OTWAY WATER BOOK 64

LEAF ANALYSIS
and
Acid Sulfate Soil in the Big Swamp.

Google in <u>2021</u>. The Big Swamp showing a healthy coverage of vegetation.

Big Swamp 2011 (see page 11).

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25 August 2021

Malcolm Gardiner
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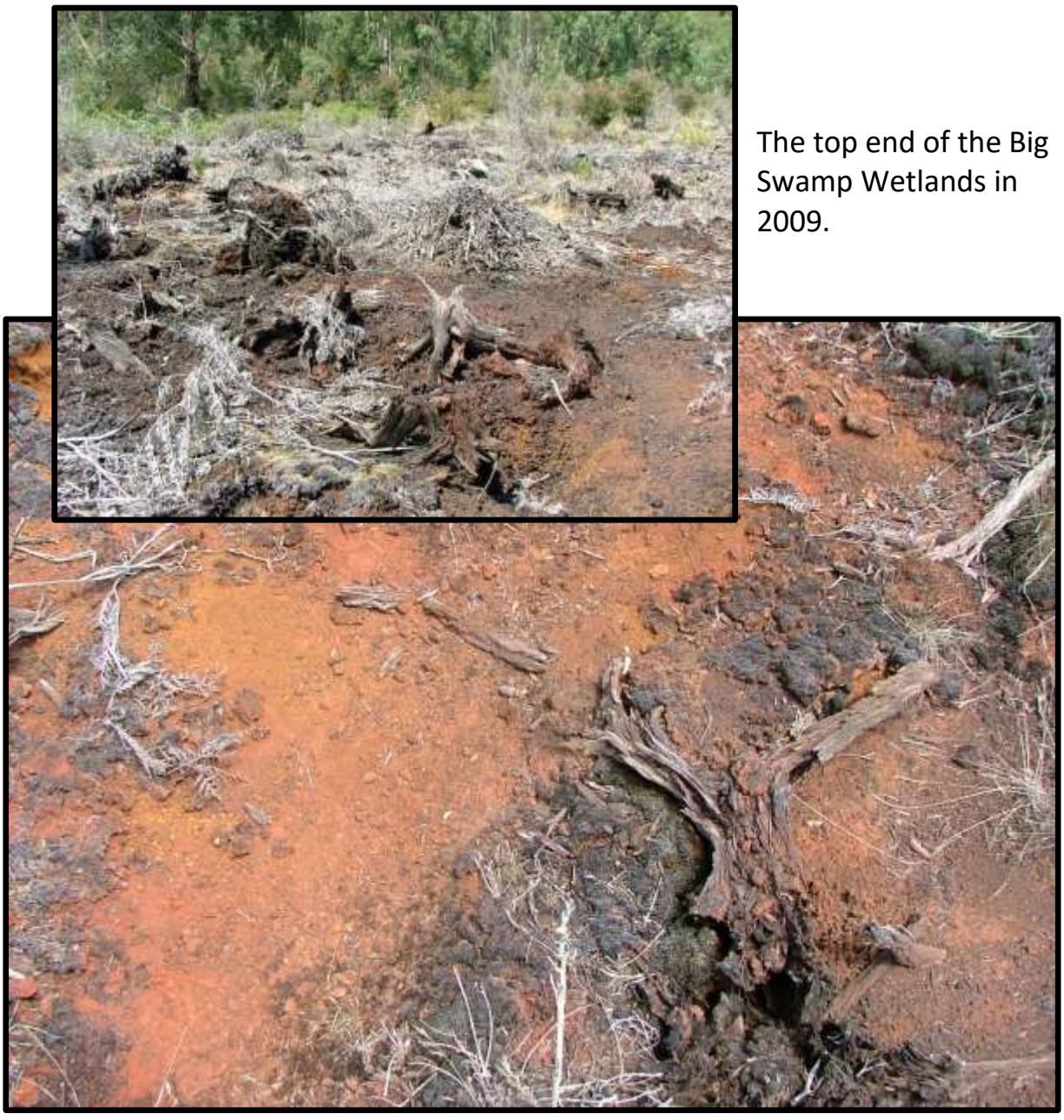
INTRODUCTION.

After the top end of the Big Swamp Wetlands at Yeodene, Victoria Australia, caught fire in the summer of 1997-1998 the vegetation never recovered to any noticeable degree until around 2015. For twelve years the site of the 1990's fire was hydrophobic and never grew a thing. It remained barren, apparently devoid of life. The Actual Acid Sulfate Soil made sure of this. From 1998 this site in the top end of the wetlands produced vegetation killing acid and heavy metals. As the Big Swamp Wetlands continued to dry out more and more pollutants spread down through the wetlands killing just about everything in its path.



The top end of the Big Swamp Wetlands in 2009.

Two photographs pre the 2010 fires showing the spread of the vegetation kill down through the wetlands.

After a series of flushes through the system the vegetation attempted to re-establish in the 1997 burnt out area with little success.

This photograph shows Tea Tree around the edges of the 1997 fire, Tea Tree that tried to establish but had succumbed to the pollutants. No regrowth in

12 years. Even bracken fern could not survive.

