analysis of Ni. Conventional foliar analysis only detected low Fe concentrations caused by Ni competition in the rhizosphere. One of the major constraints of soil analysis is that data are usually restricted to the uppermost soil horizon in plantations. Whilst many feeder roots occupy this horizon, eucalypt roots explore quite deep soil profiles (Plate 5), reaching depths of nearly 50 metres in regions with a prolonged dry season (Dell et al. 1983).

Already, in China and Australia, some forest researchers have established that nutrient concentrations in young, fully expanded leaves, sampled early in a rotation, can predict wood productivity at harvest. Before the chemical analysis of foliar samples can be widely used in the nutrient management of plantation eucalypts, a standard sampling method, chemical analysis manual and interpretation guide containing data for nutrient concentration ranges (adequate, deficient and marginal) must be developed for forest managers. The first and last of these topics are explored in Chapters 7 and 8.

Many small forest farmers, especially in developing countries, do not have access to a chemical laboratory for nutrient analyses. The visible symptoms that are described in this book will enable them to identify any severe nutrient constraints in the field and to apply corrective fertilisers. In southern Australia, the colour plates can be used to identify pockets of deficiency as they occur in eucalypt plantations established on ex-pasture sites. As the soil in a forested landscape is generally variable, it is common for some trees to show deficiency symptoms well in advance of other trees. If diagnosis by visible symptoms can be made early, the information can be used to develop fertiliser management strategies to prevent nutrient constraints in the bulk of the plantation.

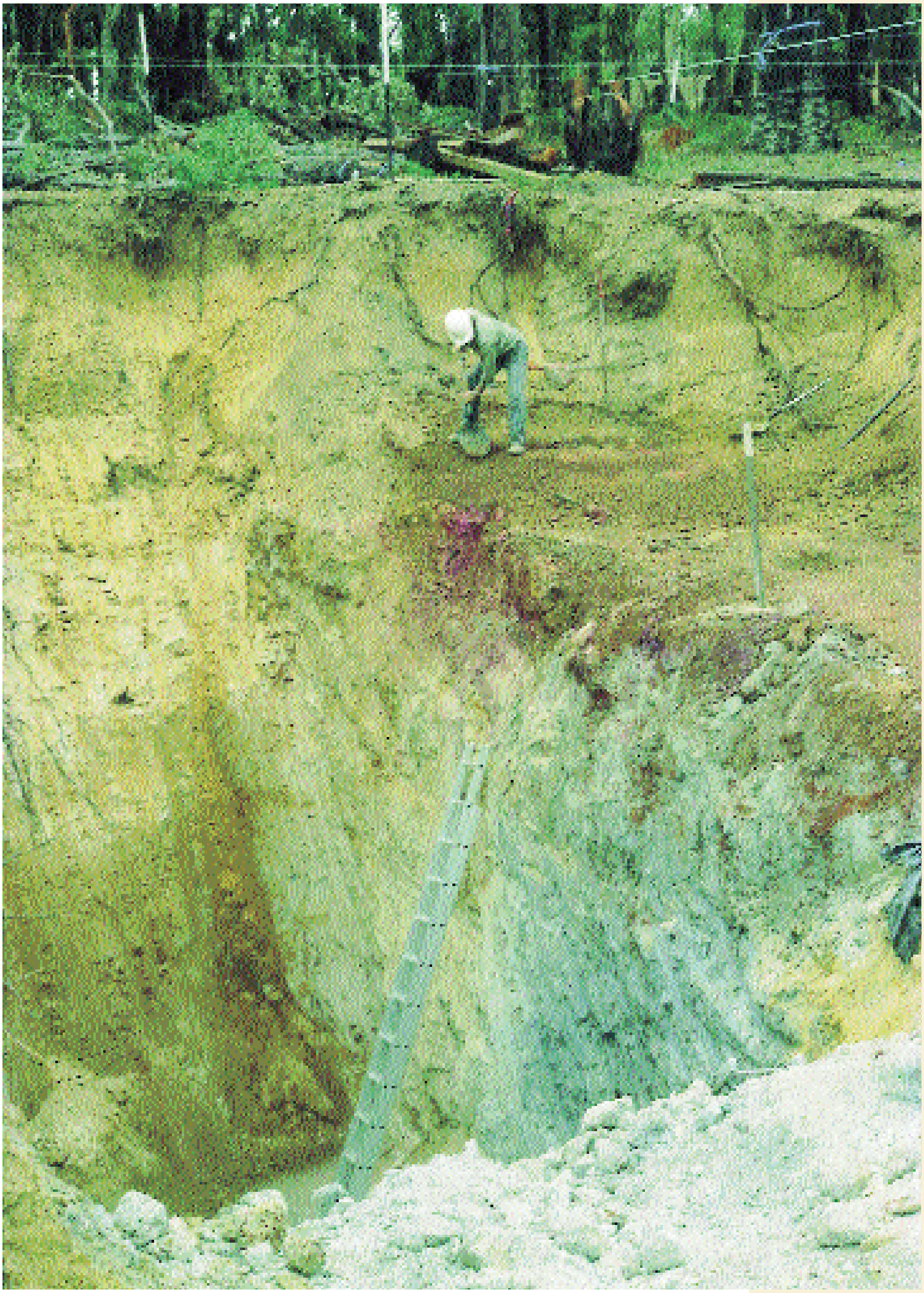


1. The sparse canopy in these eucalypts near Kaiping could have been avoided by topdressing with 200 kg N/ha and 70 kg K/ha at two years after planting. (Photo: D. Xu).

2. Loss of nutrient capital through soil erosion reduces the capacity of land to support commercial eucalypt plantations. Danzhou, Hainan. (Photo: D. Xu).

3. Where fuel is in short supply, the constant removal of sticks and leaves increases the need for the addition of fertiliser. P.R. China.

4. *E. citriodora* plantation managed for oil production. Guangdong Province, P.R. China. (Photo: N. Malajczuk)



5. Eucalypt roots can probe very deep profiles in search of water and nutrients. Here, *E. marginata* roots extend through the lateritic profile into deep clay profiles formed from the *in situ* weathering of granite (white clay) and dolerite (blue clay). Western Australia.

2. Distinguishing nutrient disorders from other causes of visual symptoms

Visual symptoms of impaired or abnormal growth in eucalypts may result from nutritional or non-nutritional causes (Fig. 1). For symptoms to be useful in the diagnosis of nutrient deficiencies, the observer must first decide whether other factors such as the presence of plant pathogens should be investigated. Fortunately, many symptoms of nutrient deficiency follow patterns of expression that can easily be distinguished from non-nutritional causes. We suggest the reader should be familiar with these patterns before attempting to diagnose deficiencies. We have included selected colour illustrations of nutrient toxicities and some non-nutritional symptoms in eucalypts to reinforce the need to fully assess all possible causes of visual symptoms .

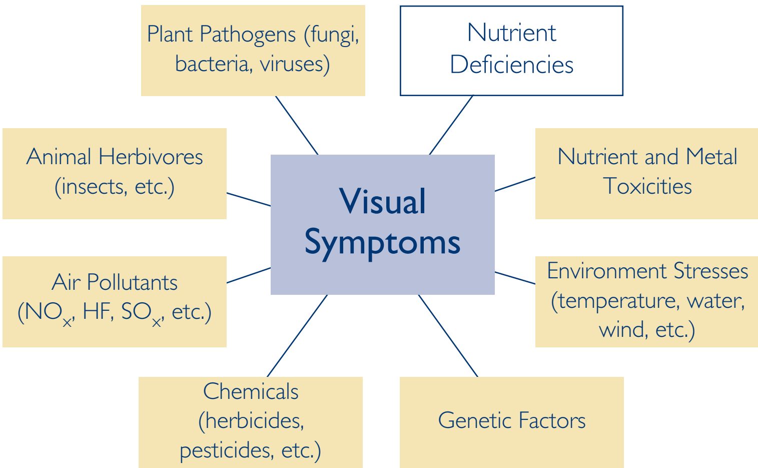


Figure 1: Many factors can cause visual symptoms in eucalypt leaves and shoots.

The main characteristics of nutrient deficiencies are:

1. Symptoms may typically take one of the following five forms: leaf chlorosis (uniform or interveinal yellowing, Plate 88, page 71), leaf necrosis (death of leaf tissue, Plate 62, page 50), leaf reddening (accumulation of anthocyanin pigments, Plate 32, page 29), leaf deformation (small leaves, irregular margins, uneven lamina, Plate 134, page 106) or death of shoot tips (Plate 140, page 110).
2. Patterns of chlorosis are related to leaf veination. Symptoms develop first in one of three areas: leaf margins (Plate 143, page 112), leaf tips (Plate 81, page 65) or interveinal areas (Plate 46, page 38).
3. Symptoms may appear initially in young expanding leaves close to the shoot tip (Plate 6, below) or in mature leaves within the canopy (Plate 7, page 9).
4. Symptoms may spread slowly from young to old leaves (Plate 77, page 63), or from old to young leaves (Plate 42, page 36).
5. Symptoms for more than one element may occur, from time to time, in the same shoot (Plate 8, 9, pages 9,10).

6. *E. globulus* with symptoms of B deficiency expressed in young leaves. Australia.

7. *E. urophylla* showing symptoms in old leaves caused by the early redistribution of K, N and Zn. Philippines.

8. *E. urophylla* showing symptoms of K deficiency (dead tissue) and B deficiency (yellowing between veins) in the same tree. P.R. China.

9. Branch of *E. urophylla* with symptoms in mature and young leaves caused by shortage of two nutrients (K and B, respectively). Philippines.

3. Nutrient use and movement in the tree

An understanding of the functional requirement of essential nutrients by plants and how nutrients are redistributed between organs according to demand are prerequisites to diagnosing nutrient deficiencies by visual symptoms. For example, because Fe is required during the synthesis of chlorophyll, Fe deficiency typically causes chlorosis (yellowing) of leaves (Plate 89). Deficiencies of nutrients that are essential for cell elongation and cell wall formation, such as Zn, B and Ca, are manifested in meristematic regions and typically result in malformed organs (Plate 144). Further information on nutrient function can be found together with colour plates of deficiency symptoms in Chapter 5.

In addition, the position in the shoot where symptoms of deficiency first appear largely depends on whether the element is redistributed from old to young leaves, and from other parts of the tree as they age. This redistribution is dependent on translocation of the element in the phloem. The mobility of nutrients within the phloem varies considerably between elements and sometimes may be species dependent.

It is conventional to group the essential plant elements into three groups, viz. elements that are exported from leaves under nutrient deficiency or as leaves senesce (phloem-mobile nutrients), nutrients that are retained in leaves (phloem-immobile nutrients) and nutrients that may be exported from leaves but only under particular conditions (variably phloem-mobile nutrients). The phloem-immobile nutrients either do not move out of leaves or, under conditions of inadequate supply of nutrients in soil, move too slowly to prevent the onset of deficiency symptoms in organs where cell division and cell expansion are occurring. Strong gradients in phloem-immobile nutrients can occur within a young tree as the external supply diminishes with tree growth (Fig. 2).

The nitrogen supply to the tree is the most important factor influencing the movement of the variably phloem-mobile nutrients. Observations on symptom development and nutrient contents in leaves of different ages suggests the mobilities proposed in Table 3. This nutrient behaviour is based on plants where the nutrient being evaluated is supplied to the plant through the root system. The fate of foliar supplied nutrients may possibly differ from these findings. The phloem mobilities suggested for essential elements in eucalypts are similar to those categorised previously for most field crops (e.g. Marschner 1995, Smith and Loneragan 1997).

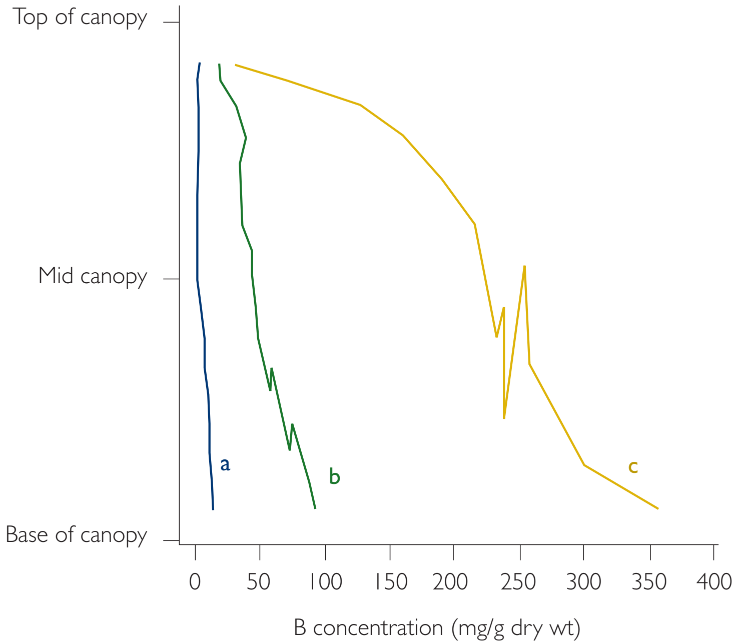


Figure 2: Gradients in foliar B concentrations up the trunk of *E. globulus*. At planting, trees were given fertiliser containing nil (a), low (b) or high (c) amounts of B. Yunnan Province, P.R. China.

Table 3: Mobility of nutrients in the phloem in eucalypts.

Highly mobile	Variably mobile	Immobile
Nitrogen	Copper	Boron
Phosphorus	Magnesium	Calcium
Potassium	Molybdenum	Iron
	Sulfur	Manganese
	Zinc	

4. Guide to nutrient deficiency symptoms

Correct diagnosis of a nutrient deficiency depends on thorough documentation of symptoms. Information on the pattern of symptom development and location of symptoms on the tree are especially important. As indicated earlier, the function and redistribution of nutrients via the phloem strongly influence symptom expression. For example, symptoms of N, P and K (all mobile nutrients) appear first in old leaves and gradually extend towards the shoot apex. As the following plates show, symptoms which are expressed in mature leaves are distinctive for these three elements because of the different roles that they play in the maintenance of cellular function. In contrast, symptoms of Ca, S, B, Fe, Cu and Zn are first expressed in young leaves and spread to mature leaves. Magnesium deficiency may appear simultaneously in expanding and fully expanded leaves. Manganese deficiency appears first on expanding leaves and extends both to younger and older leaves. The following questions will assist in directing the reader to the appropriate set of colour plates.

Questions relating to symptoms in leaves

1. Do symptoms appear first in expanding, recently mature or old leaves? (See Fig. 3 inside back cover).
2. What do the symptoms look like? Note the colour (eg. yellow, purple), presence of dead (necrotic) tissue (usually brown, grey or white), and the distribution of abnormal tissue (eg. along veins, in interveinal regions, at the leaf apex, at the leaf base, along leaf margins) (See Fig. 3).
3. Are the leaves misshapen? See B, Ca and Cu (Fig. 4, page 14).
4. Are the leaves exceptionally small? See Zn (Fig. 3).

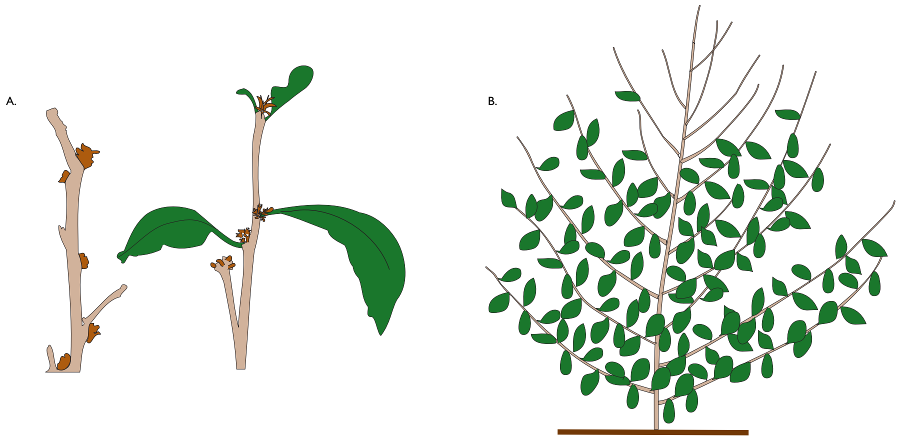


Figure 4: Distortion of apical buds and leaves (A) and loss of apical leaves (B) due to boron or copper deficiency.

Questions relating to symptoms in canopies

1. Does the canopy apex consist mostly of naked green branches? (See Plate 10, page 15).
2. Does the tree have a mop-like young canopy or a gap in the canopy exposing the trunk? (See Plate 11, page 15).
3. Has the canopy rapidly shed most of its leaves? (See non-nutritional factors, Chapter 6, and Plate 200, page 151).
4. Are the dominant branches dying back from their tips? See B and Cu (Fig. 4, see above).
5. Are there multiple leaders? See B, Cu, and Ca (Plates 12 and 13, pages 16-17). Also check for parrot damage, stem cankers and frost damage.

Questions relating to symptoms in stems

1. Is the trunk sinuous (speed-wobble is present)? See Cu (Plate 14, page 18).
2. Are the branches pendulous? See B (Plate 15, page 18) and Cu (Plate 107, page 85).
3. Are the nodes expanded with multiple shoots? See B and Cu (Fig. 4).
4. Are the stems split and bleeding? See B and Cu.
5. Is the wood poorly lignified? See B, Cu and Mn. Evaluation of lignin-like substances in cell walls can be simply carried out by applying a drop of phloroglucinol in 70% ethanol and a drop of concentrated HCl to a freshly cut branch in the field. Lignified wood stains a deep cherry red.

10. Cu-deficient *E. globulus* tree in the centre has naked branches. Australia.

11. Mn-deficient *E. globulus* canopies with bare sub-terminal branches. Australia.

12. Loss of apical dominance in *E. urophylla* due to B deficiency causing death of the leader followed by growth of lateral branches. P.R. China.

13. Wind-throw of branches in a *E. globulus* tree where the leader has been damaged. Many factors can cause this, including nutrient deficiencies (Ca, B, Cu, Mn), pests (parrots), and frost. Australia.

14. Speed-wobble in the main stem of Cu-deficient *E. globulus*. Australia.

15. Reduced lignification of the wood in B-deficient *E. globulus* causes the branches to weep. P.R. China.

5. Atlas of deficiencies

Nitrogen

Function

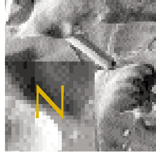
Nitrogen is a constituent of amino acids, amides, amines, N bases, nucleic acids, alkaloids, chlorophyll and many co-enzymes. Nitrogen deficiency leads to leaf chlorosis due to reduced chlorophyll formation. Since most amino acids are also precursors of the polypeptide chains of proteins, N influences many enzyme reactions. Nitrogen is also a structural component of cell walls. As old leaves senesce, protein is degraded and soluble forms of N are retranslocated in the phloem to growing parts of the plant. Hence, in N deficiency, yellowing occurs first on mature leaves. However, symptoms spread rapidly to young leaves in eucalypt seedlings. Nitrogen is absorbed by roots in both the nitrate and ammonium forms. *E. globulus* seedlings utilise nitrate less effectively than ammonium and nitrate toxicity has been observed (Shedley et al. 1995)

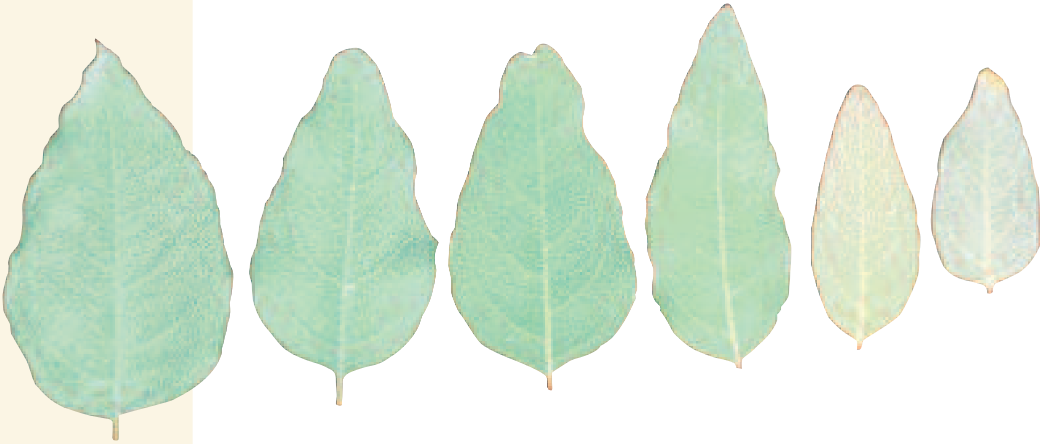
Symptoms

Initially the interveinal areas of mature leaves turn pale green leaving the major veins broadly and irregularly flanked with green (*E. grandis, E. pellita, E. urophylla*). In *E. globulus* the surface waxes mask the early stages of chlorosis. With time, yellowing spreads to include expanding leaves and all leaves become uniformly pale yellow. In *E. pellita* seedlings, the oldest leaves may develop small necrotic spots with bleached centres and purple margins. In seedlings, rapid onset of early deficiency symptoms may also appear first in recently expanded leaves. When plants are uniformly yellow the symptoms of N deficiency can easily be mistaken for those of S deficiency. However, since S deficiency symptoms spread from young to old leaves the two nutrient disorders can be distinguished in mildly affected trees.

16. *E. globulus* ssp. *globulus*: stunted, pale green N-deficient trees (foreground) and N-adequate trees (background). Ex-bush site, Western Australia.

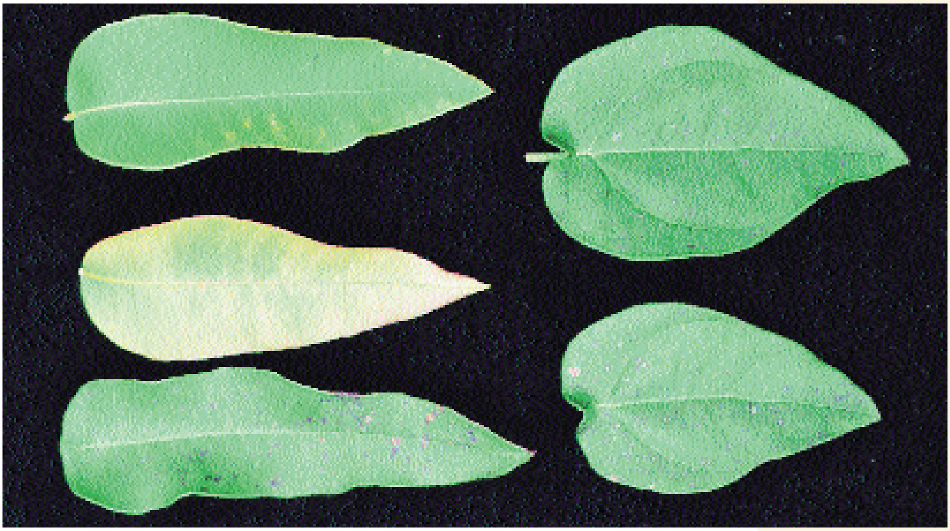
17. *E. globulus* ssp. *globulus*: uniformly pale green leaf (right) from N-deficient seedling; normal leaf (left). Western Australia.





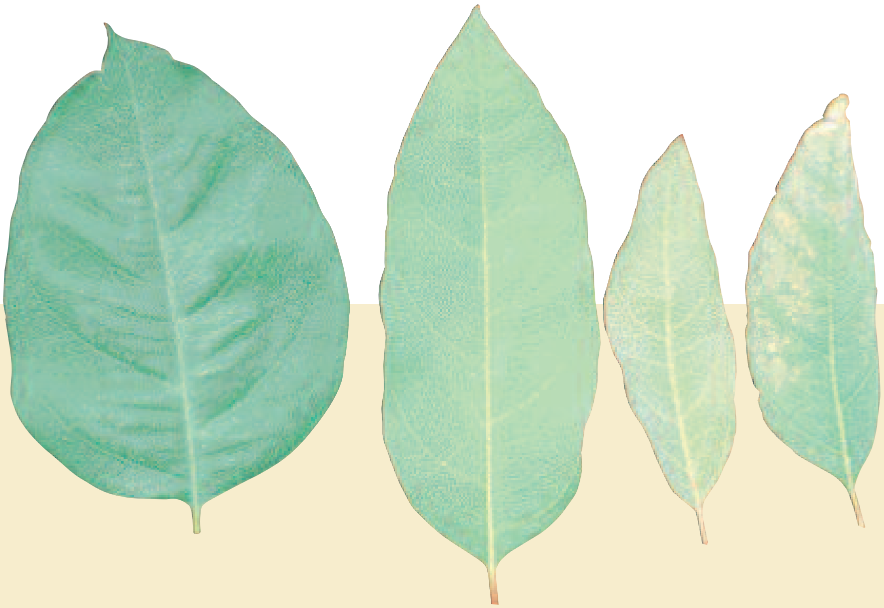
18. *E. grandis*: fully expanded leaves showing mild (yellowing in interveinal areas) to severe (uniform yellow leaves) N deficiency (right); normal leaf (left). Glasshouse.

19. *E. grandis* x *E. urophylla*: mid-canopy shoots from N-deficient (left) and N-adequate (right) one-year-old trees. Guangdong Province, P.R. China.



20. *E. maculata*: yellow leaves from N-deficient seedling (top and centre left); leaf with spots (lower left) from P-deficient seedling; normal leaves (right). Glasshouse.

21. *E. pellita*: N-deficient seedlings. Guangdong Province, P.R. China.



22. *E. pellita*: chlorosis and necrosis in mature leaves (right) from N-deficient seedling; normal leaf (left). Glasshouse

23. *E. pellita*: recently expanded N-deficient leaves on a two-year-old tree. Guangdong Province, P.R. China.

24. *E. urophylla*: uniformly pale green fully expanded leaves (right) of N-deficient seedlings; normal leaf (left). Glasshouse.

25. *E. urophylla*: branch of N-deficient two-year-old tree. Philippines.

Phosphorus

Function

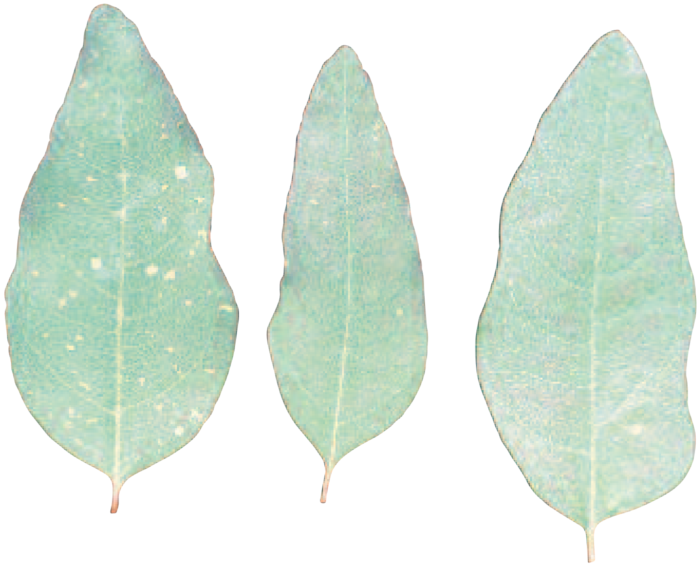
Phosphorus is essential for plant growth as it is involved in most metabolic processes. Phosphorus is a constituent of nucleic acids, phospholipids, phosphoproteins, phosphate esters, dinucleotides and adenosine triphosphate. Hence, P is required for the storage and transfer of energy, photosynthesis, electron transport processes, the regulation of some enzymes (for example, in the synthesis of sugars and starch), and the transport of carbohydrates. Phosphorus is phloem-mobile and the purple symptoms of P deficiency appear first in old leaves as P is redirected to young leaves.

Symptoms

The first sign of P deficiency is the appearance of small purple interveinal blotches on mature leaves. The centre of each blotch then becomes necrotic, typically turning brown or white. Symptoms spread from old to young leaves. Severely deficient trees are stunted and all the foliage turns purple (*E. grandis, E. pellita, E. urophylla*). In the field, severely P-deficient *E. pellita* and *E. urophylla* trees can have purple foliage without necrotic lesions. In *E. globulus* the necrotic spots coalesce into large areas of dead tissue with irregularly shaped margins.

26. *E. globulus* ssp. *globulus*: necrosis and purple areas on old leaves of a P-deficient seedling. Glasshouse.

27. *E. globulus* ssp. *globulus*: necrotic patches on mature leaves of P-deficient six-month-old tree. Yunnan Province, P.R. China.



28. *E. grandis*: necrotic patches on mature leaves of P-deficient seedlings. Glasshouse.

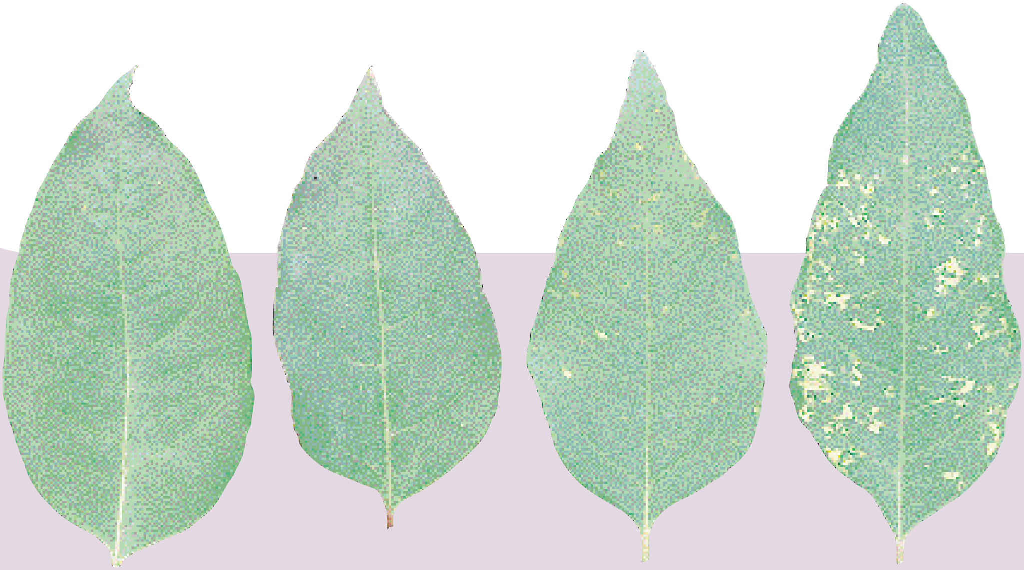
29. *E. grandis* x *E. urophylla*: six-month-old trees showing severe P deficiency (foreground). Guangdong Province, P.R. China

30. *E. grandis* x *E. urophylla*: P-deficient six-month-old tree with purple leaves and stunted main stem. Guangdong Province, P.R. China.

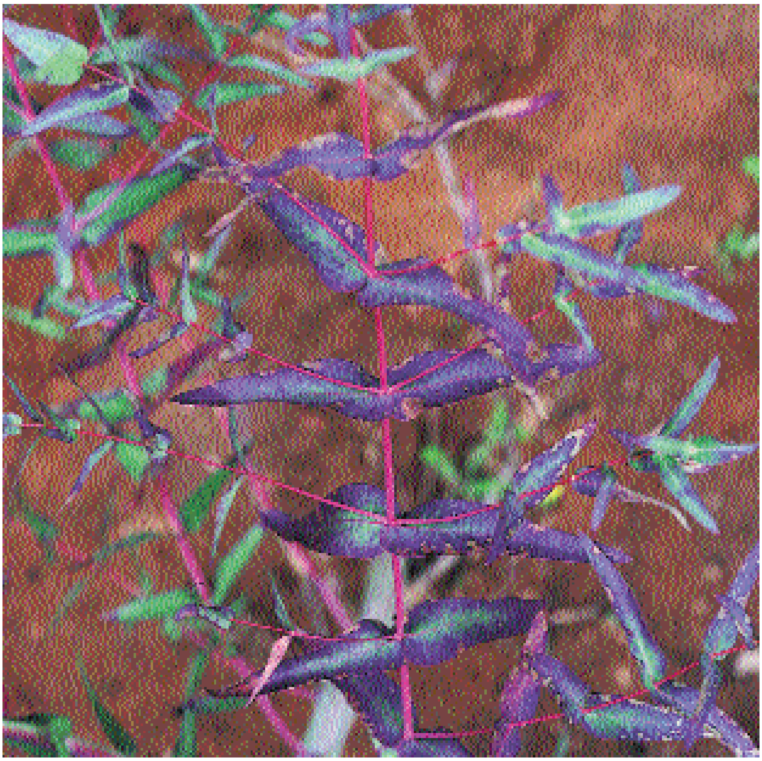
31. *E. grandis* x *E. urophylla*: necrotic spots on expanding and fully expanded leaves of P-deficient tree. Guangdong Province, P.R. China

32. *E. pellita*: severely P-deficient six-month-old trees with purple foliage. Guangdong Province, P.R. China.

33. *E. pellita*: young leaves with purple pigmentation from a severely P-deficient tree. Guangdong Province, P.R. China.

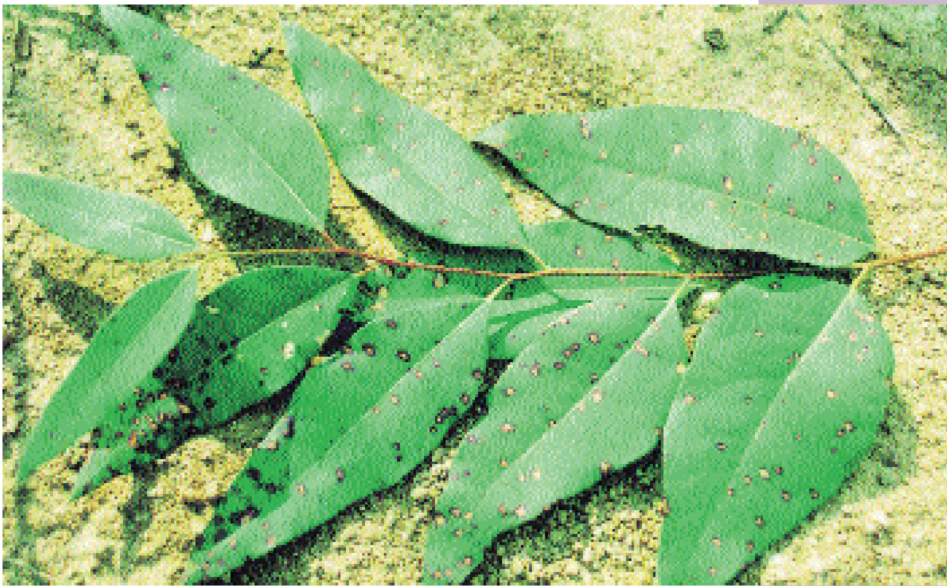


34. *E. pellita*: necrotic areas on mature leaves of P-deficient seedlings. Glasshouse.

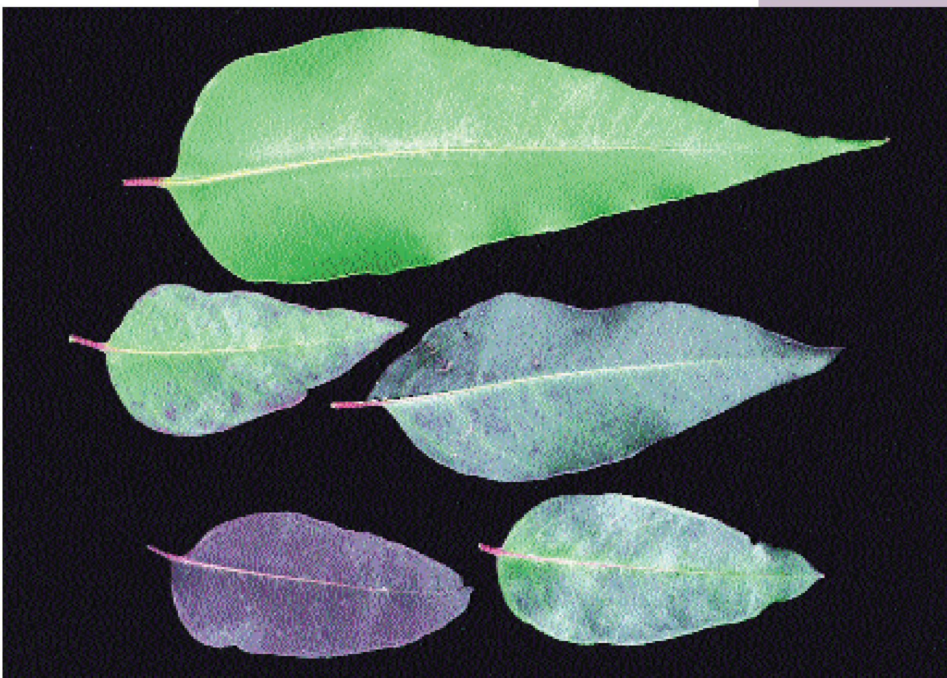


35. *E. smithii*: P-deficient six-month-old tree with purple leaves. Yunnan Province, P.R. China.

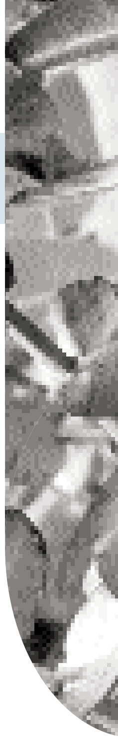
36. *E. urophylla*: part of a P trial showing treatment without P (foreground) and with adequate P (rear). Guangdong Province, P.R. China. (Photo: M. Brundrett).



37. *E. urophylla*: branch of a P-deficient tree with necrotic spots outlined in purple. Guangdong Province, P.R. China. (Photo: D. Xu).



38. *E. urophylla*: mature leaves of P-deficient six-month-old trees showing reduced leaf size and purple colour (lower); normal leaf (upper). Bukidnon Province, Philippines.



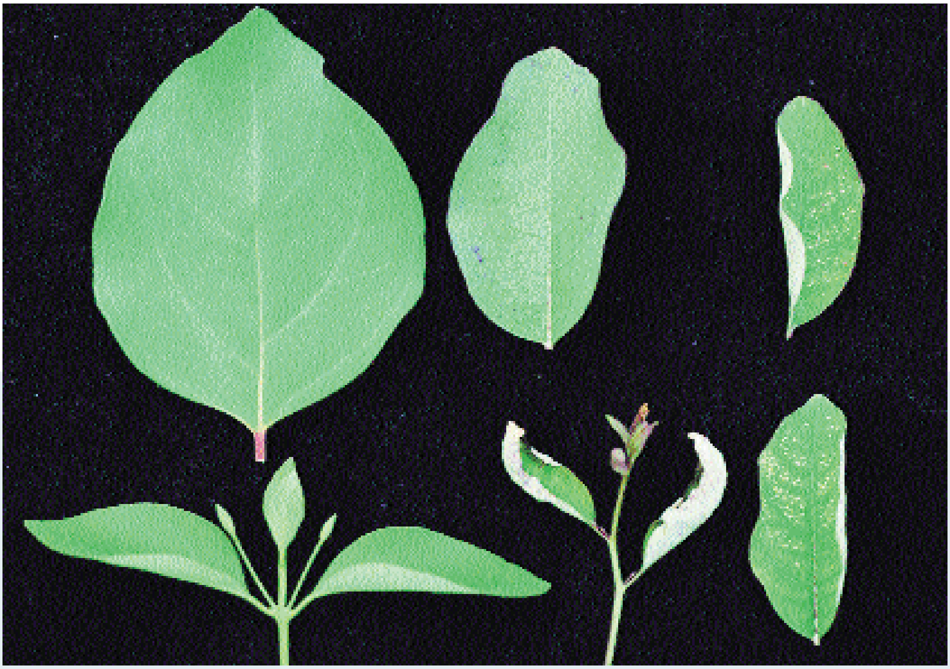
Potassium

Function

Potassium is necessary for maintaining turgor, stomatal control, pH stabilisation and osmoregulation of cells. It is required for the synthesis of proteins, for the metabolism of carbohydrates and lipids, and is an activator of numerous enzymes. In deficient plants, protein synthesis, photosynthesis and cell extension are impaired, and localised cell death occurs. Potassium moves freely in the phloem, so it is readily exported from old leaves where symptoms of deficiency first appear.

Symptoms

Potassium deficiency in eucalypts is characterised by necrosis or scorching of old leaves. Symptoms spread from old to young leaves and, in severely deficient seedlings, the shoot apex is affected resulting in dwarfing. Scorching causes severely affected leaves to cup upward or the tip to curl. In the field, symptoms have been observed up to the shoot tip but leaves were normal in size. Large differences in the appearance of symptoms occur between species. In *E. grandis* patches of interveinal tissue, approximately mid-way between the midrib and leaf margin, turn pale green then white and dry. The surrounding blade often becomes purple. Although necrotic areas form between the veins, the margins and leaf tips become scorched in juvenile foliage of *E. urophylla*. Juvenile foliage of *E. grandis* × *E. urophylla* develops interveinal chlorosis then marginal scorching. In mature adult leaves of this cross, scorching extends interveinally from the margins. In *E. pellita* K deficiency causes interveinal yellowing and the midrib and main lateral veins are flanked dark green. Gradually the chlorotic tissue dies and becomes bleached. In *E. globulus* seedlings the first sign of K deficiency is interveinal and marginal scorching. Although marginal scorching has been observed in juvenile foliage in the field, the most common symptom is the yellowing of interveinal regions of fully expanded leaves. Removal of the surface wax reveals broad green veins separated by strips of yellow tissue. In mature adult leaves, marginal chlorosis and necrosis develops but not to the same extent as in juvenile foliage.



39. *E. diversicolor*: chlorosis and necrosis in leaves of K-deficient seedlings. Normal leaves at left. Glasshouse.

40. *E. globulus* ssp. *globulus*: marginal necrosis of fully expanded juvenile leaves in a K-deficient seedling. Glasshouse.

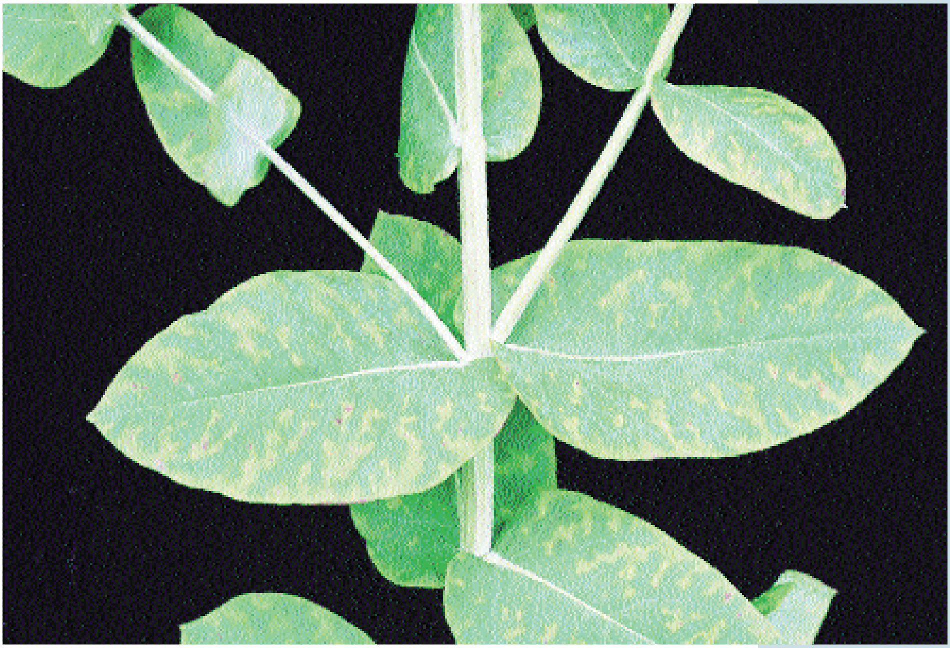
41. *E. globulus* ssp. *globulus*: leaf burn in juvenile foliage of a K-deficient tree. Western Australia.

42. *E. globulus* ssp. *globulus*: basal leaf shedding caused by K deficiency. Yunnan Province, P.R. China.

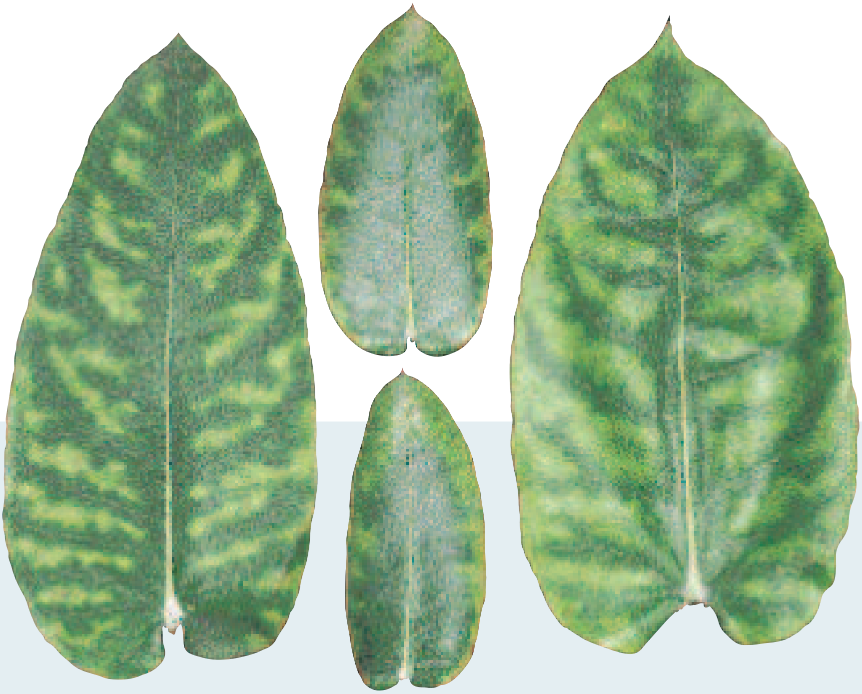
43. *E. globulus* ssp. *globulus*: sometimes K-deficient mature leaves develop reddish autumn tones as well as necrosis. Western Australia.

44. *E. globulus* ssp. *globulus*: stunted growth of a severely K-deficient one-year-old tree. Western Australia.

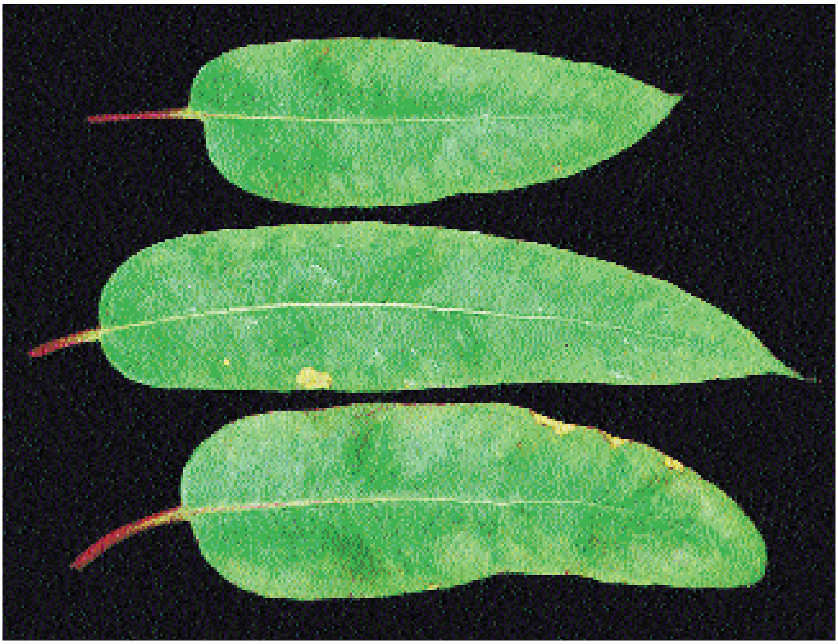
45. *E. globulus* ssp. *globulus*: one-year-old tree with symptoms of K deficiency in mature leaves. Yunnan Province, P.R. China.



46. *E. globulus* ssp. *globulus:* marginal necrosis in recent fully expanded leaves of a K-deficient nine-month-old tree. Yunnan Province, P.R. China.

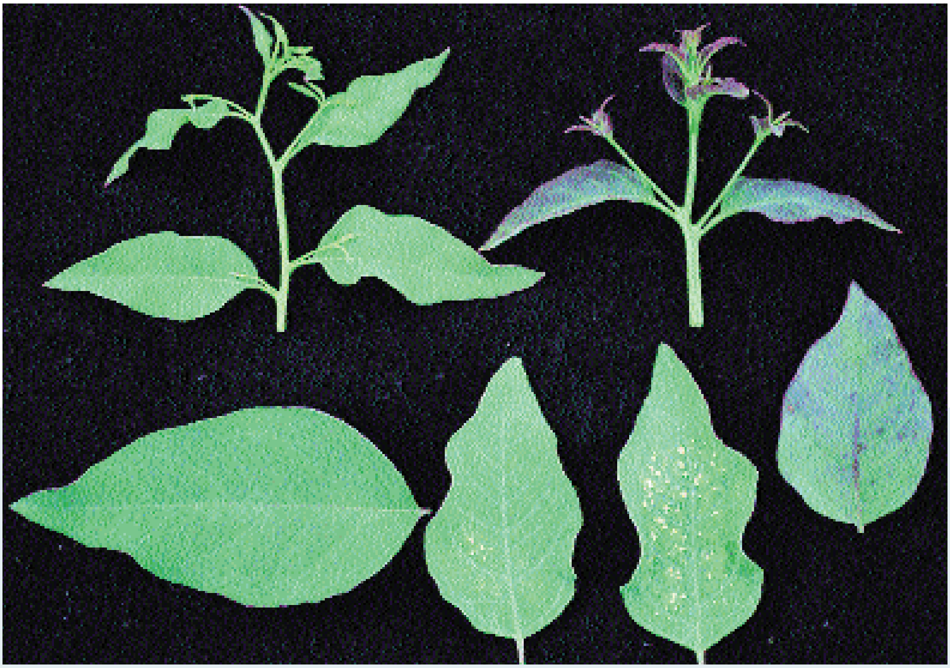


47. *E. globulus* ssp. *maidenii*: mature juvenile leaves with wax removed showing interveinal chlorosis due to K deficiency. Yunnan Province, P.R. China.

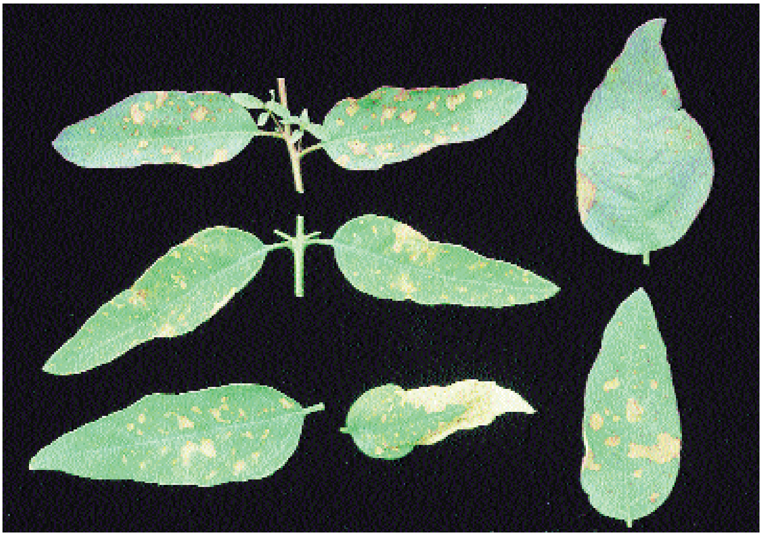


48. *E. globulus* ssp. *maidenii*: mature adult leaves showing marginal chlorosis and necrosis due to K deficiency. Yunnan Province, P.R. China.

49. *E. grandis*: K-deficient seedling with necrotic patches on fully expanded leaves. Glasshouse.

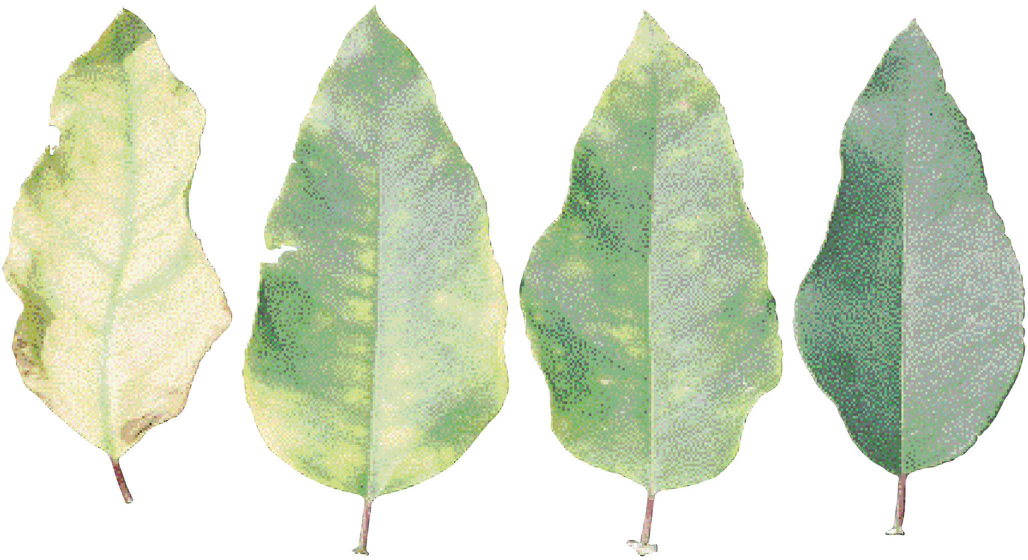


50. *E. grandis*: speckled development of interveinal chlorosis and necrosis in recently expanded leaves of K-deficient seedling (right); normal leaves (left). Glasshouse.

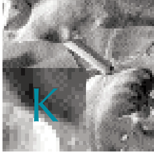


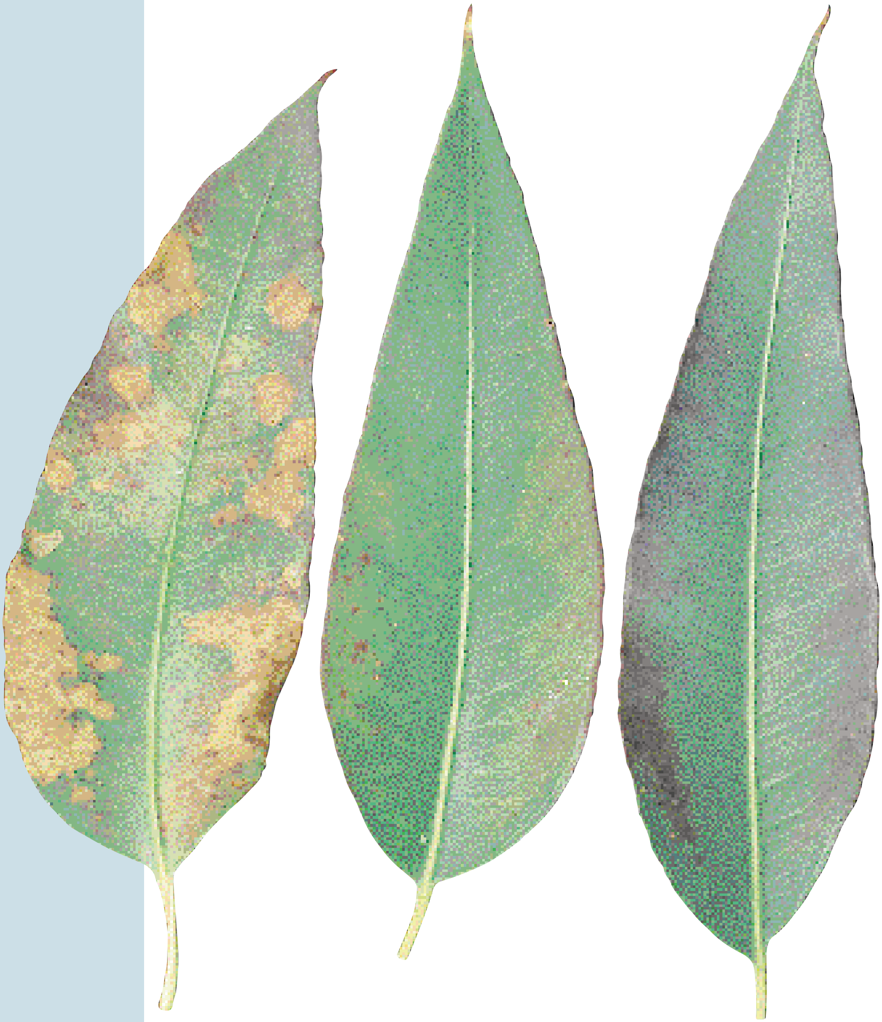
51. *E. grandis*: interveinal, marginal and tip necrosis in fully expanded leaves of K-deficient seedlings. Glasshouse.

52. *E. grandis* x *E. urophylla*: mild (left) to severe (right) symptoms of K deficiency. Note the purple pigmentation followed by apical and interveinal necrosis. Guangdong Province, P.R. China.



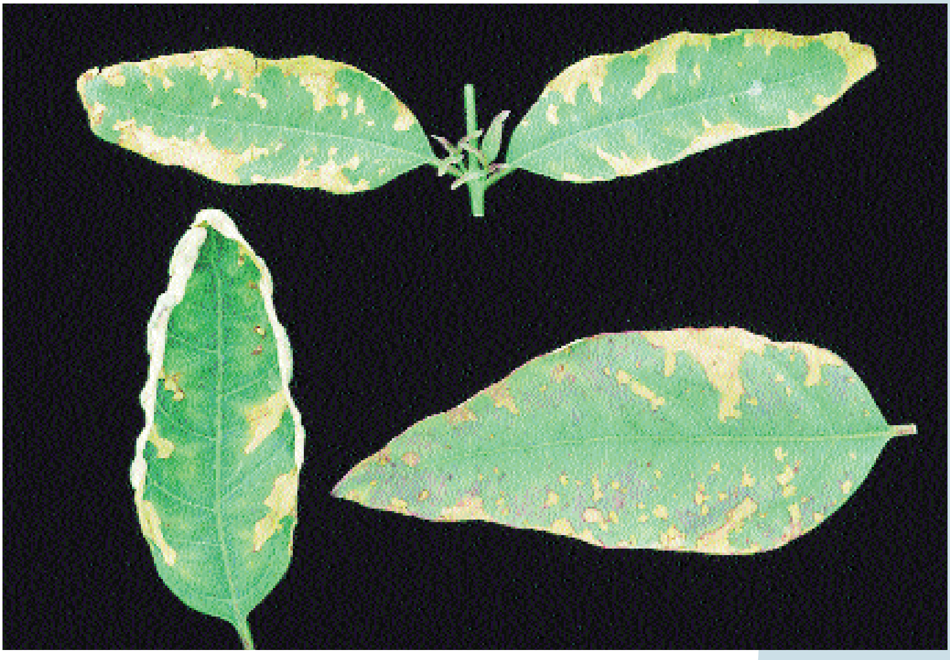
53. *E. grandis* x *E. urophylla*: interveinal chlorosis and marginal necrosis of mature leaves of a nine-month-old tree with mild K deficiency. Guangdong Province, P.R. China.



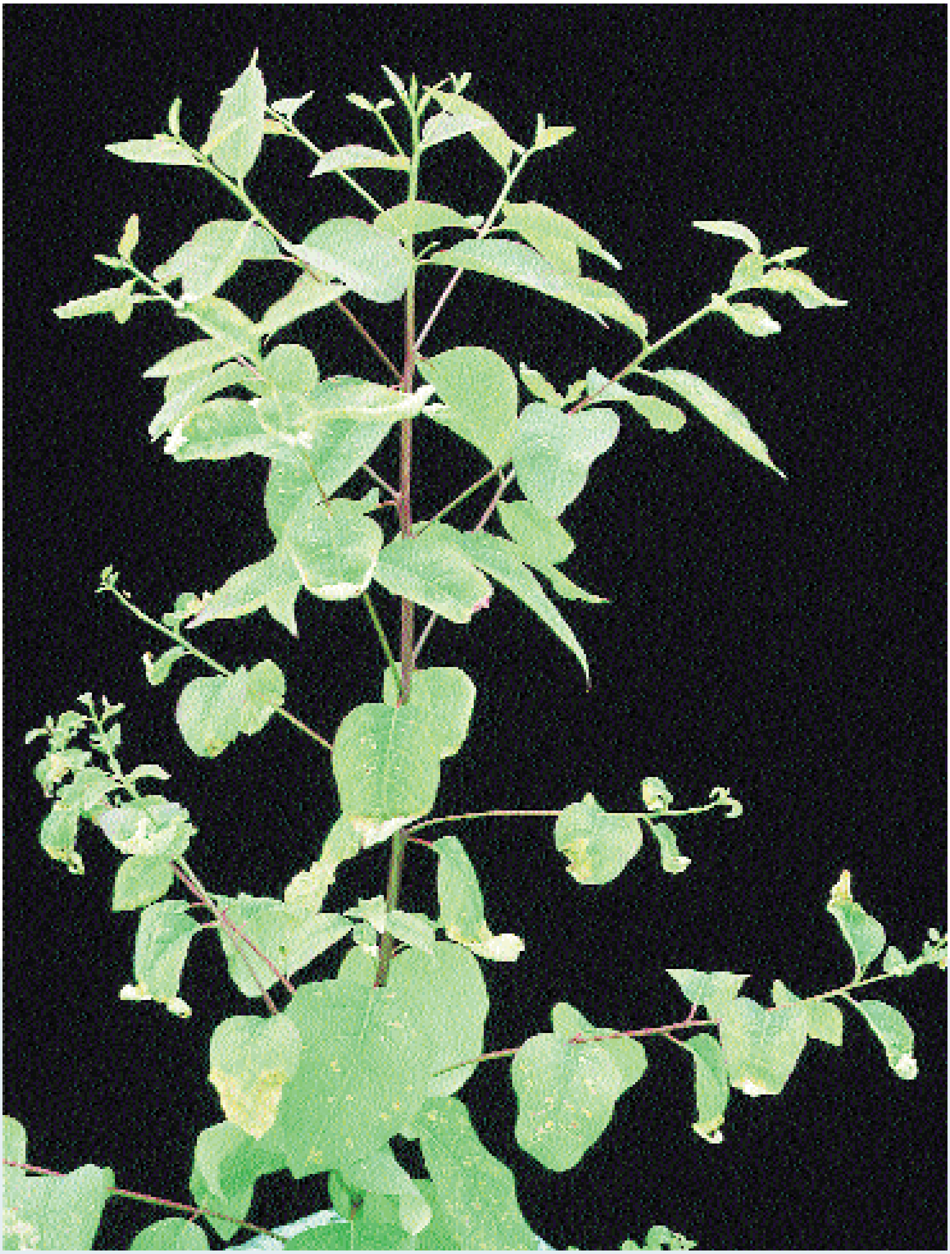


54. *E. grandis* x *E. urophylla*: necrosis of mature adult leaves from a two-year-old tree. Guangdong Province, P.R. China

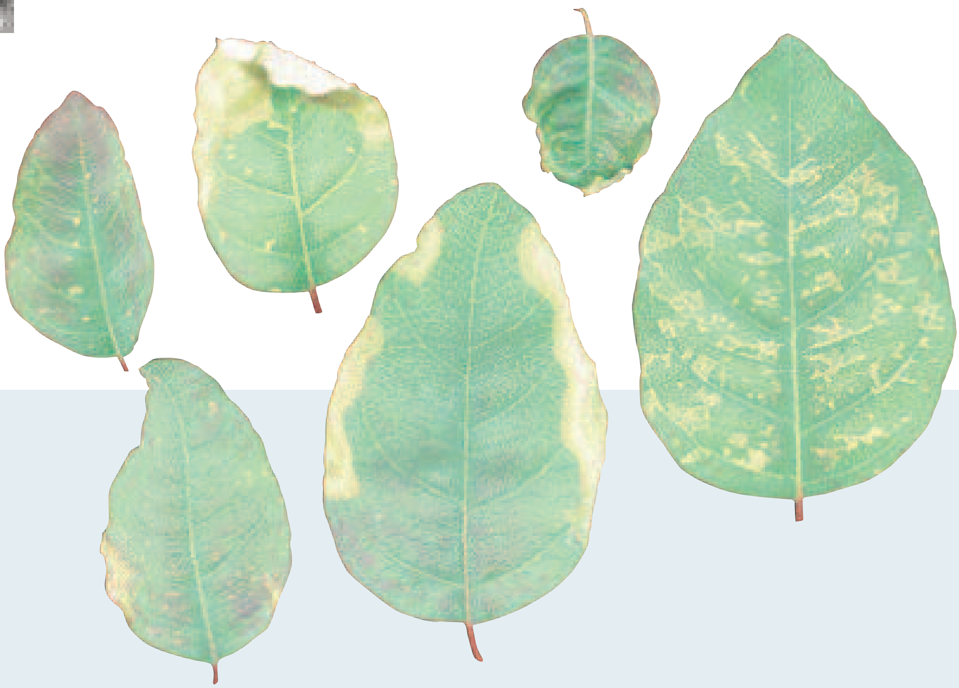
55. *E. pellita*: K-deficient seedling with severe marginal and interveinal necrosis of mature leaves. Glasshouse.



56. *E. pellita*: interveinal and marginal chlorosis and white necrosis of leaves from a K-deficient seedling. Glasshouse.



57. *E. urophylla*: K-deficient seedling with necrotic patches on expanding and fully expanded leaves. Glasshouse.

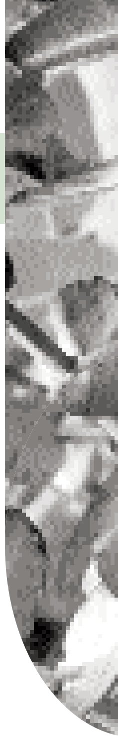


58. *E. urophylla*: speckling and marginal necrosis of fully expanded leaves of a K-deficient seedling. Glasshouse.

59. *E. urophylla*: one-year-old K-deficient tree with necrotic areas on leaves in the centre of the canopy. Guangdong Province, P.R. China.

60. *E. urophylla*: severely K-deficient tree. Leaves become dry and are then shed. Guangdong Province, P.R. China. (Photo: D. Xu).

Calcium

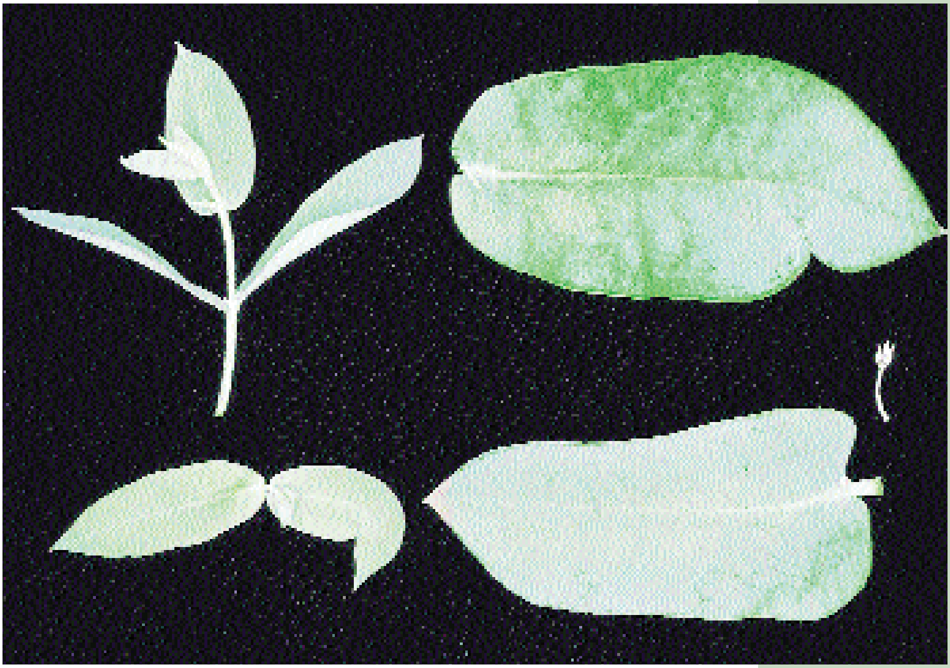


Function

Most of the Ca in the plant occurs in the cell wall where it is associated with pectin in the middle lamella. In the vacuole, Ca may be present as calcium oxalate. Calcium is also required for membrane integrity and function. Because Ca is essential for cell division and growth, the growing tips of roots and shoots are particularly vulnerable to Ca deficiency. The Ca which is delivered in the transpiration stream to old leaves can not be exported to young Ca-deficient tissues since Ca is not transportable in the phloem. Symptoms of Ca deficiency, typically cell death, appear first in developing organs such as buds and elongating leaves.

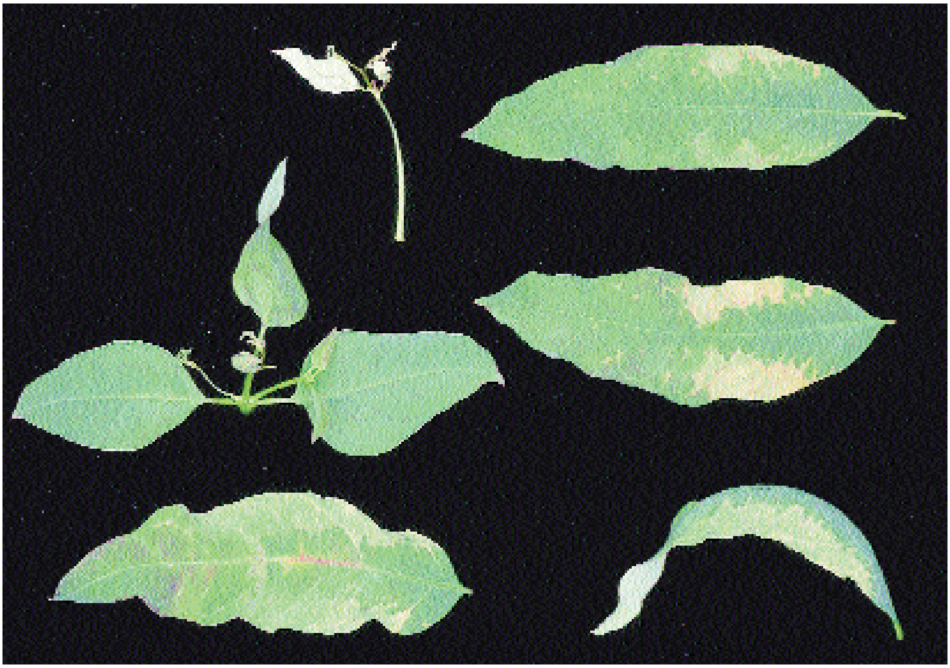
Symptoms

Deficiency symptoms are expressed at the shoot tip and in expanding leaves. Impaired expansion at the margins of young leaves causes them to buckle or become sickle shaped (*E. globulus*). Growing points die and fall off. Expanding leaves develop tip and marginal burns, and the leaves appear distorted with rolled margins or reverse cupping (*E. grandis, E. urophylla*). New leaves which appear from axillary buds also die and are shed. A form of shoot die-back has been associated with Ca deficiency.



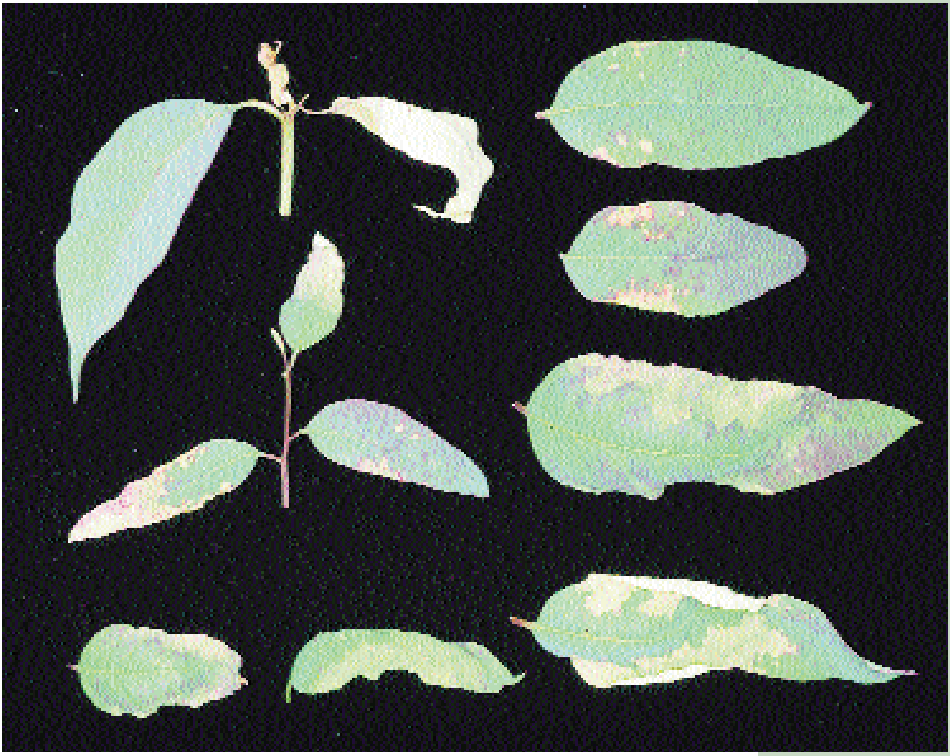
61. *E. globulus* ssp. *globulus*: early symptoms of Ca deficiency: death of axillary buds, tip burn and leaf malformation. Glasshouse.

62. *E. grandis*: necrosis of young leaves of Ca-deficient seedlings. Glasshouse.



63. *E. grandis*: severe Ca deficiency symptoms: death of growing points, leaf necrosis, reverse cupping of leaves. Glasshouse.

64. *E. maculata*: leaf distortion due to Ca deficiency. Glasshouse.



65. *E. urophylla*: tip death, necrosis and cupping of leaves of Ca-deficient seedlings. Glasshouse.

Magnesium

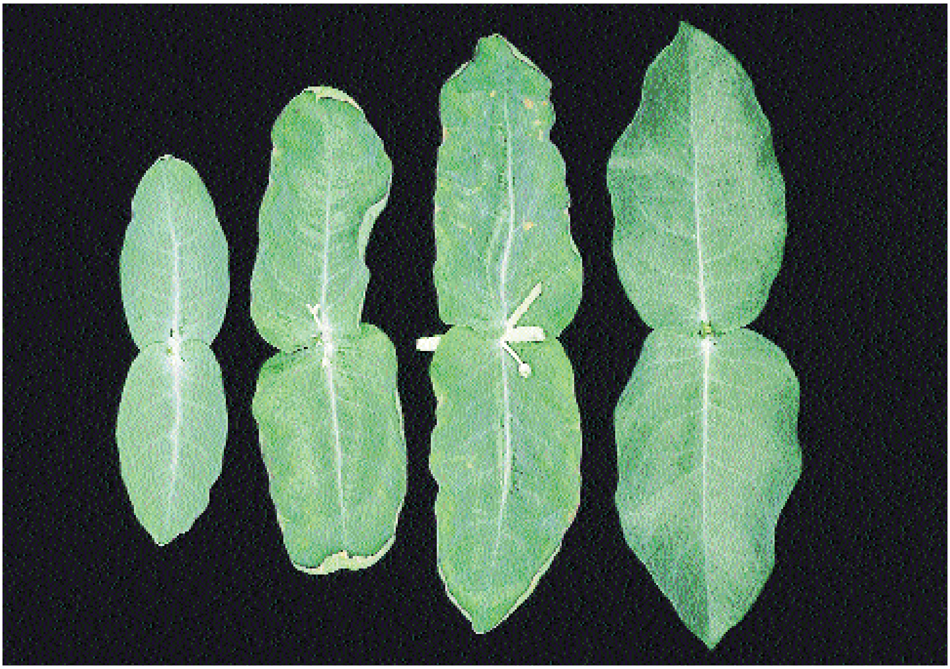
Function

A major function of Mg is as a co-ordinated metal in chlorophyll. Magnesium is also required for protein synthesis, the activation of many enzymes, and the regulation of cellular pH and cation-anion balance. Magnesium is exported in the phloem from old leaves. In woody plants, yellow deficiency symptoms commonly appear in mature leaves.

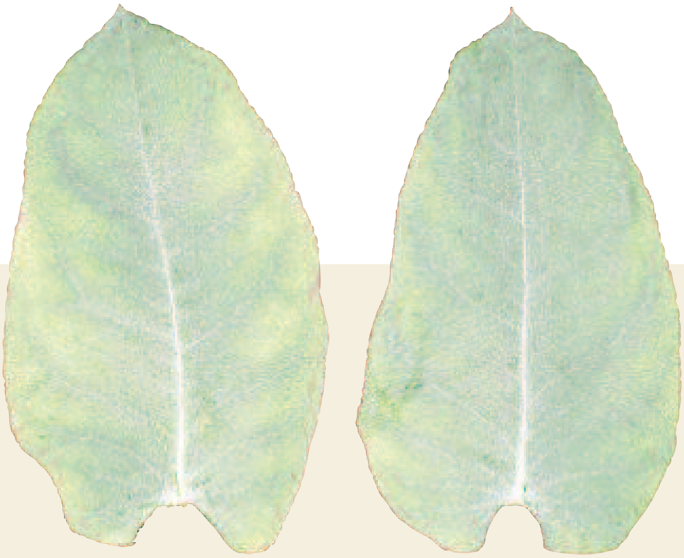
Symptoms

Magnesium deficiency symptoms are first expressed in fully expanded leaves and, when deficiency is severe, they extend into the young foliage. Green tongues of tissue along the major veins are typical of Mg deficiency in eucalypts (see *E. maculata*). In *E. globulus* the leaves become beaked and yellowing extends from the margins between the broad green veins. The chlorosis is masked by the surface wax deposits. Beaking also occurs in *E. grandis*. In juvenile foliage of *E. grandis* and *E. urophylla*, interveinal and marginal areas of leaves become purple. Small brown necrotic spots may then develop. Adult leaves of Mg-deficient *E. grandis* × *E. urophylla* trees have wide green midribs longitudinally flanked by yellow tissue. The first sign of Mg deficiency in *E. pellita* is a change in the colour of the interveinal tissue which appears ‘wet’.

66. *E. globulus* ssp. *globulus*: Mg-deficient seedling with beaked leaves.

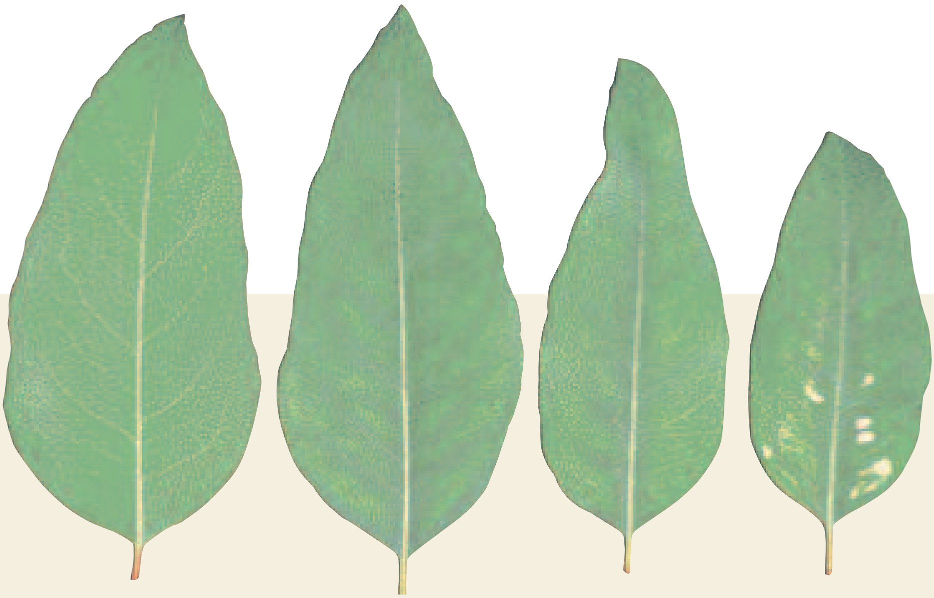


67. *E. globulus* ssp. *globulus*: expanding (left) and expanded (centre) leaves with symptoms of Mg deficiency (wax removed): marginal chlorosis followed by necrosis. Glasshouse.



68. *E. globulus* ssp. *globulus*: marginal and interveinal chlorosis in old leaves of a mildly Mg-deficient seedling. Glasshouse.

69. *E. grandis*: mature leaves from a Mg-deficient seedling showing purpling and beaking. Glasshouse.

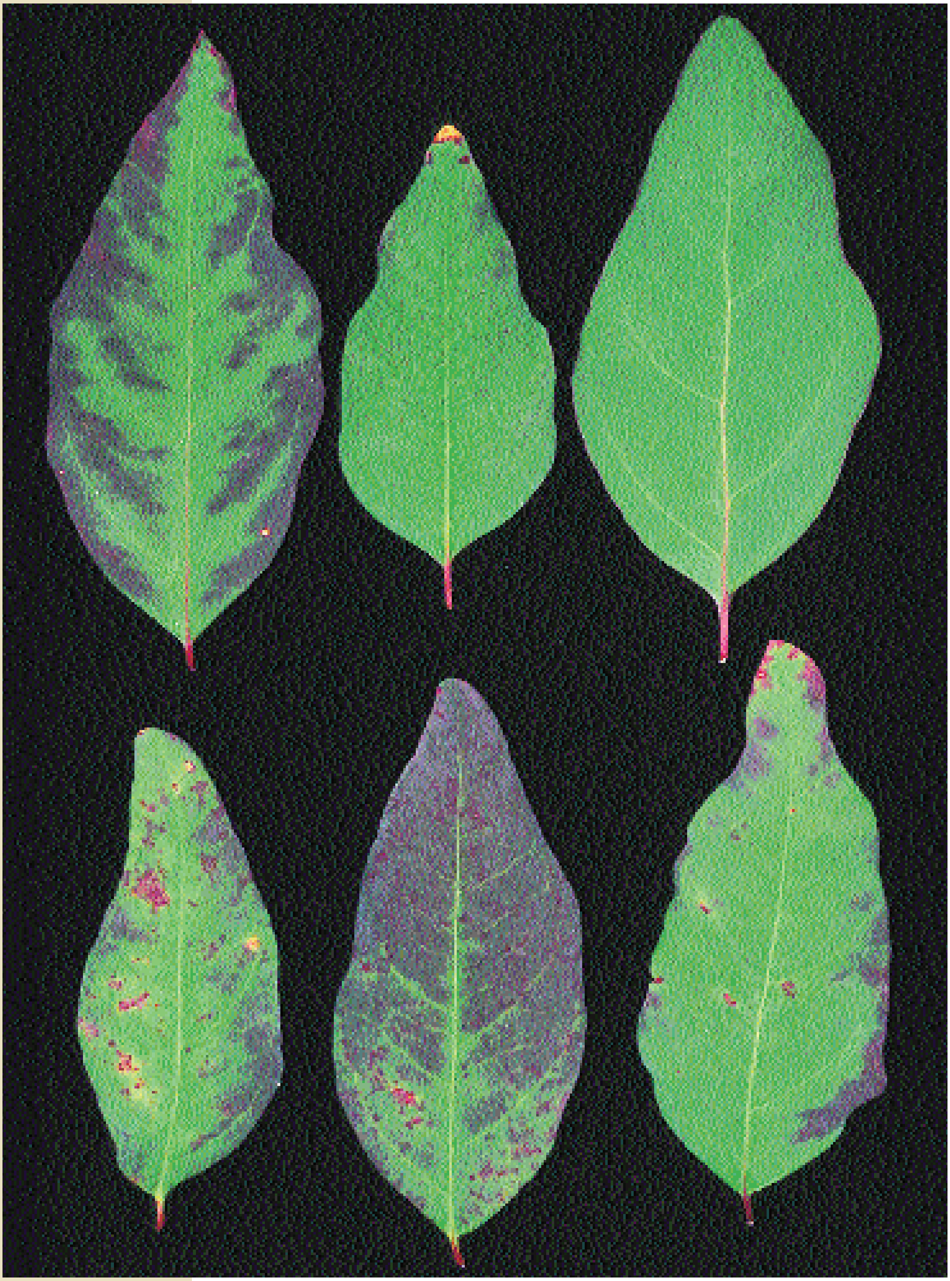


70. *E. grandis* × *E. camaldulensis*: Mg-deficient leaves on the right; healthy leaf on the left. Glasshouse.

71. *E. grandis* × *E. urophylla*: green tissue flanks the mid-rib in mature foliage of a Mg-deficient tree. Guangdong Province, P.R. China.

72. *E. maculata*: interveinal chlorosis leaving wide flanks of green tissue along the veins of mature leaves of Mg-deficient seedlings. Glasshouse.

73. *E. pellita*: leaves from Mg-deficient seedlings showing mild chlorosis and loss of reflective bloom in interveinal areas. Glasshouse.



74. *E. urophylla*: mature leaves of Mg-deficient seedlings with purple pigmentation and necrosis in interveinal areas. Glasshouse.

75. *E. urophylla*: branch from a Mg-deficient tree with symptoms in the older leaves. Indonesia.

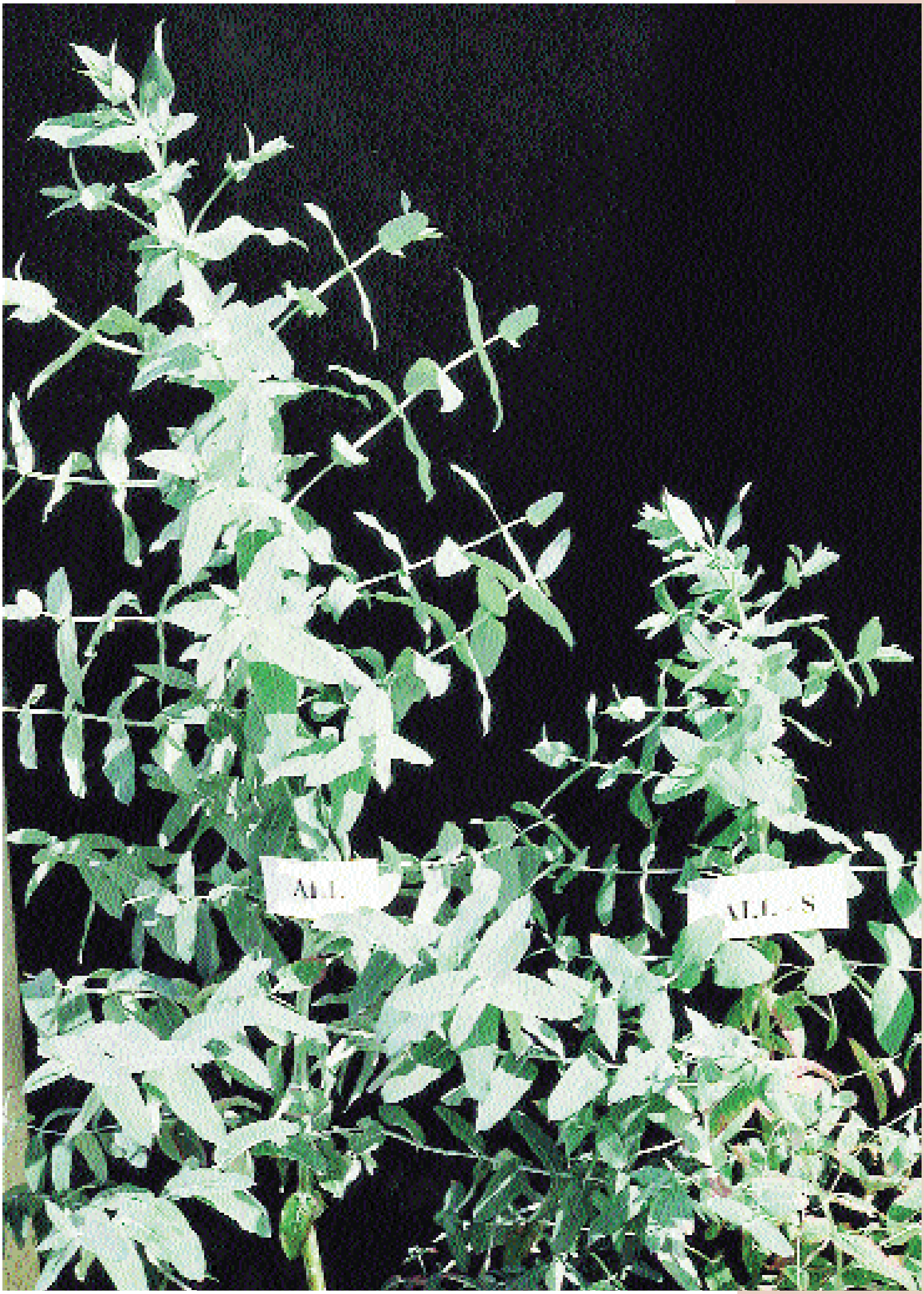
Sulfur

Function

Sulfur is essential for the formation of proteins containing the amino acids cysteine and methionine. Sulfur is also required for the synthesis of thiamin, co-enzyme A and sulfolipids. Sulfur is poorly phloem-mobile. Consequently, symptoms of S deficiency first appear on young leaves which become uniformly yellow as the chlorophyll content declines.

Symptoms

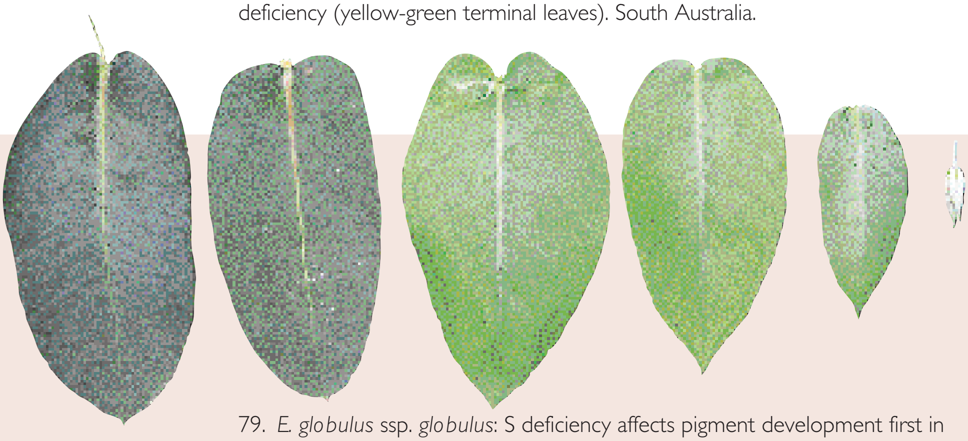
In S-deficient eucalypts the interveinal areas of expanding leaves turn pale green. With time, the leaves become uniformly yellow and symptoms spread from expanding to fully expanded leaves. In *E. globulus* vertical growth may be impaired before symptoms become obvious. Under severe deficiency the young leaves of *E. grandis, E. pellita* and *E. urophylla* become pale red, the leaf tips die and shrivel and the terminal bud aborts. Leaves which are uniformly yellow are indistinguishable from those suffering N deficiency. However, N deficiency symptoms first appear in mature leaves and the young leaves do not develop a red blush unless the N deficiency is quite severe.



76. *E. globulus* ssp. *globulus*: mild S deficiency (right) can reduce growth without the appearance of deficiency symptoms in juvenile foliage; normal plant (left). Glasshouse.

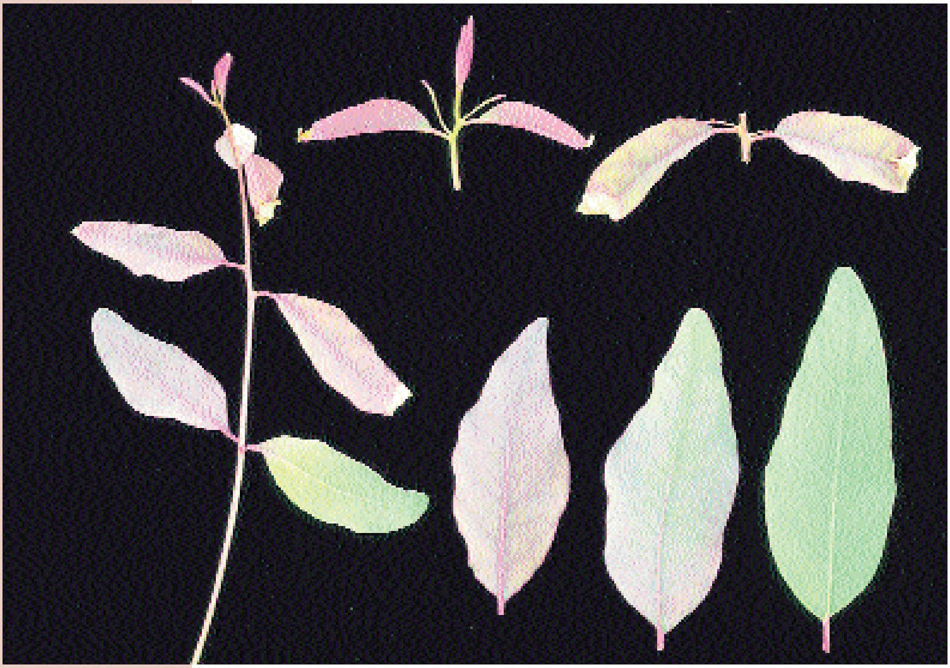
77. *E. globulus* ssp. *globulus*: yellowing of young leaves due to mild S deficiency in a one-year-old tree. Western Australia.

78. *E. globulus* ssp. *globulus*: branches from two-year-old tree with mild S deficiency (yellow-green terminal leaves). South Australia.

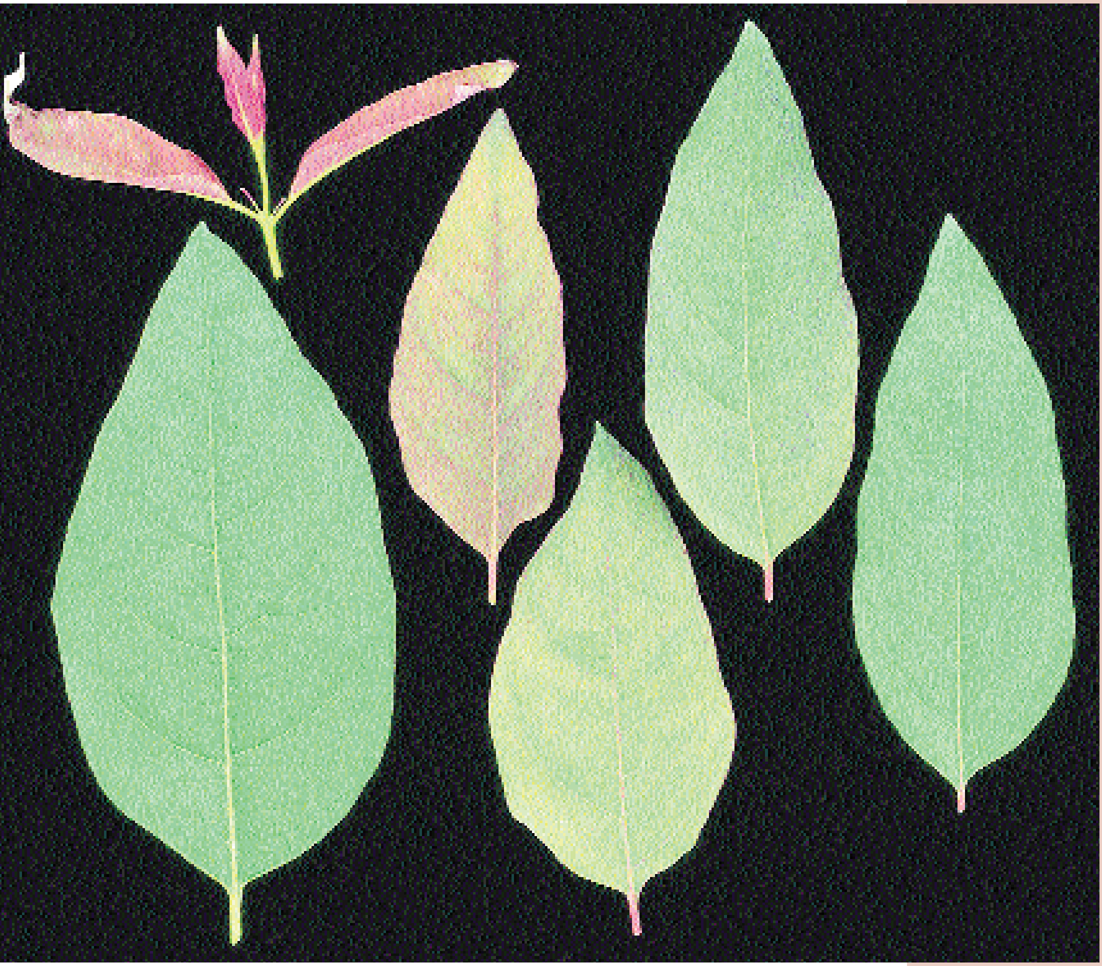


79. *E. globulus* ssp. *globulus*: S deficiency affects pigment development first in expanding leaves (centre) and later the leaves at the shoot apex. The leaves are from one branch. South Australia.

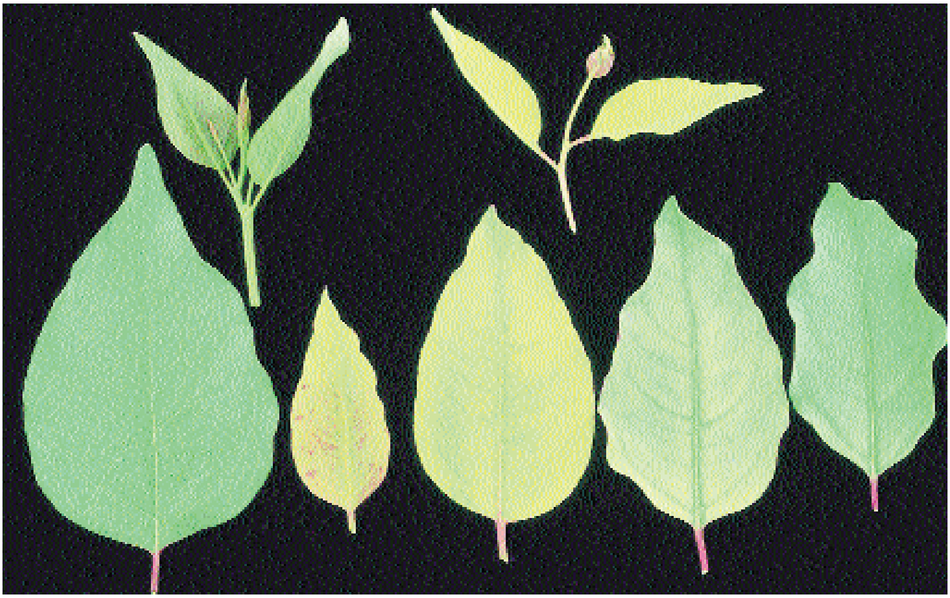
80. *E. grandis*: yellowing and purple pigmentation of young leaves of mildly S-deficient seedlings. Glasshouse.



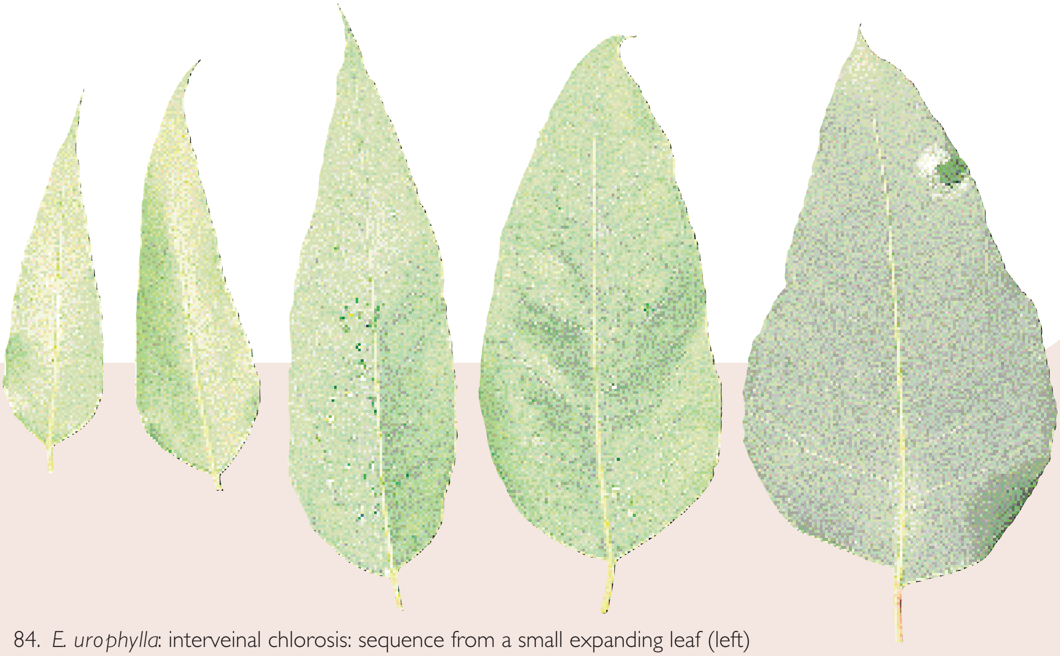
81. *E. grandis*: young leaves and shoot tips from severely S-deficient seedlings. The expanding leaves have necrotic tips and purple blades. Glasshouse.



82. *E. pellita*: yellowing, reddening and tip necrosis in young leaves of a S-deficient seedling: sequence from shoot tip (upper left) to mature leaf (right); normal mature leaf (lower left). Glasshouse.



83. *E. urophylla*: yellow expanding leaves and interveinal chlorosis in young fully expanded leaves of a S-deficient seedling (right); normal shoot tip and fully expanded leaf (left). Glasshouse.



84. *E. urophylla*: interveinal chlorosis: sequence from a small expanding leaf (left) to a fully expanded leaf (right) of S-deficient two-year-old tree. Guangdong Province, P.R. China.

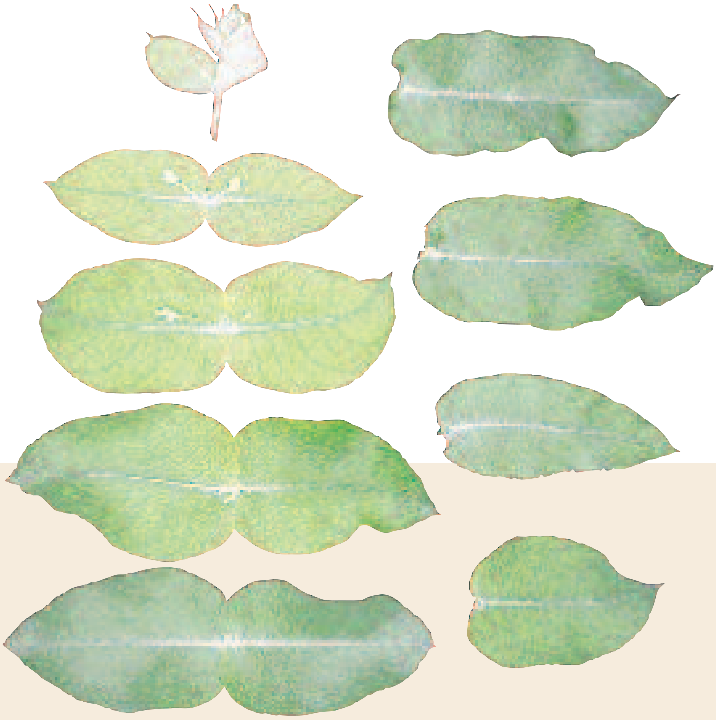
Iron

Function

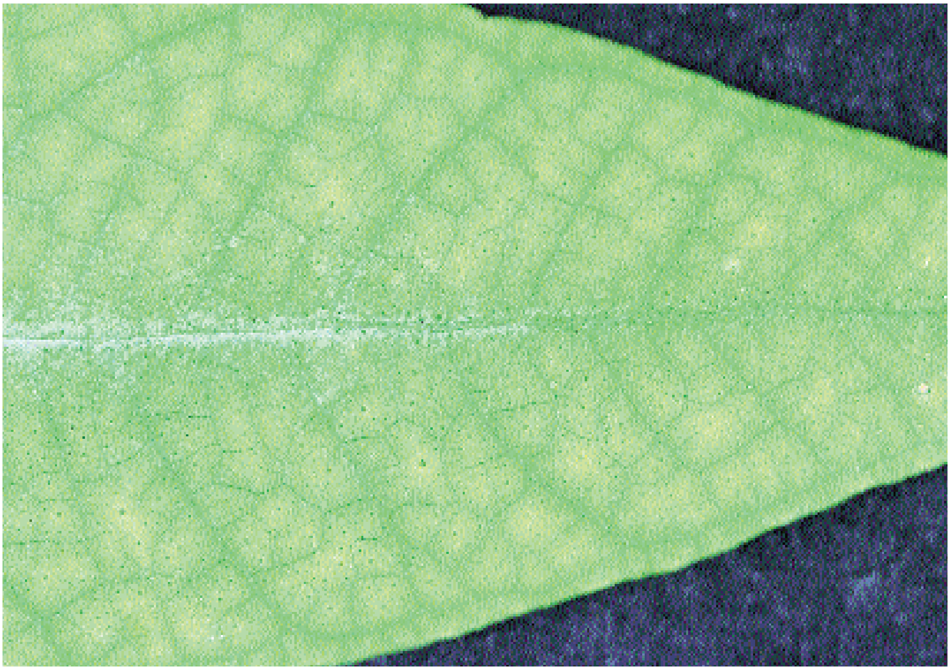
Iron is essential for the synthesis of chlorophyll. Since Fe does not move out of old leaves, even under deficiency, chlorosis of young leaves is an early symptom of Fe deficiency. Iron, with its reversible oxidation states (Fe^{2+}, Fe^{3+}), is involved in many redox reactions of photosynthesis and respiration. Much of the Fe in a leaf is bound to hemoproteins (e.g. the cytochromes and peroxidases). It is also a component of some Fe-S proteins such as ferredoxin.

Symptoms

At the early stage of Fe deficiency, the interveinal areas of expanding leaves develop a pale green chlorosis. As symptoms advance, the young leaves become intensely yellow and normal green tissue is restricted to the major and minor veins. This distinctive pattern, of yellow leaves with narrow green veins, is characteristic of Fe deficiency in all species of eucalypts. Chlorosis spreads from young to older leaves. In the field, there may be a sharp transition between chlorotic young foliage and dark green foliage of an earlier growth flush. In *E. globulus*, symptoms are often more severe in the lower third of the canopy.



85. *E. globulus* ssp. *globulus*: sequence of leaves on the main stem of an Fe-deficient seedling shows that symptoms are worse in young leaves. Glasshouse.



86. *E. globulus* ssp. *globulus*: detail of young Fe-deficient leaf with green veins and interveinal chlorosis. Western Australia. (Photo: W. van Aken).

87. *E. globulus* ssp. *globulus*: coppice with pale yellow leaves indicative of Fe deficiency. Yunnan Province, P.R. China.

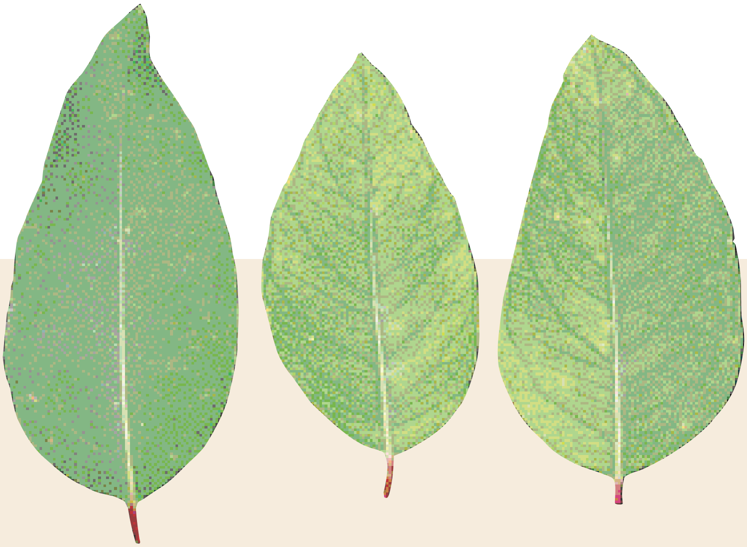
88. *E. globulus* ssp. *globulus*: young leaves with interveinal chlorosis due to secondary Fe deficiency. Detail of leaves in coppice of Plate 87. Yunnan Province, P.R. China.

89. *E. globulus* ssp. *globulus*: yellow adult foliage of two-year-old tree growing on calcareous soil. Victoria, Australia.

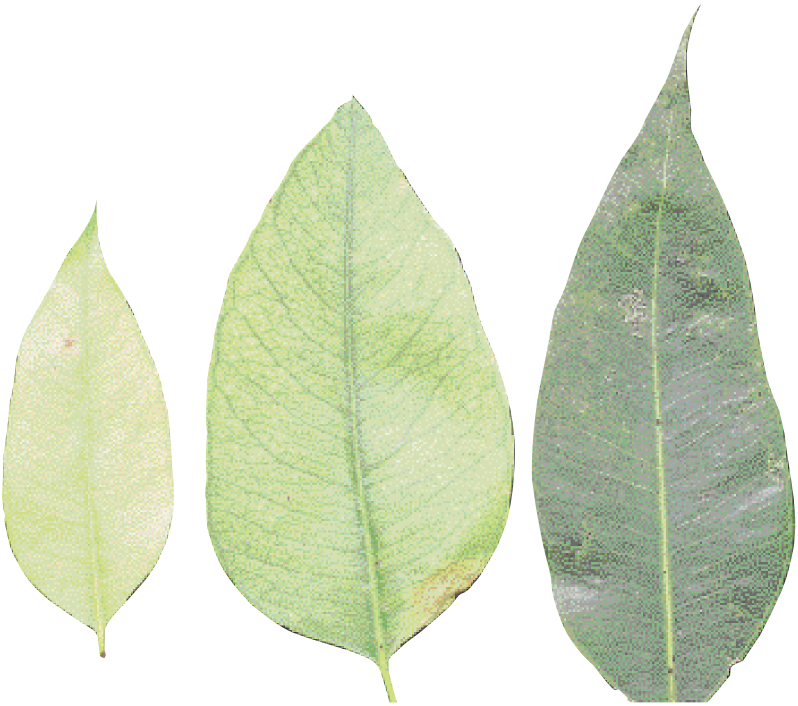
90. *E. grandis* × *E. urophylla*: interveinal chlorosis in young leaves of a mildly Fe-deficient two-year-old tree. Guangdong Province, P.R. China.

91. *E. maculata*: interveinal chlorosis and green veins in young leaves from Fe-deficient seedlings; normal leaf colour (right). Glasshouse.

92. *E. marginata*: mild Fe deficiency in young leaves. Western Australia.

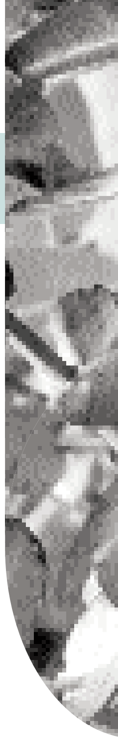


93. *E. urophylla*: early symptom of Fe deficiency in young leaves of three-year-old tree. Hainan, P.R. China.



94. *E. urophylla*: young leaves (left) with green veins and interveinal chlorosis, and mature leaf (right) from an Fe-deficient tree. Surigao Province, Philippines.

95. *E. urophylla*: iron deficiency induced in young leaves in a one-year-old tree growing on serpentine soil. Bukidnon Province, Philippines.



Copper

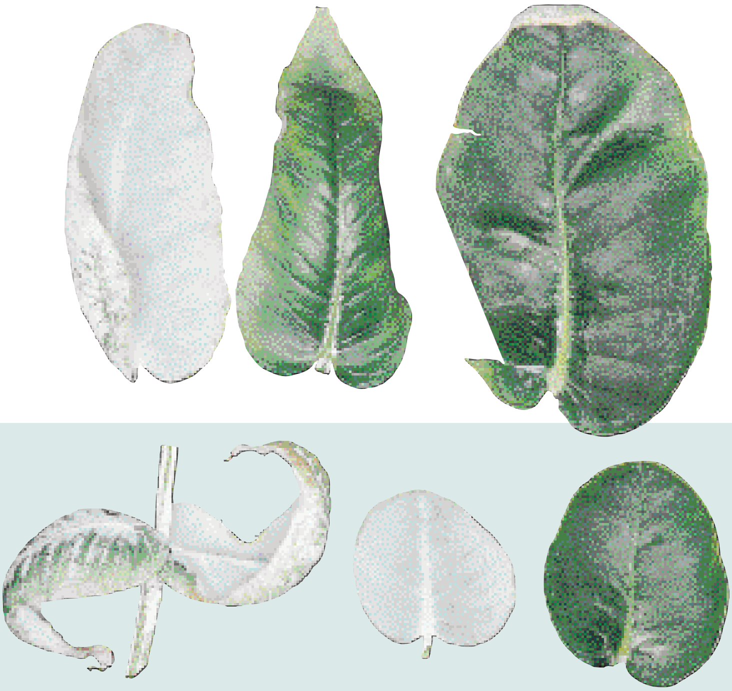
Function

Much of the Cu in the plant is bound to plastocyanin in the leaf. Copper is essential for photosynthesis since plastocyanin is a major component of the electron transport chain. Copper is also a constituent of a number of Cu-metalloenzymes such as cytochrome oxidase and phenolase. Phenolase is required for normal lignification of wood. Symptoms of Cu deficiency first appear in apical buds and expanding leaves, and the wood that develops is poorly lignified. Branches can be pendulous in some Cu-deficient eucalypts. Unlike in N deficiency, senescence of old leaves is not accelerated in Cu-deficient trees. Thus, Cu is poorly phloem-mobile in Cu-deficient trees.

Symptoms

Copper deficiency affects the young growth. Often the first sign of deficiency is the pendulous habit of lateral branches due to impaired lignification of wood in the young stem. This is exacerbated by high soil nitrogen. Expanding leaves become twisted, cupped or recurved and the margins may be irregular in outline. In *E. globulus* the young leaves also develop marginal chlorosis and some necrosis which becomes obvious when the surface waxes are removed. Chlorosis and necrosis also occur in *E. urophylla*. In advanced Cu deficiency, the shoot apex dies back, axillary buds are shed and dwarf, multistemmed trees form without a main leader. In *E. maculata*, and sometimes in *E. globulus*, nodes become enlarged due to the formation of large numbers of short-lived lateral branches. Severely Cu-deficient trees are vulnerable to invasion by pathogenic canker-forming fungi and other organisms. Under conditions of high evaporative demand, Cu-deficient trees wilt and may lose their leader more readily than Cu-adequate trees.

96. *E. globulus* ssp. *globulus*: shoot from the upper crown of a two-year-old Cu-deficient tree. Western Australia.



97. *E. globulus* ssp. *globulus*: reverse rolling and tip necrosis of young leaves from a Cu-deficient two-year-old tree. The surface waxes have been removed from three leaves to reveal marginal and interveinal chlorosis. Western Australia.

98. *E. globulus* ssp. *globulus*: seasonal production of small leaves is one of the many symptoms of Cu deficiency in young trees. Western Australia.

99. *E. globulus* ssp. *globulus*: recurved lateral branches of a Cu-deficient seedling (lower); normal branches (upper). Glasshouse.

100. *E. globulus* ssp. *globulus*: stem twisting (speed-wobbles) and pendulous branches in a one-year-old Cu-deficient tree. Western Australia.

101. *E. globulus* ssp. globulus: canopy thinning in Cu-deficient three-year-old tree. Western Australia.

102. *E. globulus* ssp. *globulus*: malformed and aborted leaf development in four-year-old Cu-deficient tree. Western Australia.

103. *E. globulus* ssp. *globulus*: wilting of the canopy apex in a Cu-deficient tree during hot windy weather in late summer. Western Australia.

104. *E. grandis*: Cu-deficient seedling with twisted leaves, dead axillary shoots and pale green expanding leaves. Glasshouse.

105. *E. maculata*: branch of a Cu-deficient tree with enlarged nodes bearing numerous shoots. Western Australia.

106. *E. maculata*: malformed leaves in a two-year-old Cu-deficient tree. Western Australia.

107. *E. nitens*: pendulous branches and bud death of a Cu-deficient tree. Tasmania. (Photo: R. Cromer).

108. *E. urophylla*: dark green, malformed leaves in a Cu-deficient one-year-old tree. Indonesia.

Zinc

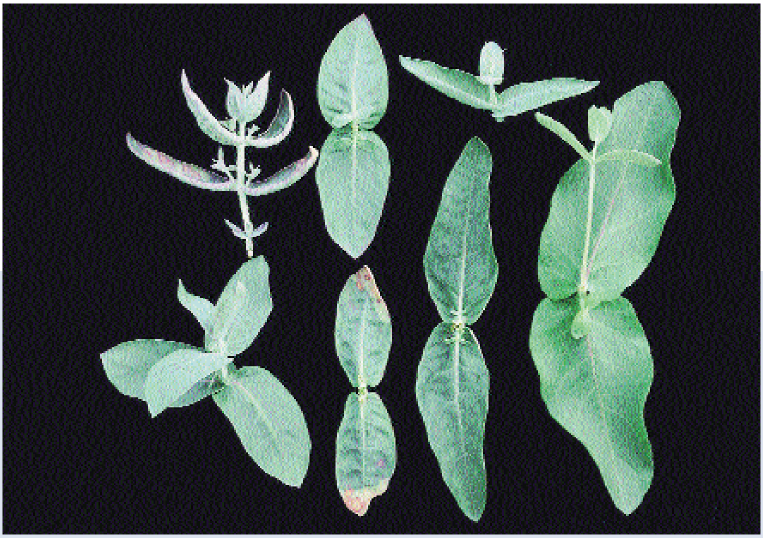
Function

Zinc is a constituent of some metallo-enzymes, including carbonic anhydrase and alcohol dehydrogenase, and is required for the activity of many other enzymes. Zinc is also required for photosynthesis. Reduced leaf size and shortened internodes in Zn-deficient trees is related to the requirement of Zn for the synthesis of auxin, a growth hormone that facilitates cell expansion. Zinc is not readily redistributed from old leaves so symptoms of deficiency (small leaves with some interveinal yellowing) first appear in expanding leaves.

Symptoms

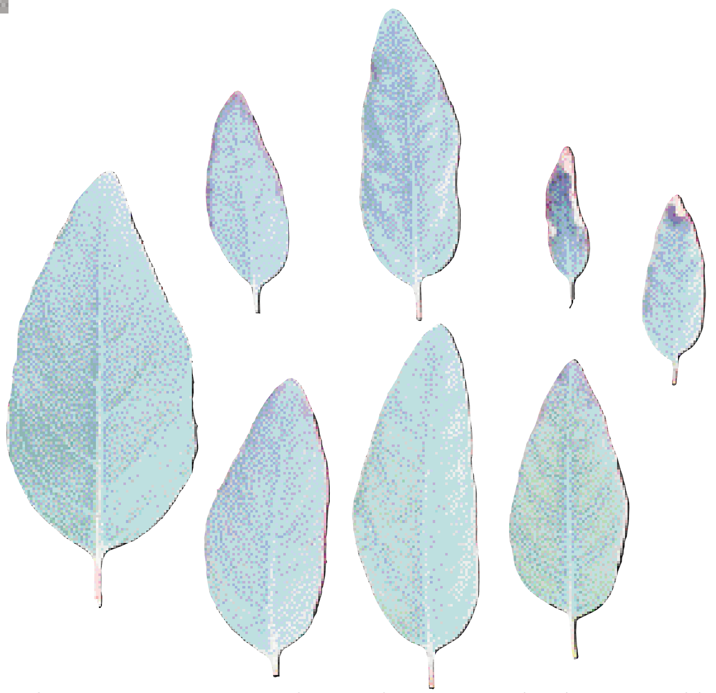
Zinc-deficient trees are stunted and the leaves are small and crowded. In *E. grandis, E. pellita* and *E. urophylla*, expanding leaves initially develop interveinal chlorosis. Purple areas may also appear. With increasing severity of deficiency, the new leaves decrease in size and the internodes shorten. Leaf tips and interveinal patches of tissue may become necrotic. Some of the necrosis can be attributed to the accumulation of high concentrations of P in heavily fertilised trees. In juvenile *E. globulus* foliage, chlorosis is masked by the surface waxes and the first signs of Zn deficiency are leaf stunting and leaf rolling.

109. *E. globulus* ssp. *globulus*: crowded leaves near the apex of the main stem of a Zn-deficient seedling. Glasshouse.

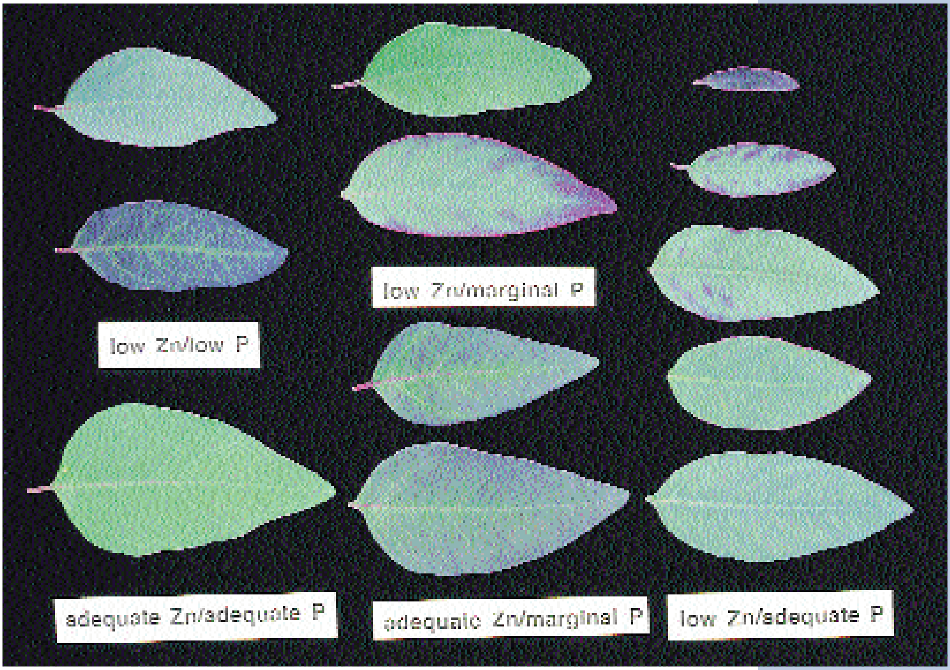


110. *E. globulus* ssp. *globulus*: shoot tips and expanding leaf pairs of a Zn-deficient seedling showing stunting, leaf-rolling, patches of interveinal chlorosis and tip necrosis. Glasshouse.

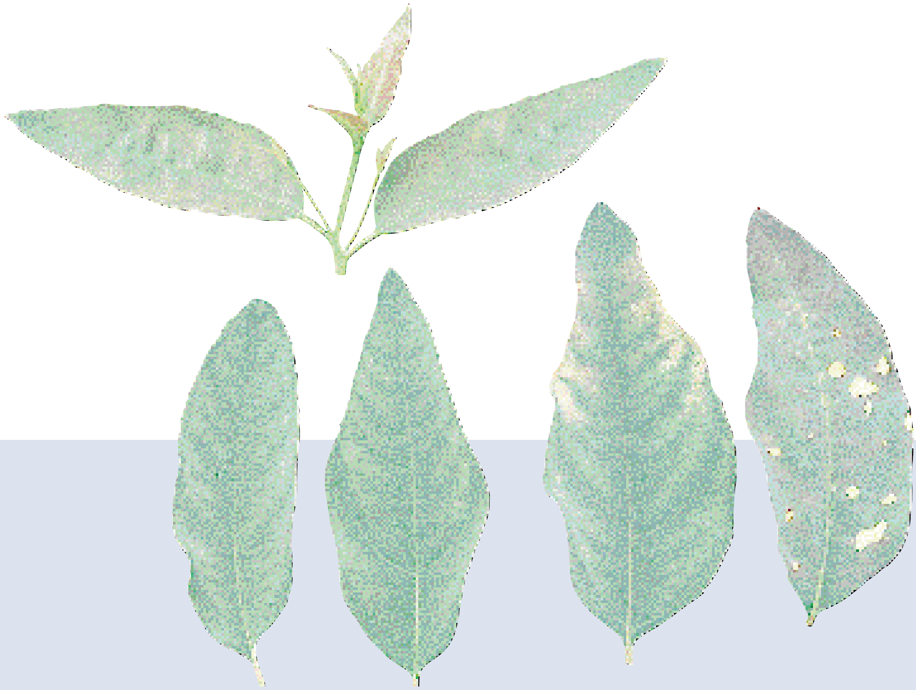
111. *E. globulus* ssp. *globulus*: young leaves are small in this two-year-old Zn-deficient tree. Western Australia.



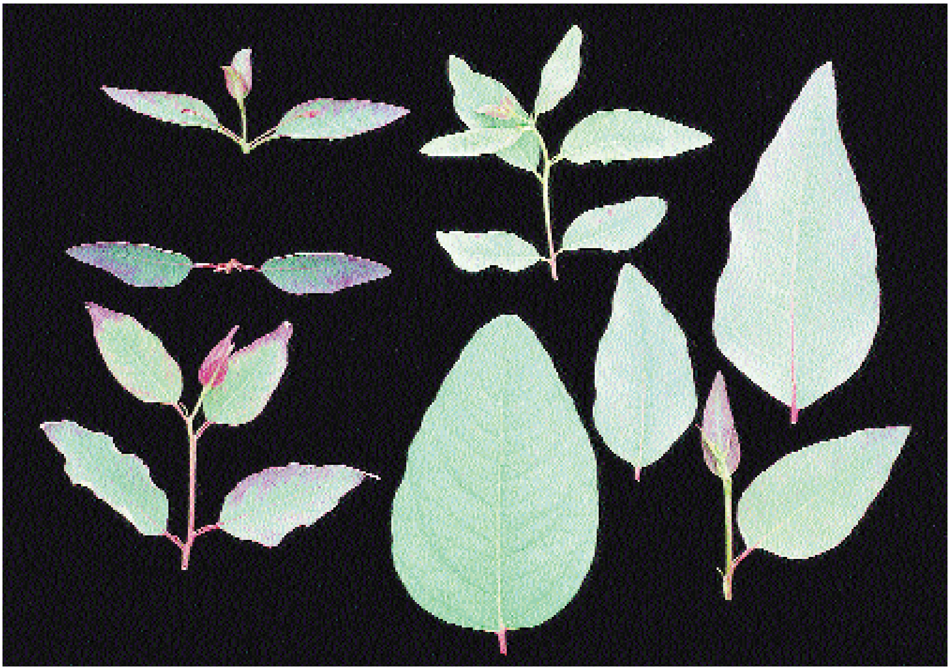
112. *E. grandis*: young leaves of a Zn-deficient seedling (right); normal leaf (left). Interveinal chlorosis extends between the dark green veins, the leaves are smaller than normal and the tips become necrotic in severely deficient plants. Glasshouse.



113. *E. grandis*: P-Zn interactions influence symptom expression in young leaves. The formation of purple blotches (which later become necrotic) is associated with the accumulation of P in leaf tissue. Glasshouse.



114. *E. pellita*: young leaves from Zn-deficient seedling with chlorosis extending from the margins between dark green veins (left and centre). In severely deficient plants, purple patches form and become bleached (right). Glasshouse.

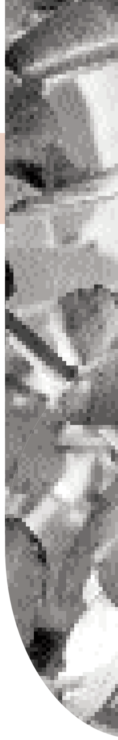


115. *E. urophylla*: dwarf young leaves with purple pigmentation and tip necrosis from a Zn-deficient seedling; normal leaf (lower centre). Glasshouse.

116. *E. urophylla*: stunted Zn-deficient one-year-old tree. Bukidnon Province, Philippines.

117. *E. urophylla*: dwarf leaves with purple areas and necrotic spots on a one-year-old Zn-deficient tree. Bukidnon Province, Philippines.

Manganese



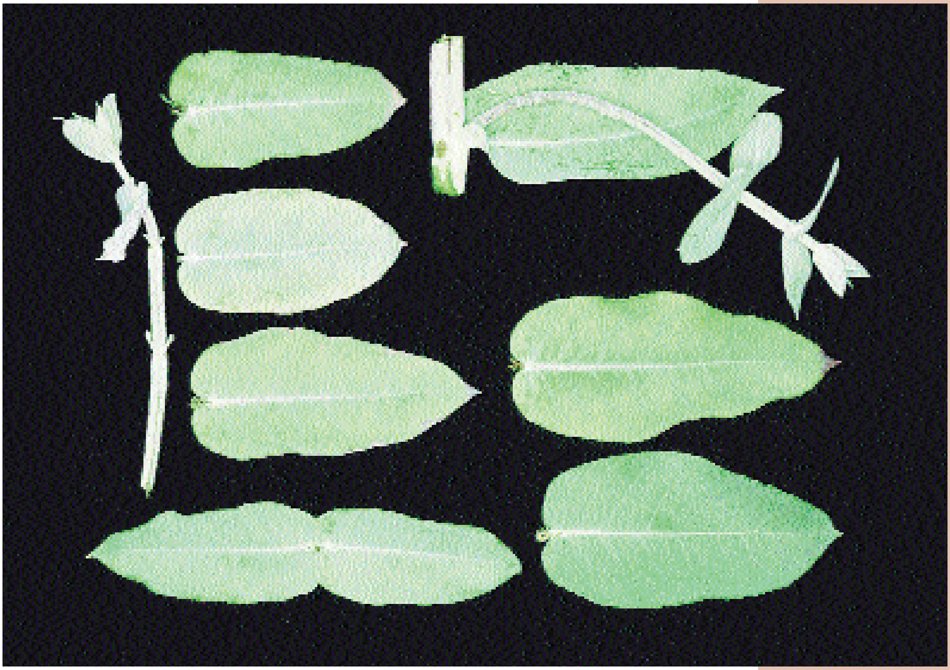
Function

Manganese is required for the evolution of oxygen from the splitting of water in photosynthesis. It is also involved in redox reactions and electron transport in chloroplasts. Manganese is essential for a few metallo-proteins such as superoxide dismutase. In deficient plants, necrosis develops in recently expanded and expanding leaves, and lignification of wood may be impaired. Hence, there is limited movement of Mn in the phloem from old leaves towards the shoot apex.

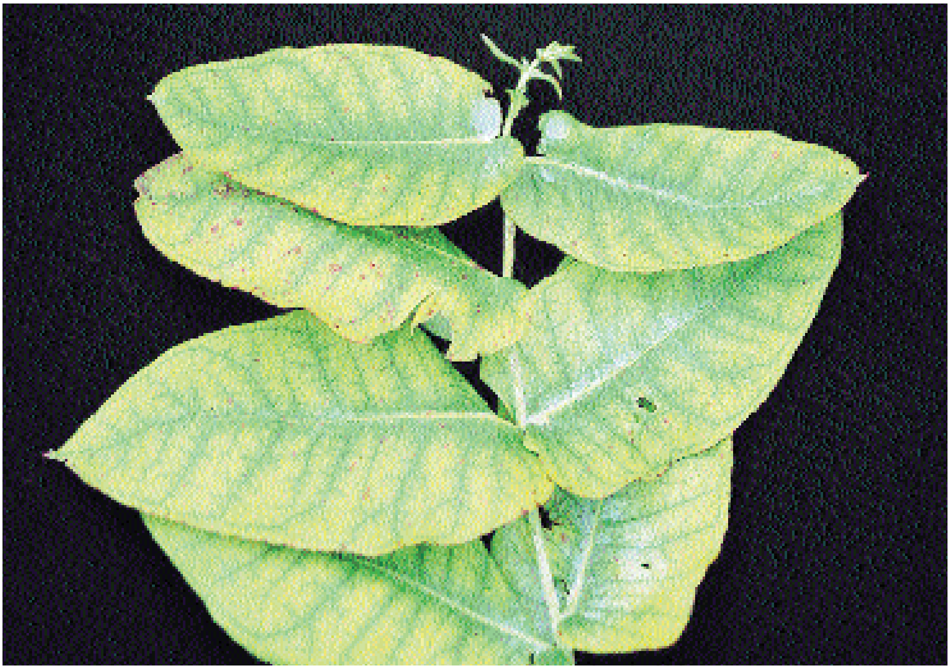
Symptoms

The margins of juvenile and adult expanding leaves become pale green. Chlorosis extends between the lateral veins towards the midrib. Leaves are normal in size. With time, the chlorotic tissue becomes yellow. The major veins are always flanked with wide margins of green tissue. Symptoms spread from young to mature leaves. Reduced lignification of stem wood has been observed in *E. globulus* causing lateral branches to weep. Terminal meristems can die in severely deficient trees and the resulting die-back symptoms can be confused with Cu deficiency.

118. *E. camaldulensis*: sequence of leaves from near the shoot apex (left) along a branch to a mature leaf (right) with chlorosis due to Mn deficiency. Western Australia.



119. *E. globulus* ssp. *globulus*: interveinal chlorosis, leaf death, leaf tip necrosis and recurved lateral branches of Mn-deficient seedlings. Glasshouse.



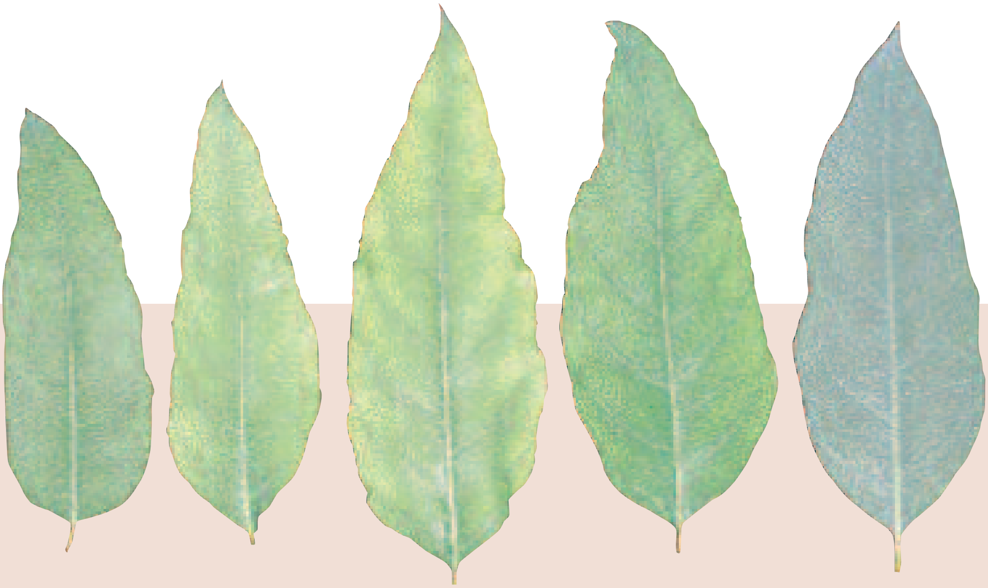
120. *E. globulus* ssp. *globulus*: apical necrosis and interveinal chlorosis in juvenile foliage of a Mn-deficient tree. Western Australia. (Photo: I. Dumbrell).

121. *E. globulus* ssp. *globulus*: marginal and interveinal chlorosis on adult (left) and intermediate leaves (right) of a Mn-deficient two-year-old tree. Surface wax has been removed from two of the intermediate leaves. Western Australia.

122. *E. globulus* ssp. *globulus*: shoot at the transition from juvenile to adult foliage of a Mn-deficient tree. Western

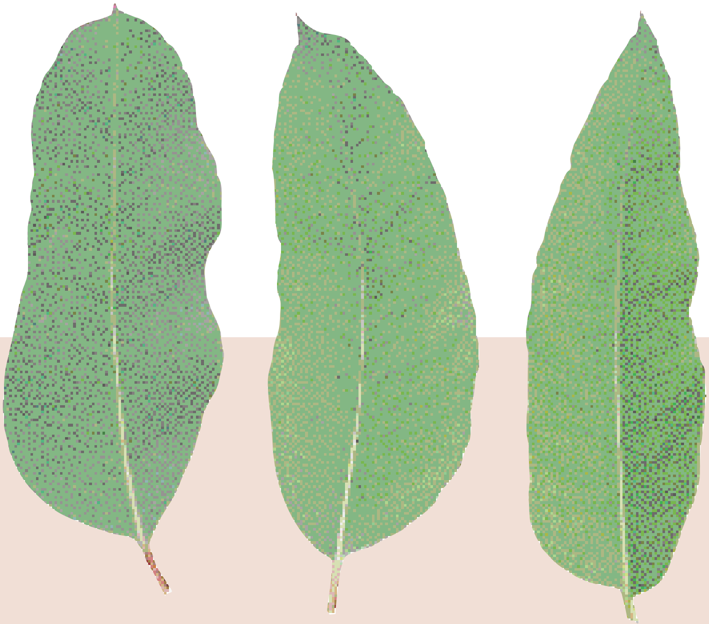
123. *E. globulus* ssp. *globulus*: adult foliage with marginal chlorosis due to Mn deficiency. Western Australia. (Photo: I. Dumbrell).

124. *E. globulus* ssp. *globulus*: branch from Mn-deficient tree showing that symptoms develop first in young foliage. Western Australia.(Photo: I. Dumbrell).



125. *E. grandis*: symptoms of Mn deficiency first appear in expanding leaves (centre) and spread to younger (left) and older (right) leaves. Glasshouse.

126. *E. grandis*: marginal chlorosis in adult leaves from a Mn-deficient four-year-old tree. Western Australia

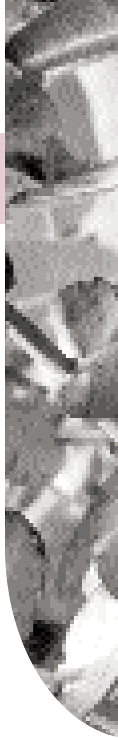


127. *E. grandis* × *E. camaldulensis*: interveinal chlorosis (right) in young leaves of a Mn-deficient seedling; normal leaf (left). Glasshouse.

128. *E. maculata*: marginal chlorosis due to Mn deficiency (centre and right); normal leaf (left). Glasshouse.

129. *E. urophylla*: young leaves of Mn-deficient seedlings develop interveinal chlorosis (centre) followed by white necrotic spots (right); normal leaf (left). Glasshouse.

Boron



Function

The role of B in cell metabolism is still being unravelled. Boron is required for cell division, cell growth and possibly membrane function. Much of the B in the plant is located in the cell wall where it forms a structural borate-rhamnogalacturonan II complex. Like Ca, B that is delivered in the transpiration stream to the shoot in eucalypts is not retranslocated in the phloem. Hence, growth of the shoot and root tips is seriously impaired in B-deficient trees. Shoot tips may die leading to multiple lateral branches, and reduced lignification of the wood may cause branches to weep.

Symptoms

Boron deficiency severely impairs growth at the shoot tip. Initially there is a change in pigmentation in the young leaves; either the accumulation of purple pigments basipetally along the margins (*E. grandis*) or chlorosis. Yellowing can extend from the leaf margin interveinally over the whole blade (*E. grandis* × *E. urophylla*), be confined to the margins (*E. camaldulensis, E. pellita, E. tereticornis*) or extend over the whole leaf except for the basal region (*E. globulus, E. urophylla*). Developing leaves may be malformed with missing sectors at the margin or within the blade. The first signs of deficiency in *E. globulus* are leaf rolling and weeping branches. In severely deficient trees, all the branches can be prostrate because the stems do not have enough lignin in the wood to support the foliage. Shoots die back from the growing points. The remaining upper nodes become enlarged and bear multiple, short-lived shoots (*E. deglupta, E. grandis, E. urophylla*). In *E. robusta* and *E. urophylla*, the leaves are often brittle and have raised corky tissue associated with the veins.

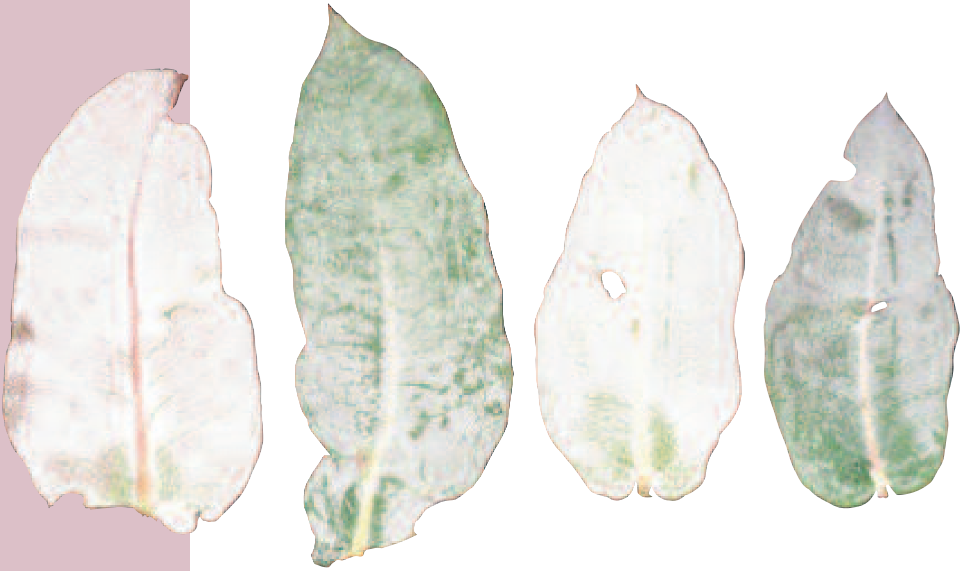
130. *E. camaldulensis*: marginal and interveinal necrosis, and leaf splitting in a B-deficient two-year-old tree. Philippines.

131. *E. deglupta*: branch from a one-year-old tree showing chlorosis, leaf distortion and leaves with missing segments. Bukidnon Province, Philippines.

132. *E. deglupta*: shoot dieback and multiple budding of a B-deficient two-year-old tree. Bukidnon Province, Philippines.

133. *E. globulus* ssp. *globulus*: B-deficient six-month-old tree with prostrate branches and rolled leaves. Yunnan Province, P. R. China.

134. *E. globulus* ssp. *globulus*: deformed leaves of a B-deficient two-year-old tree. Yunnan Province, P.R. China.



135. *E. globulus* ssp. *globulus*: B-deficient leaves with small windows. Victoria, Australia.

136. *E. globulus* ssp. *globulus*: apical and marginal chlorosis of a young leaf from a B-deficient tree. Yunnan Province, P.R. China.

137. *E. globulus* ssp. *globulus*: branch from a two-year-old tree showing B deficiency symptoms that were expressed during the dry season (centre of branch). The leaves that developed during the subsequent wet season were normal. Yunnan Province, P.R. China.

138. *E. globulus* ssp. *globulus*: chlorosis and marginal splitting in adult leaves from a three-year-old B-deficient tree. Western Australia.

139. *E. globulus* ssp. *globulus*: boron deficiency causes the accumulation of phenolic compounds that oxidise when the bark is removed. Top – wood from tree with adequate B fertiliser, centre – marginal B, bottom – B-deficient stem. Yunnan Province, P.R. China.

140. *E. globulus* ssp. *maidenii*: twig die-back of a B-deficient tree. Yunnan Province, P.R. China.

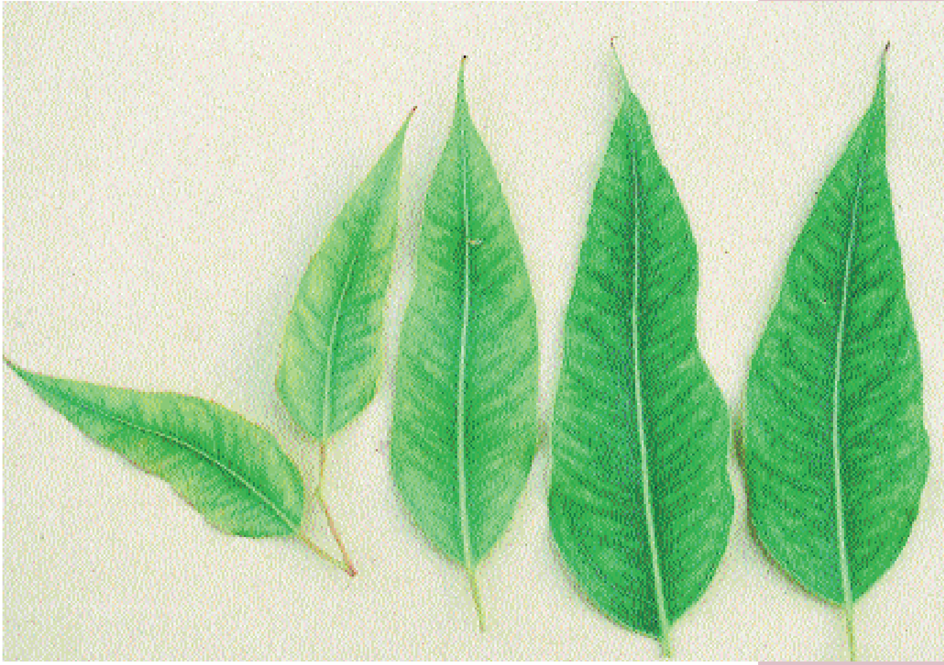
141. *E. globulus* ssp. *maidenii*: misshapen leaves of B-deficient coppice. Yunnan Province, P.R. China.

142. *E. grandis*: B-deficient six-month-old tree with necrosis of young leaves, purple pigmentation of expanded leaves and bud death. Guangdong Province, P.R. China.

143. *E. grandis*: purple pigmented leaves with a triangular area of green tissue from a B-deficient six-month-old tree. Guangdong Province, P.R. China

144. *E. grandis*: enlarged nodes, multiple shoots, and bud death of a severely B-deficient six-month-old tree. Guangdong Province, P.R. China.

145. *E. grandis* x *E. camaldulensis*: misshapen leaves with areas of chlorosis from B-deficient seedling. Glasshouse.



146. *E. grandis* x *E. urophylla*: young leaves with interveinal chlorosis from a B-deficient two-year-old tree. Guangdong Province, P.R. China.

147. *E. pellita*: loss of leaves and twig die-back of a B-deficient tree. Guangdong Province, P.R. China.

148. *E. pellita*: marginal and interveinal chlorosis in a young expanded leaf from a B-deficient tree. Guangdong Province, P.R. China.

149. *E. robusta*: misshapen young leaves, and marginal and apical chlorosis in leaves of a B-deficient two-year-old tree. Yunnan Province, P.R. China

150. *E. smithii*: apical and marginal purpling in B-deficient leaves of a six-month-old tree. Yunnan Province, P.R. China

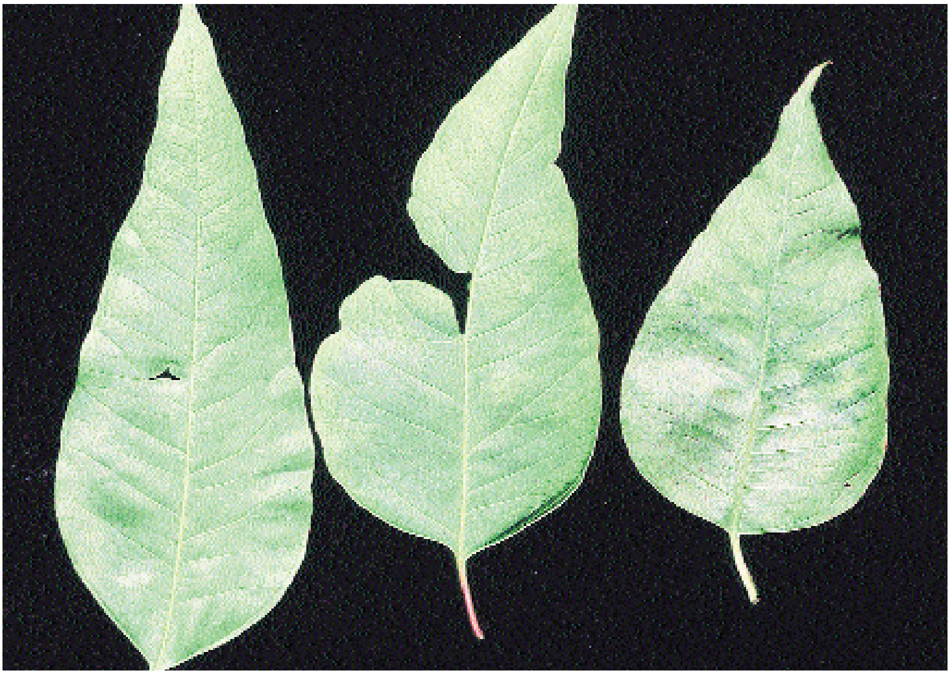
151. *E. tereticornis*: leaves of a B-deficient three-year-old tree with marginal necrosis and incomplete wavey blades. Guangdong Province, P.R. China.

152. *E. urophylla*: tip death and multiple shooting from axillary buds of a B-deficient two-year-old tree. Guangdong Province, P.R. China.

153. *E. urophylla*: one-year-old tree with severe leaf symptoms of B deficiency. Guangdong Province, P.R. China.

154. *E. urophylla*: shoot tip from a severely B-deficient tree showing multiple shooting from axillary buds. Guangdong Province, P.R. China.

155. *E. urophylla*: raised areas of brown corky tissue over lateral veins on the abaxial surface of old leaves. Guangdong Province, P.R. China.



156. *E. urophylla*: boron-deficient leaves may develop small windows or sectors of the leaf blade may fail to develop. Guangdong Province, P.R. China.

157. *E. urophylla*: boron deficiency is more severe in the dry season, as shown by the chlorotic leaves in the centre of the branch. Philippines.

158. *E. urophylla*: sometimes the early symptoms of B deficiency are similar to Fe deficiency. Philippines.

159. *E. ABL 12 W5 clone* (left) is better able to grow on soils deficient in B than *E. Leizhou No. 133 clone* (right). Guangdong Province, P.R. China.

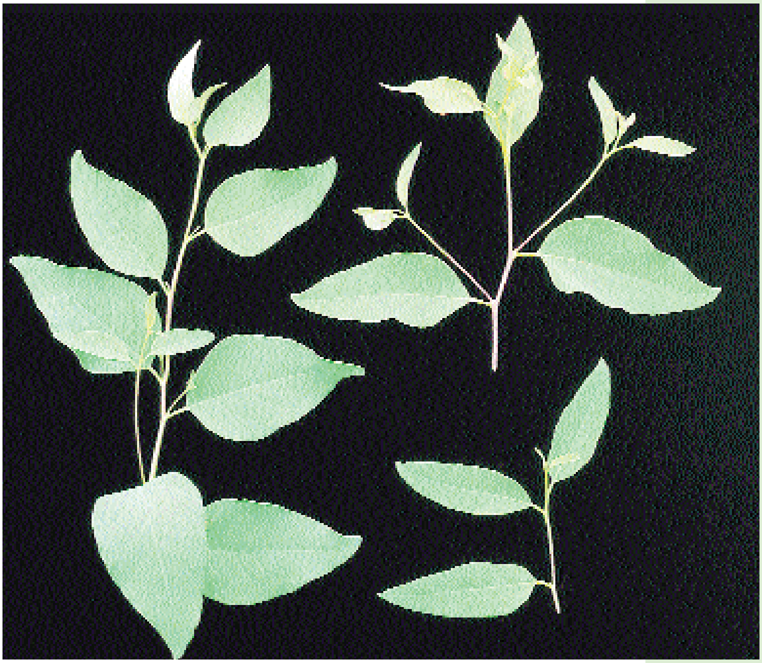
Molybdenum

Function

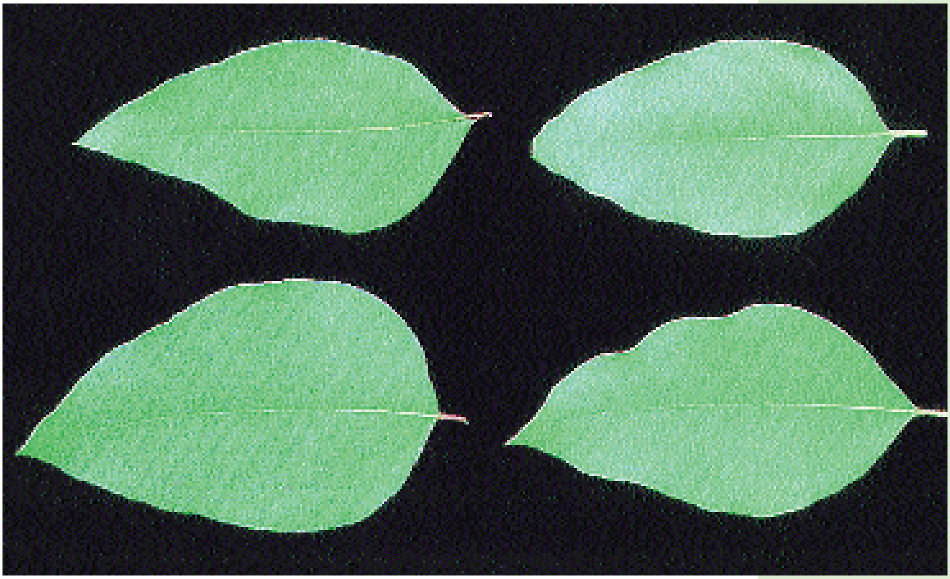
Molybdenum is essential for the action of nitrate reductase, one of the two enzymes required for the reduction of nitrate to ammonium. It is required in eucalypts where inorganic N is taken up as nitrate.

Symptoms

Molybdenum deficiency has only been observed in seedlings grown on acidic soil where N has been supplied as nitrate. Deficiency symptoms (interveinal chlorosis) resemble the early symptoms of N deficiency but, unlike those of N deficiency, they appear first in young leaves.



160. *E. grandis*: yellow young leaves of Mo-deficient seedlings supplied with nitrate-N. Glasshouse.



161. *E. urophylla*: young expanded leaves with light green interveinal areas of a Mo-deficient seedling supplied with nitrate-N. Glasshouse.

6. Toxicities and non-nutritional symptoms

Earlier we suggested (Fig. 1) that factors other than nutrient deficiencies can cause visible disorders in eucalypts. Since many of these can be mistaken for nutrient deficiencies, a select few are illustrated here. It is important to realise that this is not a complete panorama of possible disorders, rather they illustrate stresses that we have encountered in the field.

The principles outlined in Chapter 2 also apply when interpreting non-deficiency symptoms. Early stages of leaf necrosis caused by fertiliser toxicity (Plate 163), sodic soils (Plate 164) and some air pollutants (Plate 173) can resemble acute K deficiency (Plate 59). However, as stress increases, and leaf and branch death occurs, the resemblance to K deficiency diminishes. Where sudden tree decline occurs in patches (Plate 178), likely causes are drought, salinity, waterlogging or disease. In addition to pathogenic fungi, insects can damage roots and girdle the collar region of young trees. Where the tops of trees are damaged and then recover, frost (Plate 181) is a possible cause. Examine symptoms in the understorey as they can help to confirm diagnosis for air pollutants.

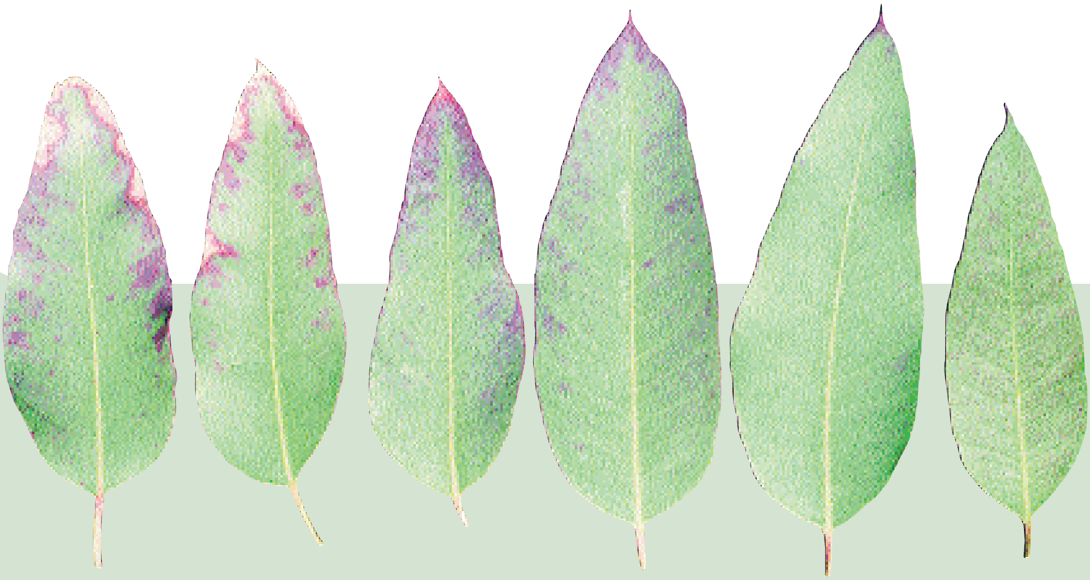
Although nutrient deficiency symptoms on a leaf may superficially resemble necrosis from insect and fungal attack (Plates 187, 193), they are readily distinguished with a hand lens (x5 magnification). Examine both sides of the leaf. Features to look for include small pigmented fungal fruiting bodies in leaf spots, aerial fungal hyphae, insect eggs, larvae inside leaf blisters (Plate 187), insect frass, bite marks, exudation fluids and stem cankers (Plate 197). Be careful when interpreting old damage as the primary agent resulting in the initial attack may have long disappeared.

Note that trees suffering from a nutrient disorder are also vulnerable to non-nutritional problems. Individual leaves may, therefore, exhibit a number of unrelated symptoms (Plate 195). Another factor to consider is the frequency of occurrence of a particular symptom in a plantation. The occasional dwarf tree with small leaves is more likely to result from genetic factors (Plate 182) than Zn deficiency. Natural variegation (Plate 184), another genetic disorder, is also expressed in the nursery.

Where heavy metal toxicities are suspected these can be confirmed with analysis of leaves and roots. However, unless the element causing the toxicity is measured, the conclusion drawn may be invalid. It is not uncommon for Fe (in eucalypts) or N (in acacias) to be diagnosed as the cause of yellowing in plantations on ultramafic soils where excessive Ni is the primary factor limiting growth (Dell 1997; Plates 168, 171).

Nutrient and metal toxicities

162. *E. grandis* x *E. urophylla*: abnormal leaf pattern due to excess application of boron. Guangdong Province, P.R. China.



163. *E. grandis* x *E. urophylla*: tip burn of old leaves due to excess application of boron. Guangdong Province, P.R. China.

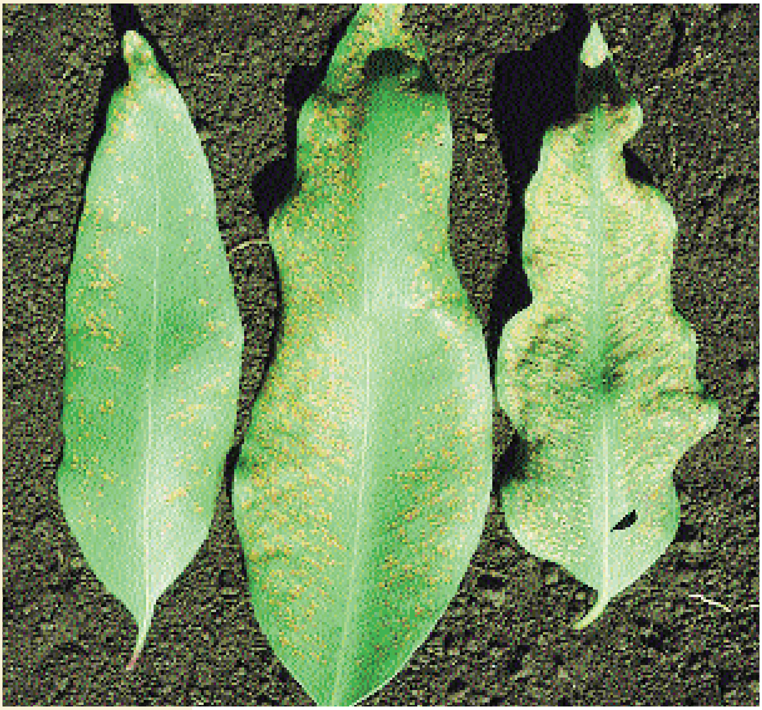
164. *E. globulus* ssp. *globulus*: marginal and tip burn of mature leaves from a tree growing in sodic soil. Western Australia.

165. *E. calophylla*: marginal burn from sea spray. Western Australia.

166. *E. camaldulensis*: symptoms of Ni toxicity in a one-year-old tree growing on ultramafic rocks. Bukidnon Province, Philippines.

167. *E. grandis*: symptoms of Ni toxicity in a one-year-old tree growing on ultramafic rocks. Bukidnon Province, Philippines.

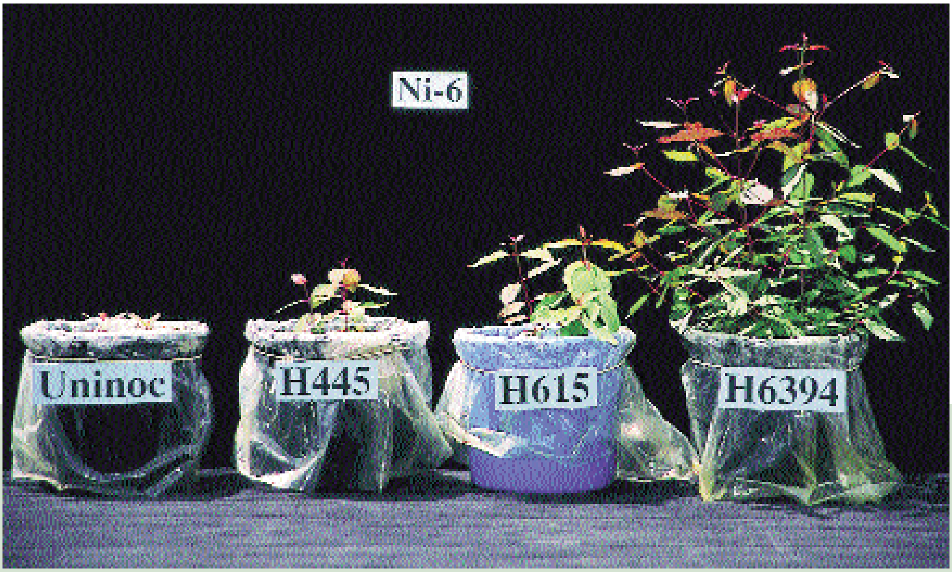
168. *E. pellita*: symptoms of Ni toxicity in a one-year-old tree growing on ultramafic rocks. Bukidnon Province, Philippines.



169. *E. tereticornis*: symptoms of Ni toxicity in a one-year-old tree growing on ultramafic rocks. Bukidnon Province, Philippines.

170. *E. urophylla*: symptoms of Ni toxicity in a one-year-old tree growing on

171. Eucalypt species selection trial on serpentine soil showing superior growth of *E. tereticornis*. Bukidnon Province, Philippines.



172. *E. urophylla*: certain Ni-tolerant fungi can form beneficial associations with the roots of eucalypts resulting in increased growth and reduced toxicity. The pot on the right was inoculated with an isolate of *Pisolithus* collected from the residue from a nickel mine. The pot on the left was uninoculated and those in the centre were inoculated with fungi from habitats low in Ni. The soil contains 6 mg Ni/kg. (Photo: N. Aggangan).

Air pollutants

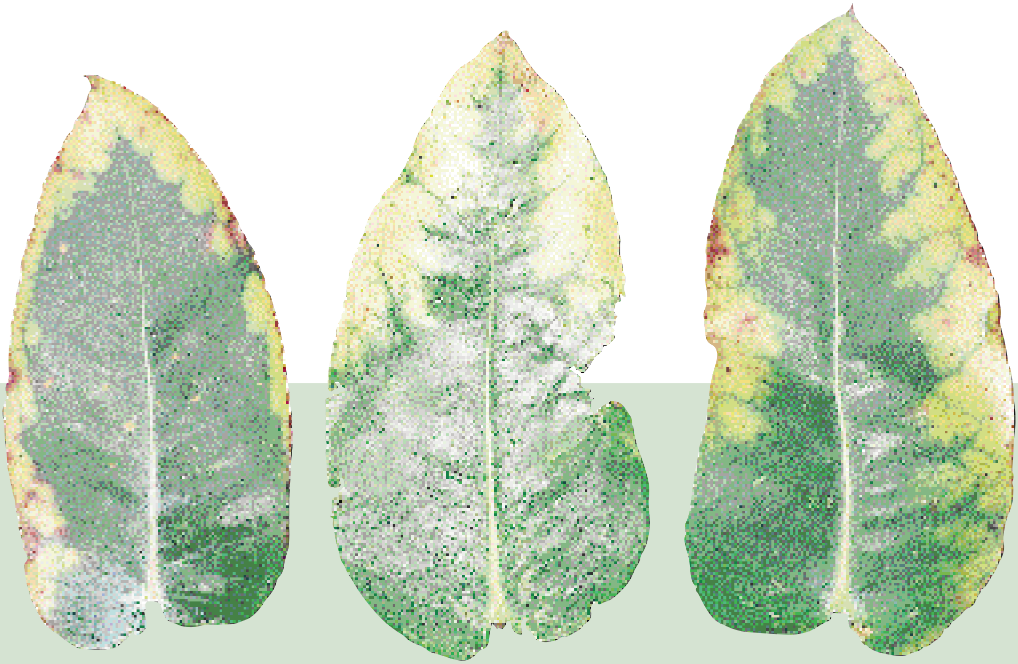
173. *E. urophylla*: leaf burn in a young plantation affected by fluoride emission from brick kilns in an adjacent valley. Guangdong Province, P.R. China.

174. *E. urophylla*: leaf necrosis caused by fluoride. Guangdong Province, P.R. China.

175. *E. excerta*: terminal leaf necrosis caused by fluoride. Guangdong Province, P.R. China.

176. *E. tereticornis*: necrosis in leaf exposed to high doses of SO_2 in an experimental chamber. (Photo: F. Murray).

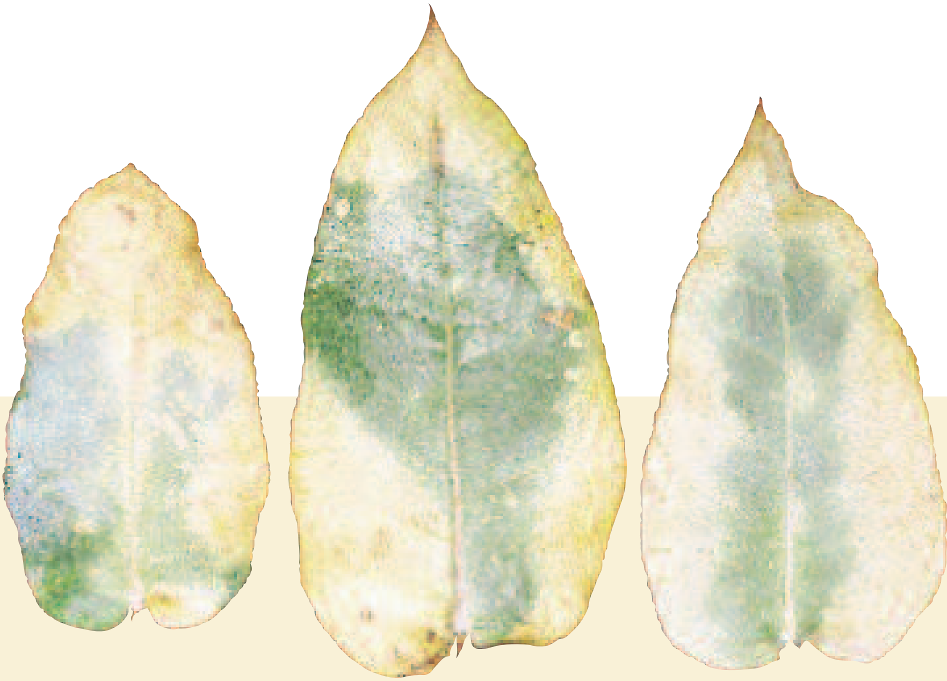
Chemical damage



177. *E. globulus* ssp. *globulus*: marginal chlorosis caused by spray drift during application of herbicide. Western Australia.

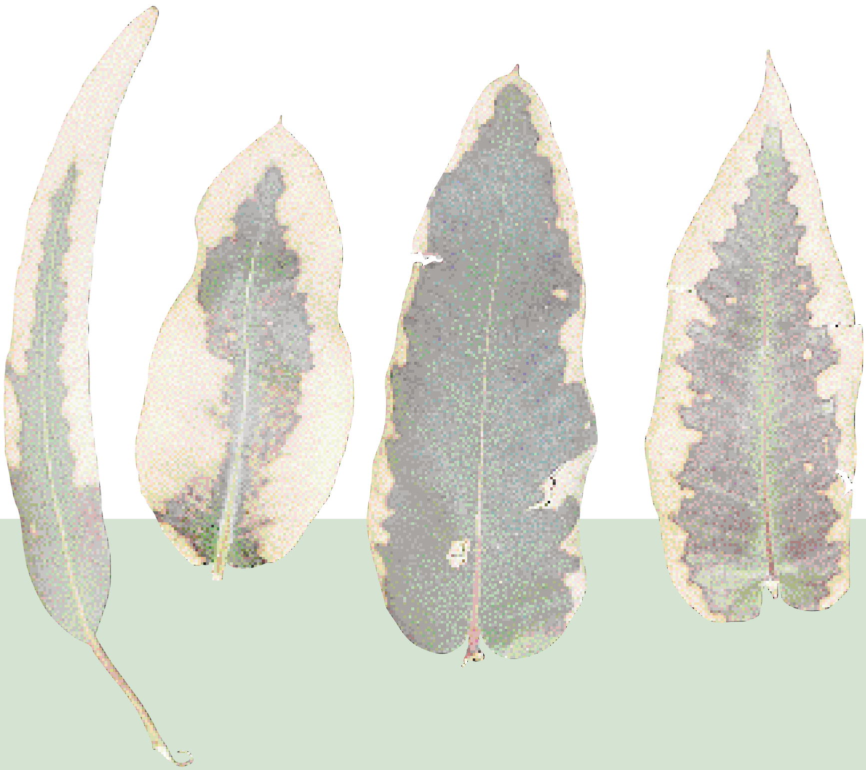
Environmental stresses

178. *E. globulus* ssp. *globulus*: one-year-old tree with bleached leaves after six months drought. Western Australia.



179. *E. globulus* ssp. *globulus*: drought-affected leaves with dead patches of bleached tissue. Western Australia.

180. *E. globulus* ssp. *globulus*: one-year-old trees with shoot death due to waterlogging. Western Australia. (Photo: G. Hardy).



181. *E. globulus* ssp. *maidenii*: frost damage on mature leaves. Yunnan Province, P.R. China.

Genetic factors

182. *E. globulus* ssp. *globulus*: one-year-old plantation with normal tree (right) and a genetic dwarf (left). Western Australia.

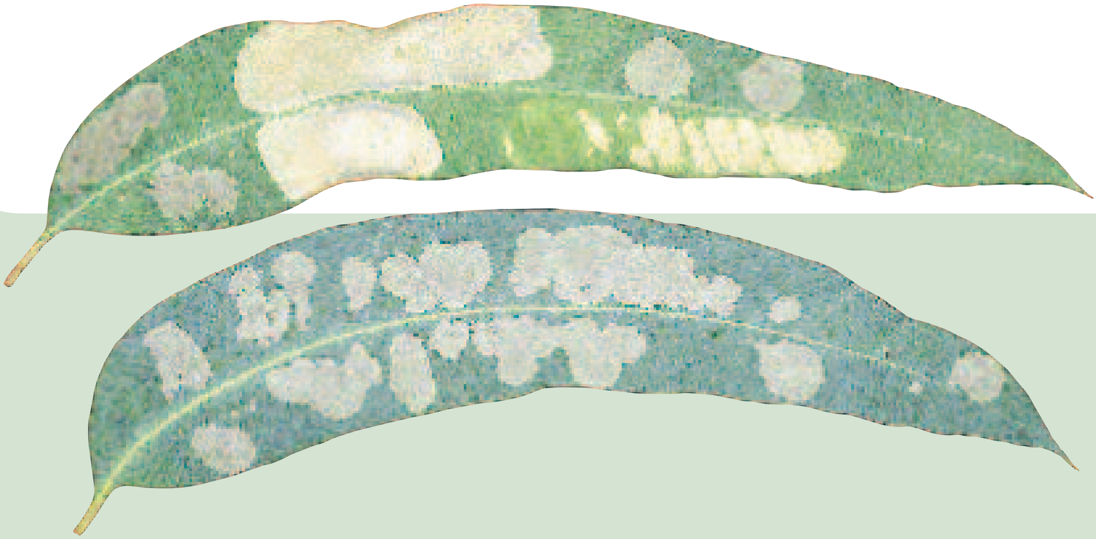
183. *E. globulus* ssp. *globulus*: natural variegation in juvenile leaves. Western

184. *E. grandis*: chlorotic leaves resulting from the lack of chlorophyll synthesis. Indonesia.

185. *E. grandis*: natural variegation. Indonesia.

Herbivores

186. *E. globulus* ssp. *globulus*: two-year-old plantation with tree in the foreground heavily attacked by leaf-blister sawfly (*Phylacteophaga frogatti*). Western Australia. (Photo: G. Hardy)

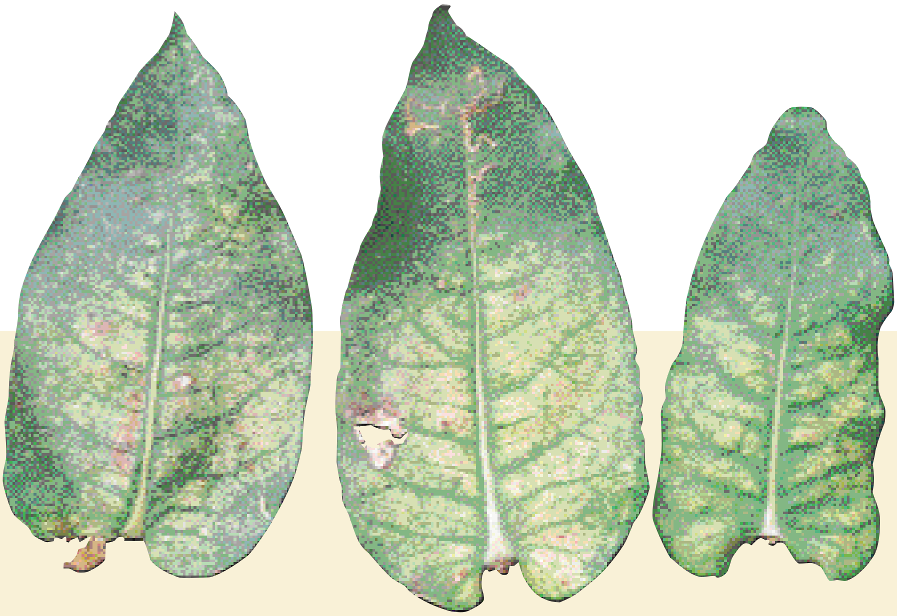


187. *E. camaldulensis*: leaf damage caused by leaf-blister sawfly. Western

188. *E. globulus* ssp. *globulus*: adult foliage consumed by autumn gum-moth (*Mnesampela privata*). Western Australia. (Photo: G. Hardy).

189. *E. globulus* ssp. *globulus*: the blue gum psyllid (*Ctenarytaina eucalypti*) has caused the malformation of young juvenile leaves. Western Australia.

190. *E. globulus* ssp. *globulus*: leaf malformation and multiple shooting in a shoot from a one-year-old tree which has been attacked by psyllids. Western Australia.



191. *E. globulus* ssp. *globulus*: mature juvenile leaves with chlorosis caused by sap-sucking insects and mild K deficiency. Western Australia.

192. *E. globulus* ssp. *globulus*: parrot damage to the bark can result in the death of the leader. Western Australia.

Pathogens

193. *E. globulus*: leaf-spot on juvenile foliage caused by *Mycosphaerella cryptica*. Western Australia.

194. *E. globulus*: leaf-spot on adult foliage caused by *Mycosphaerella cryptica*. Western Australia.

195. *E. globulus*: juvenile leaf with symptoms of Fe deficiency (interveinal chlorosis), fungal disease (leaf spots caused by *Mycosphaerella* sp.) and insect herbivory. South Australia.

196. *E. globulus*: Cu-deficient one-year-old tree with branches heavily infected by canker fungi. Western Australia.

197. *E. globulus*: bark-splitting caused by *Endothia gyrosa*, a canker-forming fungal pathogen. Western Australia. (Photo: G. Hardy).

198. *E. grandis*: defoliation caused by *Cylindrocladium* sp. Near Ingham, Queensland. (Photo: S. Rance).

199. *E. grandis*: disturbed growth in leaves and stems of six-month-old tree due to infection by the rust fungus *Puccinia psidii*. Brazil.

200. *E. urophylla*: defoliation of lower and middle canopy by an unidentified fungal pathogen. Guangdong Province, P.R. China. (Photo: D. Xu).

7. Confirmation of diagnosis by plant analysis

The diagnosis of a nutrient disorder from visual symptoms (Chapter 5, Will 1961, Malavolta et al. 1962, Karschon 1963, Kaul et al. 1968, Marcos de Lanuza and Marzo Munzo-Cobo 1968, Marcos de Lanuza and Marzo Munzo-Cobo 1969, Truman and Turner 1972, De Rocha Filho et al. 1978, Stewart et al. 1981, Cheng and Horng 1992, Dell and Robinson 1993, Dell and Malajczuk 1994, Grundon et al. 1997), can be verified by chemical analysis of affected plant tissue. Plant analysis is often the preferred method of diagnosing nutrient deficiencies, as concentrations of nutrients in plant tissues are determined by the combined effects of all the processes affecting the supply of nutrients in soil and uptake by roots. Although plant analysis has been used most widely in agriculture and in horticulture, there has also been considerable development of plant analysis methods for diagnosing nutrient deficiencies in forest trees (Ballard 1980, Schönau 1981, Schönau and Herbert 1983, Lambert 1984, Mead 1984).

A number of approaches have been used for the diagnosis of nutrient disorders from plant analysis (Dell 1996). These include: (a) comparison of nutrient concentrations in healthy and unhealthy plants; (b) the estimation of plant nutrient status from a knowledge of relationships between concentrations of nutrients in plant tissues and plant growth; (c) examination of nutrient balances within plants through calculation of nutrient ratios; and (d) development of biochemical techniques. These approaches are outlined below.

Comparison of healthy and unhealthy trees

Comparing nutrient concentrations in healthy and unhealthy plant tissues is a commonly used method for confirming the cause of visual symptoms of a nutrient disorder. It will be most useful where uncertainty arises because of (i) changes in symptoms with the severity of deficiencies and with plant age, (ii) the possibility of incorrect interpretation where symptoms are caused by other environmental factors (e.g. disease, pollution), (iii) the similarity of some symptoms associated with different nutrients, and (iv) the compounding of effects where more than one nutrient imbalance is involved. Where visual symptoms of nutrient disorders are observed in eucalypt plantations (Plates 201 and 202), samples of healthy and unhealthy leaves of uniform age should be collected from within the same area.

Information on sampling is given in Chapter 8. Typical nutrient concentration ranges observed in nutrient-deficient trees and trees with adequate levels of nutrients are given in Table 4.

201. *E. globulus*: pockets of yellow, Fe-deficient two-year-old trees associated with calcareous soil. Australia.

Table 4: Nutrient concentration ranges observed in the youngest fully expanded leaves of one-to two-year-old eucalypts in plantations. The *E. globulus* data are from plantations in Australia and P.R. China, and the data for the other species are from P.R. China, Indonesia and the Philippines. A question mark indicates that further work is required to confirm the data.

	E. globulus		*E. grandis*		*E. urophylla*		*E. grandis x E. urophylla*	
	Deficient	**Adequate**	**Deficient**	**Adequate**	**Deficient**	**Adequate**	**Deficient**	**Adequate**
	mg g^{-1} Dry Wt							
N	10-17	19-27	5-15	18-34	<11	15-30	8-13	18-29
P	0.7-1.0	1.3-2.7	0.2-0.9	1-3	0.5-0.9	1.2-3.1	0.8-1.0	1.2-2.6
K	3-7	8-15	3-5	6-18	2-5	6-16	2-6	9-15
Ca	?<1	3-17	<1	3-8	?<1	3-15		2.1-7.5
Mg	?<0.8	1-7	0.3-0.7	1-3	?<0.8	1.7-6.4	0.2-0.4	1.1-3.6
S	<1.2	1.3-2.2	<1	1.5-3	?<1	1-3		1.2-2.9
	mg kg^{-1} Dry Wt							
Fe	8-10*	25-700	10-14	25-130	?<20	25-100		40-100
Zn	8-11	15-50	5-9	15-50	9-12	16-47		13-29
Mn	2-19	40-2000	?<15	60-2300	?<15	130-4000		130-2300
Cu	0.5-2	3-24	0.4-1.5	2-11	0.8-1.5	3-19		3.5-13.4
B	4-10	14-38	5-8	15-27	4-12	16-69	8-12	13-30

* <20 on calcareous soils

A disadvantage of restricting plant analysis to investigation of nutrient concentrations in unhealthy tissue is that symptoms may only appear when nutrient supply to roots has markedly reduced growth (Fig. 5) or caused stem malformation (Mead 1984). In a similar approach not dependent on the expression of deficiency symptoms, multiple regression analysis to relate concentrations of nutrients in tissues to stand vigour can also be an effective tool in the intensive management of forest stands (Bevege 1978).

Critical nutrient concentrations

Plant analysis methods used in diagnosing nutrient disorders and predicting nutrient requirements of plants are based on the relationships which occur between plant growth and concentrations of nutrients in plant tissues (Fig. 5). An example of a response for B is illustrated in Fig. 6. These relationships, established by experimentation with a number of rates of application of a single nutrient, are used to determine the 'critical nutrient concentration', the concentration in the plant tissue above which there is no significant increase in growth (Ulrich and Hills 1967). The critical concentration is often set for 90% of maximum yield (Table 5).

Alternatively, relationships between nutrient function and concentrations of nutrients in plant tissues can be used (Table 5). Loneragan (1968) introduced the concept of a 'functional nutrient requirement' defined as 'the minimal concentration of nutrient within the organism which can sustain its metabolic function at a rate which does not limit growth'. Nutrient functions that can be measured include the activity of nutrient-dependent enzymes (Dell and Wilson 1989, Gherardi et al. 1999) and biochemical processes such as photosynthesis in leaves or the lignin content of wood (Dell 1996). The relationship for a specific nutrient can be markedly affected by limitations of other nutrients so that critical concentrations need to be determined under conditions which ensure an adequate supply of all other nutrients. However, many other factors can also affect critical concentrations, the most common being declines in the nutrient concentrations of tissues, and in critical concentrations, with increasing plant age. In developing plant analysis methods for diagnostic and predictive purposes, a common objective is therefore to

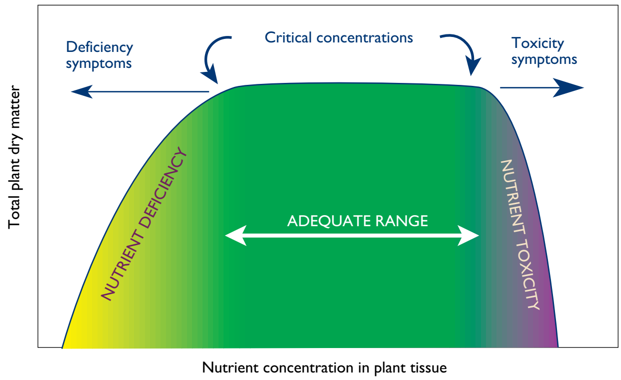


Figure 5: Relationship between nutrient concentration in plant tissue and plant growth.

Table 5: Critical nutrient concentrations for the diagnosis of nutrient deficiencies in eucalypt seedlings determined from nutrient rate experiments.

Species	N(mg/g)	P(mg/g)	K(mg/g)	Zn(mg/kg)	Cu(mg/kg)	B(mg/kg)
E. calophylla						17.9 [a, e, f]
E. globulus	24.9 [a, e, o]				2.4 [c, a, e, g]	11.7 [a, e,h]
E. grandis					2.1 [c,a,e,g]	
E. maculata				9-10 [a, e, j]	1.5 [b, e, i]	
E. marginata				10-12 [b, e, k]		
E. pellita			7 [a, e, l]			
E. urophylla		1.7 [a, d, m]		21 [a, e, n]	2.6 [c, a, e, g]	

a = youngest fully expanded leaf; b = shoot apex; c = functional Cu requirement for catechol oxidise activity; d = field (one-year-old trees); e = glasshouse (seedlings); f = Asad et al. (Unpub. Data); g = Gherardi et al. 1999; h = Sakya et al. (Unpub. Data); i = Dell 1994; j = Dell and Wilson 1989; k = Wallace et al. 1986; l = Dell (Unpub. Data); m = Dell and Xu (Unpub. Data); n = Dell and Xu 1995; o = Shedley et al. 1995

analyse plant tissues of constant developmental age on each sampling occasion or to quantify the variation in critical nutrient concentration with increasing plant age. Furthermore, as environmental factors can markedly affect maximum growth rates and critical nutrient concentrations, relationships between tissue nutrient concentrations and growth need to be defined in the field as well as under glasshouse conditions (Bates 1971). However, we have found that functional nutrient concentrations for a leaf at a defined stage of development can be quite similar for leaves formed in different environments. Also, critical P concentrations for the diagnosis of deficiency in *E. urophylla* were the same for one-year-old trees in China and seedlings in a glasshouse in Australia (Xu and Dell, unpublished data).

The extent to which nutrients can be retranslocated within the plant determines the selection of tissue which is most suitable for predicting the current nutrient status of the plant. With phloem-immobile nutrients (e.g. Ca, Fe) and variably mobile nutrients (e.g. Zn, Cu, S), young tissues provide the best indication of current nutrient status, whereas for mobile nutrients (e.g. N, P, K) which can be rapidly retranslocated from old plant tissues to young growing shoots, the analysis of mature tissues gives the best guide to nutrient status (Hill 1980, Marschner 1995). In eucalypts and other evergreen trees, there are generally large gradients in foliar concentrations

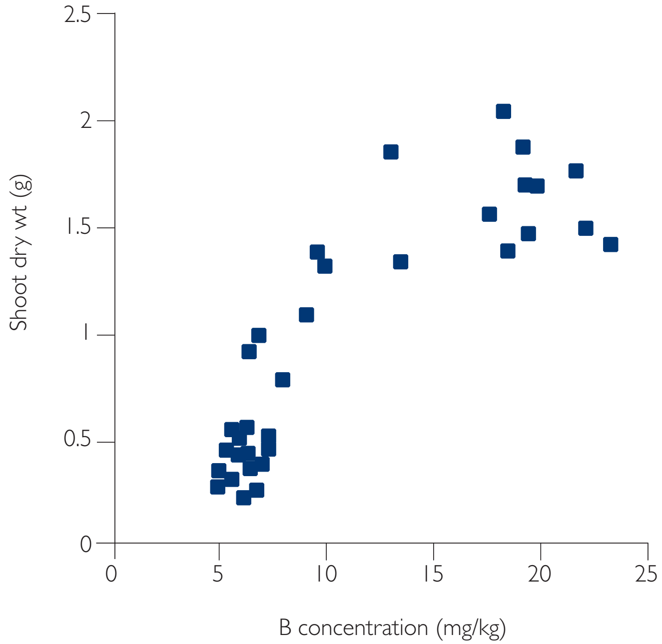


Figure 6: Relationship between shoot dry weight and the B concentration in the youngest fully expanded leaves of *E. globulus* seedlings. (Data: Amalia Sakya).

of both mobile and immobile nutrients throughout the crown as a result of variation in the age of foliage (Bara Temes 1970, Lamb 1976, Mead 1984, Leuning et al. 1991). Strong gradients also occur with leaf position on a branch (Fig. 7), so great care must be taken in selecting the appropriate position for sampling (Chapter 8).

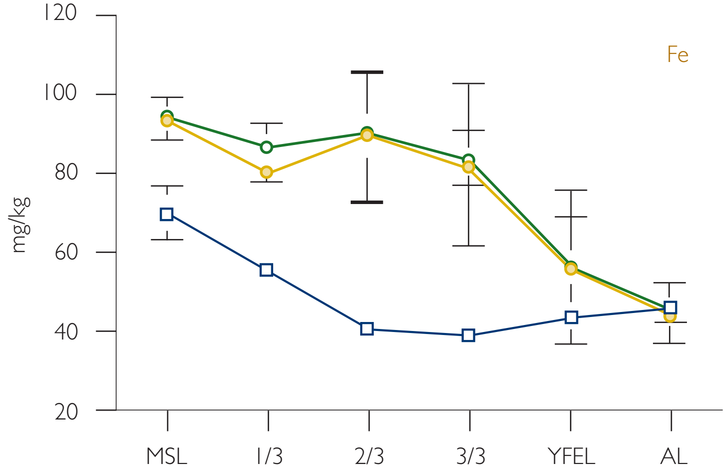
The variation within eucalypt crowns, and seasonal changes in foliar nutrient concentrations (Knight 1988) associated with differences in the seasonal patterns of nutrient uptake and growth (Fife and Nambiar 1982), means that sampling procedures for plant analyses need to be rigorously standardised. *For the practical purpose of using one organ for the analysis of all essential nutrients, we recommend sampling the first fully expanded leaves of an actively growing branch in the upper third of the canopy (See Plates 203 and 204)*. This position is based on data from many fertiliser response trials and field sampling.

For monitoring purposes (Chapter 8), trees should be sampled towards the end of a vigorous period of growth. For *E. globulus* in Western Australia, late summer or early autumn is recommended because this is a period of stability that best reflects the nutrient status of the tree and the supply characteristics of the soil. Examples of data that are used to define this

202. Most commonly, micronutrient deficiencies are expressed in pockets within a plantation. Here, B-deficient, prostrate *E. globulus* trees occur adjacent to trees with good form. Yunnan Province, P.R. China.

period are shown in Fig. 8. In south China, October-November is the recommended sampling time. Variations to the recommended sampling time may be used on an irregular basis if there is a requirement for management. For example, in Western Australia Cu levels in leaves produced in December give an early warning of potential tip die-back from Cu deficiency in late summer. Early prognosis allows corrective foliar Cu to be applied in time to prevent the onset of the problem. Note, however, that the foliar concentration ranges given in Table 4 are for leaves sampled in the season recommended above.

Plant analysis methods for predicting the nutrient status of forest species



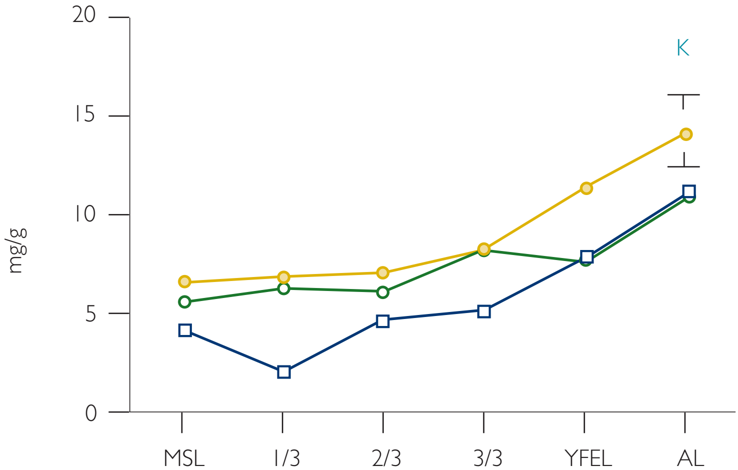


Figure 7: Nutrient concentrations can vary with leaf age along a branch. The data are for *E. globulus* and show contrasting patterns for Fe and K in three one-year-old trees. MSL are leaves where the branch joins the trunk, AL are leaves at the tip of the branch, YFEL are the youngest fully expanded leaves. (Data: Catherine Chamberlain).

have in most cases favoured the use of foliar tissues. Leaves are generally chosen on the basis that they are a site of high chemical activity, they are convenient to sample, and they are often the tissue where symptoms of nutrient disorders are first expressed. In some situations, tissues other than leaves (e.g. twigs, bark, roots, xylem sap) have been shown to be useful in the diagnosis of nutrient deficiencies. Whilst petioles are favoured for analysis for some deciduous plants, they are too small in eucalypts to be considered here. Studies of eucalypt growth in relation to soil P supply have suggested that P concentrations in young stems of seedlings (Dell et al. 1987, O'Connell and Grove 1987) or in twigs of older trees (Grove 1990) may be a more sensitive measure of plant P status than P concentrations in leaves. In older eucalypt stands where access to the crown becomes difficult, analysis of inner bark tissue may be suitable in predicting tree requirements for some nutrients. However, this approach is not recommended until extensive studies have been undertaken to establish that the bark is sensitive to changes in nutrient supply of most essential nutrients. Suggestions that roots be sampled as alternatives to above-ground parts are impractical. In the future, sap analysis may prove to be a useful predictor of requirement for N fertiliser.

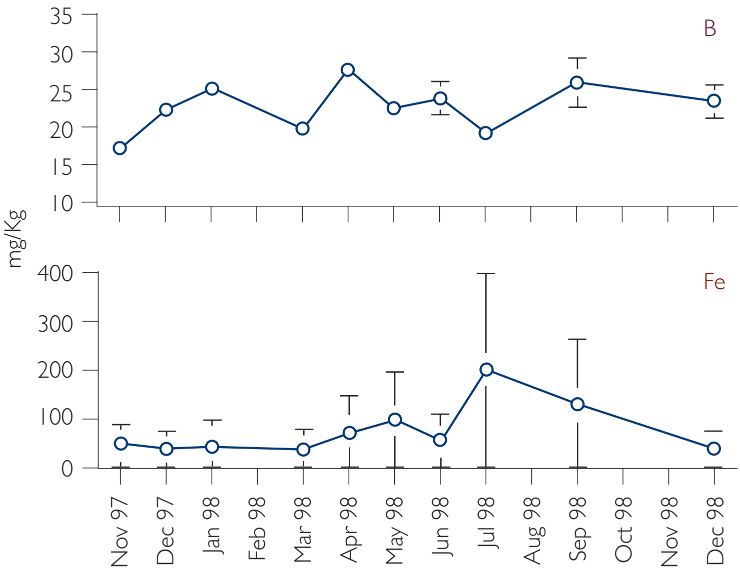


Figure 8: Effect of season on the concentration (mg/kg dry wt) of B and Mn in the youngest fully expanded leaves of *E. globulus*. Trees were 16 months of age at the first sampling date. Foliar Mn concentrations increased markedly during winter in some trees. February to April is the recommended sampling period in this region (Albany, Western Australia).

Nutrient ratios

Another approach often used in the diagnosis of nutrient deficiencies is the examination of nutrient 'balance' within the plant. In its simplest form, this method involves measurement of the ratio of two nutrients within a plant tissue and comparison with a predetermined range where plant growth will not be limited. A more comprehensive development of this approach is the Beaufils' Diagnostic and Recommendation Integrated System (DRIS) which is based on the calculation of ratios of several nutrients and their use in combination to indicate nutrient deficiencies (Beaufils 1973, Walworth and Sumner 1987). DRIS is a predictive rather than a diagnostic technique and has not been shown to be more useful than the traditional approach discussed above (Smith and Loneragan 1997). For example, the DRIS method was found to be only partially successful in predicting the responses of *Eucalyptus saligna* saplings to addition of N, P and K fertilisers (Ward et al. 1985, Yost et al. 1987). Disadvantages of this approach are the greater analytical requirements and, with the exception of N and S, the lack of a sound physiological basis for interpreting nutrient ratios.

Biochemical tests

There has been a rapid development of biochemical techniques for the diagnosis of nutrient deficiencies in agricultural and horticultural plants over the past two decades but there has been relatively little use of the techniques in forestry. Dell (1996) suggested that the biochemical assays which show the most promise for eucalypts are those developed for P and Cu. Phosphatase activity in tissues of *E. diversicolor* seedlings was shown to be a more useful indicator of P deficiency than tissue P concentration in one glasshouse study (O'Connell and Grove 1987). Phosphatase activity was elevated in P-deficient plants. Dell (1994) used a lignin stain to distinguish between Cu-deficient and Cu-adequate *E. maculata.* The activity of phenolase is reduced in Cu-deficient eucalypts (Dell 1996) and the activity of this enzyme was used to define functional nutrient requirement referred to earlier (Gherardi et al. 1999). Dell and Wilson (1989) found that the activity of carbonic anhydrase was severely depressed in Zn-deficient *E. maculata* but concluded that foliar Zn analysis was superior for diagnosis. Biochemical assays may be better than nutrient analysis for predicting the nutrient status of a plant when significant amounts of nutrients are held in physiologically inactive forms (Leece 1976) but lack of specificity and often greater complexity in their measurement are seen as disadvantages. Presently, there is a need to develop a biochemical test for Fe as Fe-deficient *E. globulus* on calcareous soils has higher leaf total Fe concentration than Fe-adequate trees on acid soils.

8. Collecting and handling leaf samples

Introduction

In the previous chapter, some of the principles regarding the use of foliar nutrient concentrations for diagnosing nutrient disorders were explored. In this chapter, we give requirements and procedures for sampling trees and handling samples prior to their receival in analytical laboratories. *It is important that recommended procedures are followed otherwise the analytical data may be worthless.*

There are two main reasons for sampling trees, and the intensity and pattern of sampling will vary accordingly.

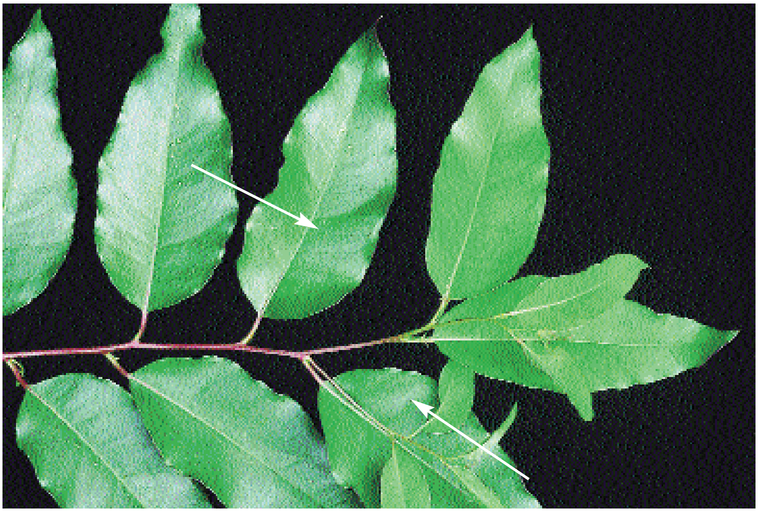
203. Leaves from the upper third of the canopy should be taken when sampling trees for monitoring purposes. The age-class of leaf to collect is shown in Plate 204. *E. globulus*, Western Australia.

Sampling trees for the diagnosis of nutrient disorders

Samples are collected using a sampling strategy based on the occurrence of trees with symptoms compared to healthy trees (see Plates 201 and 202).

1. Take leaves from matched healthy and affected trees.
2. Choose trees at random within the affected and unaffected areas.
3. Where a range of symptoms exist, collect separate samples of each symptom group.
4. Do not combine samples from trees with different symptoms.
5. Consider sampling the following leaf positions (see Plates 204 and 205): the shoot apex including stem and undeveloped leaves; the youngest fully expanded leaves and the mature leaves up to 6 months of age.
6. Record the following sample and site information:

    Location of plantation
    Date of sampling
    Sample No.
    Number of samples
    Compartment/Block
    Tree species (include clone, coppice, etc.)
    Stocking
    Tree age
    Leaf age and position
    Tree condition including symptoms in sampled leaves (see Chapter 5)
    Damage from pests, disease, drought, water-logging, salt, chemical sprays, etc. (see Chapter 6)
    Reason for submission
    Surface soil type and depth
    Sub-soil type and depth
    Drainage
    Topography
    Previous land-use
    Fertiliser history
    Soil cultivation
    Weed control
    Annual rainfall



204. The youngest fully expanded leaves (arrowed) from a branch with an actively growing tip are the recommended organs for foliar analysis in eucalypts.

Recent weather events

Sampling for monitoring purposes

Samples are collected in a repeated and systematic way to assess plantation management.

Typical questions that can be addressed with monitoring are:

Is the nutrient content of the trees in balance?

Have the nutrient concentrations in trees responded favourably to silvicultural treatments such as thinning, weed control, addition of fertiliser, etc.?

Is there a need for the addition of fertiliser?

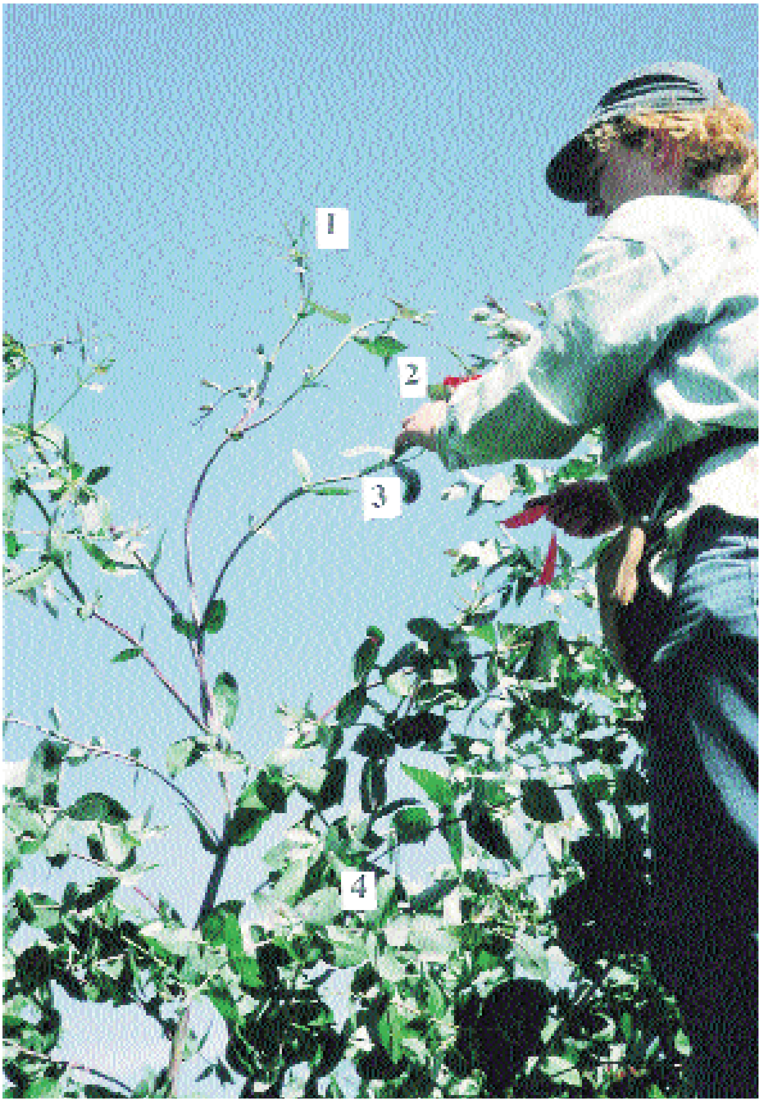
Will the rate of nutrient decline depress wood yield at harvest?

Proceed as follows:

1. Establish permanent sampling plots of 25-50 trees in compartments representative of the plantation. These can also serve as inventory plots. The sampling plots should not be placed on the edge of the plantation.
2. Select the appropriate sampling season (see page 158)
3. Sample 5-10 trees at random within the sample plot (see box on

sampling intensity, page 168)

4. Sample the upper third of the canopy (Plate 203).
5. For each tree, select two actively growing branches and remove two of the youngest fully expanded leaves (Plate 204).
6. Combine the leaves from the 5-10 trees into one sample.



205. Sampling an *E. globulus* tree to confirm Cu-deficiency. When the purpose of sampling is to diagnose an existing nutrient disorder, it may be necessary to sample the parts illustrated: 1 – shoot apex, 2 – expanding leaves, 3 – youngest fully expanded leaf, 4 – mature leaves. Western Australia.

7. Place the leaves into clean, labeled paper bags. Leaves may be wrapped in white, unscented tissue paper.

Handling leaves for both sampling methods

1. Avoid contaminating samples. Common sources of contamination include: soil, gum on envelopes, hand creams, sun-blocks, insect repellents, cigarettes, copper bracelets, metal containers, galvanised ladders (Plate 206) and packaging tapes.
2. Disposable latex gloves can be used but many of the above risks of contamination still remain.
3. Do not collect leaves that are covered in dust.
4. Do not wash leaves.
5. Avoid overheating fresh samples. Do not put samples in plastic bags. In hot climates samples can be placed in an ice chest for transport to base.
6. Dry samples in a ventilated oven at 65–70 °C to constant weight (usually a few days).

206. Sampling trees with galvanised metal ladders, as shown here, is not recommended as it is difficult to prevent contamination of samples with Zn and other heavy metals.

7. Dried samples in paper bags can be wrapped in plastic for transport to the laboratory for analysis.

Sampling intensity

When monitoring the nutrient status of trees in a plantation a critical question is: 'How many trees should be sampled to estimate the mean level of a particular nutrient in the plantation?' This is a straight forward task if one assumes that the distribution of the chosen nutrient follows a normal distribution and one has an estimate of the likely standard deviation of nutrient levels in the plantation. The steps are as follows:

- Choose a level of precision for the sampling (e.g., you wish the 95% confidence limit for your estimate of the mean to be ± 1 mg/g). The choice of precision is arbitrary and depends on how confident you need to be of your estimate of the mean.
- Estimate the standard deviation in the study population. This can come from a pilot study, or from prior sampling of a similar population such as a nearby plantation.
- Estimate the required sample size using:

$$n \approx \left(\frac{2(s)}{d}\right)^2$$

where n = the estimated sample size

s = the standard deviation

d = desired absolute error

Strictly speaking, the 2 in the above equation should be the t distribution value for sample size n. However, for simplicity this can be approximated as 2 for most cases unless n is very small.

As an example, consider the case of a plantation which you wish to monitor for levels of copper and manganese. Previous sampling of 100 trees has shown that the standard deviations for the concentration of these elements (mg/kg) in the plantation trees are 1.87 and 49.07 respectively.

What sample size is necessary to estimate mean copper concentration with a 95% confidence interval of ±0.5 mg/kg?

Applying the above equation:

$$n \approx \left[\frac{2(1.87)}{0.5}\right]^2$$

$= 56$

What sample size is necessary to estimate mean manganese concentration with a 95% confidence interval of ±20 mg/kg?

Applying the above equation:

$$n \approx \left[\frac{2(49.07)}{20}\right]^2$$

$= 24$

However, sometimes in forestry applications the question is: 'Given a sample size of x (often 5 or 10 trees), what is the 95% confidence limit for an estimation of the mean nutrient concentration of the trees in the plantation?' This question can be answered by a simple rearrangement of the formula to:

$$d = \frac{2s}{\sqrt{n}}$$

For clarity, this can be written as:

absolute error = 2(standard deviation)/sqrt(number of trees)

Sample size	Copper	Manganese
5	1.67	44.01
10	1.18	31.06

Consider the above example of the levels of copper and manganese in a population. The table below shows the 95% confidence limits in mg/kg of each nutrient for sample sizes of 5 and 10 trees respectively.

9. Fertiliser and correction of nutrient deficiencies

Introduction

The productivity of eucalypt plantations on infertile soils can be substantially improved by the application of fertiliser and appropriate site management (Xu et al. 2000). Yet, in many parts of the world, plantation eucalypts are being grown with below optimum levels of fertiliser (Plate 207). Contributing factors include: the high cost/availability of quality fertiliser; lack of data on soil constraints to tree growth; target yields set well below potential optimum tree yields; poor management of soil fertility; incidence of pests and diseases; and ignorance of fertiliser requirements for the sustainable yields of food and other crops within the region. Unlike other industrial tree and field crops, eucalypt plantations are often established in new geographical regions without extensive silvicultural trials. The response of eucalypts to the application of fertiliser is, however, a relatively simple technique for diagnosing nutrient deficiencies and for establishing fertiliser prescriptions. The success of fertiliser trials depends on many factors, the two most important being fertiliser quality and experimental design.

207. On infertile soils or soils with a nutrient imbalance, large responses to fertiliser application can occur. Changing from the traditional fertiliser practice (centre row) to a complete fertiliser (other rows) has dramatically increased growth of *E. globulus*. Yunnan Province, P.R. China.

Fertiliser quality

The use of single salt fertilisers is recommended for trials concerned with deficiency diagnosis. This is because compound fertilisers may vary considerably from their certified values in some areas. This can be overcome by having samples analysed by a reputable laboratory before experiments are established. The diagnosis of micronutrient deficiencies requires special care to protect plots from contamination. The main sources of contamination are:

Micronutrients present as minor contaminants of commercial fertilisers. For example, B levels are usually low in urea (<5 mg B/kg), moderate in single superphosphate (approx. 50 mg B/kg) and high in triple superphosphate (>100 mg B/kg).

Use of dirty equipment for sieving, weighing, mixing or spreading fertiliser.

Preparing fertiliser mixes too close to a field trial.

Inadvertent movement of fertiliser with soil on machines, boots, or hand tools, and

Surface runoff on slopes after heavy rain.

208. Sieving fertiliser to ensure that uniform amounts of nutrients are distributed to trees in research plots. P.R. China.

209. Preparing a micronutrient mix for a research trial in south China. It is crucial that small amounts of micronutrients are well mixed and placed into a carrier.

Often it is necessary to sieve fertilisers (Plate 208) that have been stored in bags, to remove lumps that can then be pulverised, thus ensuring a uniform blend. Small batches of micronutrient mixes (Plate 209) may need to be made up for field trials because the incidence of micronutrient deficiencies varies between sites. Micronutrient salts should be dry, without lumps, and components should be mixed as close to the time of use as possible, as chemical reactions may occur. Magnesium or iron sulfate, or another fertiliser, can be used as a carrier for micronutrient treatments.

Experimental design

The value of fertiliser trials is diminished unless careful planning is undertaken prior to establishment. The three most common problems encountered are:

The failure to include micronutrients as a basal dressing in macronutrient trials. This creates uncertainty about the maximum productivity measured as productivity can be modulated by induced secondary micronutrient deficiency.

The use of small plots and lack of buffer rows between treatments. Single row plots can provide useful information on short-term responses to fertiliser but larger plots (40-50 trees) are ideal for following responses through a rotation.

Treatment selection that makes it difficult to formulate fertiliser prescriptions. The following is an example of a poor design that should be avoided.

7 treatments: -N-P-K, +N-P-K, -N+P-K, -N-P+K, +N+P-K, -N+P+K, +N+P+K. Each element has only one rate.

210. Fertiliser is being applied by volume to a research trial in Yunnan Province, P.R. China.

In addition to selecting treatments that are appropriate to the objectives of the research trial, decisions need to be made regarding the size and arrangement of plots, the number of replicates, and the allocation of treatments to plots. A statistician can provide effective advice at the planning stage that will help to maximise the effectiveness of the trial. The most commonly used design for fertiliser trials is the randomised block design. More complex designs, such as the incomplete block design, are useful in accounting for variability over sites (Williams and Matheson 1994).

Research trials

Identifying limiting nutrients

The omission fertiliser trial is recommended to identify nutrient limitations for tree growth and is particularly useful for the clarification of multi-element deficiencies in the field. In the omission trial, the effects of omitting single elements or groups of elements on yield are compared with a complete fertiliser treatment. Three examples of omission trial designs are given below. 'All' refers to a complete fertiliser containing all essential macro- and micronutrients at rates that are optimal for tree growth.

Trial A	Trial B	Trial C
All	All	All
All-N	All-N,P,K	All-B
All-P	All-Ca,S,Mg	Nil
All-etc	All-micronutrients	
Nil	Nil	

In trial A, the aim is to determine which macro- and/or micronutrients are likely to be limiting growth. In B, the aim is to identify whether a particular group of nutrients is limiting growth. In C, the aim is to establish whether boron is limiting tree growth (Plate 211). Figure 9 shows the results of an omission trial that was undertaken to assist setting fertiliser recommend-ations for plantation eucalypts on problem soils in the Philippines. Nitrogen was not included as a factor in this design because N was known to be limiting at this site. The site has multiple nutrients limiting growth, the main ones being the macronutrients N, P and K, and the micronutrients B and Zn.

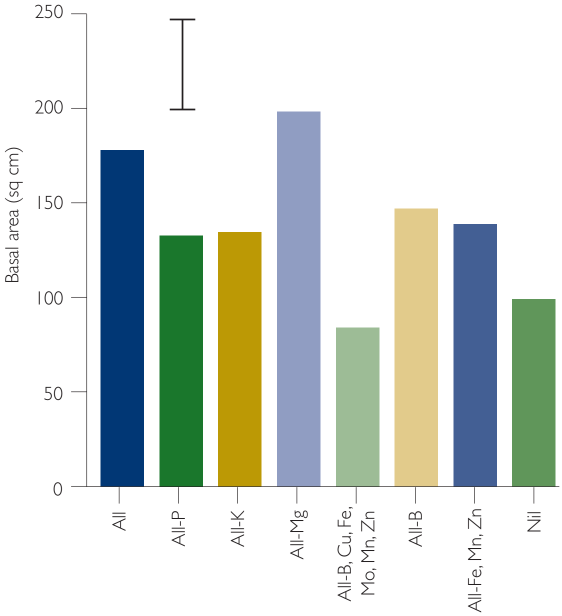


Figure 9: Growth response of *E. urophylla* to treatments in a fertiliser deletion trial five years after outplanting. Bukidnon, Philippines. LSD (P=0.05)

211. A demonstration trial shows the extent to which B is limiting productivity of *E. globulus*. Trees in the foreground received basal fertiliser without B. Trees in the background received basal fertiliser and 0.6 g B/tree. Yunnan Province, P.R.

Confirming nutrient deficiencies

In general, plant analysis (Chapter 7) is more reliable and precise in determining the capacity of eucalypts to obtain nutrients from soil than soil analysis. In a study on *Eucalyptus* ABL 12, a hybrid used extensively on the Leizhou Peninsula, Guangdong Province and in Hainan, MAI was closely related to nutrient concentrations in leaves (Cha et al. 1999a, b). For example, for K, the MAI (m^3/mu/y) = 0.119 + 0.0949 × [K in mg/g] (1ha = 15mu). The foliar K concentration was closely related (r=0.941) to total soil K but poorly related to exchangeable K concentration in the top soil. Foliar P concentration, a good predictor of growth, was not related to available P (r=-0.17) nor total P concentration (r=0.387) in the top soil. Although soil analysis can sometimes be used to confirm deficiencies of P, K and B, very few soil test standards have been set for eucalypt plantations and the soil profiles sampled for soil analysis rarely correspond to the root distribution profiles. Therefore, it is often better to confirm nutrient deficiencies by measuring the response in tree growth and foliar nutrient concentrations to the addition of fertiliser.

212. A phosphorus rates trial with *E. grandis* × *E. urophylla* consisting of six rates of P, ranging from 0 to 312 kg P/ha, and four blocks, one year after establishment in Guangdong Province, P.R. China. Each treatment plot contains 40 trees. The plots with nil or low P have small trees. The application of 50-100 kg P/ha at planting gave a satisfactory yield at this site provided N, K and B were also applied.

Determining fertiliser rate

There is no substitute for undertaking fertiliser rates trials in the field that quantify tree nutrient demand throughout the rotation period (Plate 212, Figure 10). If an appropriate distribution of rates is used from deficiency to adequacy, plant and soil tests can be calibrated and fertiliser models developed. Unless calibrated, foliar and soil analysis can only identify a need for fertiliser but not the rate necessary to sustain yield. The amount of fertiliser required will vary with soil type, depending on the nutrient storage capacity, the organic carbon content, the buffering capacity, the number and type of reactive surfaces, soil depth, soil drainage, and other factors. Also, the placement of fertiliser, whether banded, spot applied, placed in the planting hole, or broadcast will affect the availability of fertilisers that react strongly with soil constituents. Issues concerning fertiliser type, composition, application, placement, etc. are not covered here as they are dealt with elsewhere (Anon 1998, Tisdale et al. 1998, Glendinning 2000).

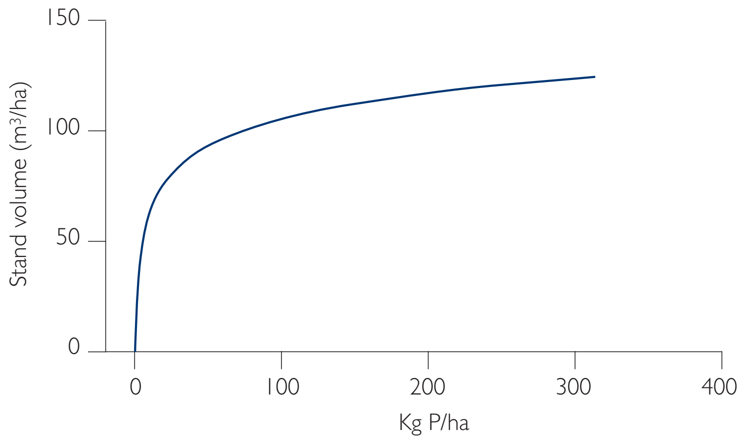


Figure 10: Results of a phosphorus rates trial (See Plate 212) showing the response of six-year-old *Eucalyptus grandis* × *E. urophylla* to superphosphate applied at planting as a band 0.6 m wide along each row. Guangdong Province, P.R. China.

213. In many parts of Asia, B limits the productivity of eucalypts in plantations. If deficiency is diagnosed early, tree form and wood yield can recover quickly. The four rows of tall *E. urophylla* have responded to the application of soil-applied B (10 g B/tree) when trees were in their second year. Trees in the foreground and on the left are of the same age (5 years). Here, it is more economical to apply
B in the form of crushed ulexite than as agricultural borates. Bukidnon, Philippines.

Correction of nutrient deficiencies

Banding or broadcasting solid inorganic fertiliser is usually the preferred method for correcting nutrient deficiencies. The timing of fertiliser application should optimise both nutrient availability and uptake by the tree. Whilst rates will vary considerably depending on soil type, climate, tree age and tree species, the following is a guide to the range that may be appropriate for 1–3-year-old trees.

Element	Rate (kg/ha)	Compound
K	25-75	Kieserite, Potassium chloride, Potassium sulfate
N	50-150	Ammonium nitrate, Ammonium sulfate, Calcium nitrate, Urea
P	20-200	Superphosphate, Triple phosphate
B	0.5-1.5	Colemanite, Borate, Borax, Boric acid, Ulexite
Cu	1-4	Copper chloride, Copper sulfate, Cupric oxide
Mn	1-10	Manganous chloride, Manganous nitrate, Manganous sulfate, Manganous oxide
Zn	0.5-2	Zinc oxide, Zinc sulfate

Dramatic growth responses can occur particularly where a micronutrient deficiency has been debilitating for growth (Plate 213). However, where deficiencies of elements have resulted in severe crown dieback and loss of form, it is very difficult to restart growth. Uncertainty remains whether it is possible to kick-start rapid growth in trees, aged from 3 to 5 years, where the canopy area has declined markedly due to run down in soil nutrient capital (Plate 1).

Where foliar analysis detects the onset of micronutrient deficiency during the dry season, it can be advantageous to immediately apply foliar corrective treatments by fixed-wing aircraft or helicopters. Usually, foliar treatments with micronutrients can only deliver enough nutrient into the leaf to sustain growth for one season. They should be followed up by application of solid fertilisers to the soil after rain occurs. Rates of foliar fertiliser to apply are still being worked out (Plate 214) but starting dosages are available in the literature for other tree crops. On soils with a high peat content, very high rates of Cu (>10 kg/ha) have not alleviated Cu deficiency. On these sites it may be necessary to apply foliar Cu over 2-3 years in order to achieve satisfactory tree growth. This is because of the binding of Cu to the organic matter and the poor access of feeder roots to surface-applied Cu during the dry season.

214. Foliar application of Cu is an option for correcting Cu deficiency, especially on peaty soils. In this research trial on *E. globulus,* trees are being sprayed by hand to determine appropriate corrective treatments. Western Australia.

Concluding remarks

There is a need to reduce the gap between the deficient and adequate concentration ranges (Table 4) that analytical laboratories are using at the moment. These data can only be derived from well designed fertiliser rates trials. Ideally, these trials should be sufficiently visionary that they can help to set plant and soil tests, set fertiliser prescriptions, determine fertiliser needs across rotations, formulate growth models, help to establish sustainability indices for plantations, determine residual fertiliser values for soils, and quantify above and below-ground carbon sequestration. They can also be used for integrative studies on insect pests and various disease organisms. A further challenge, particularly on infertile soils, is meeting the anticipated increase in nutrient demand as wood volumes increase with the availability of improved tree genetics.

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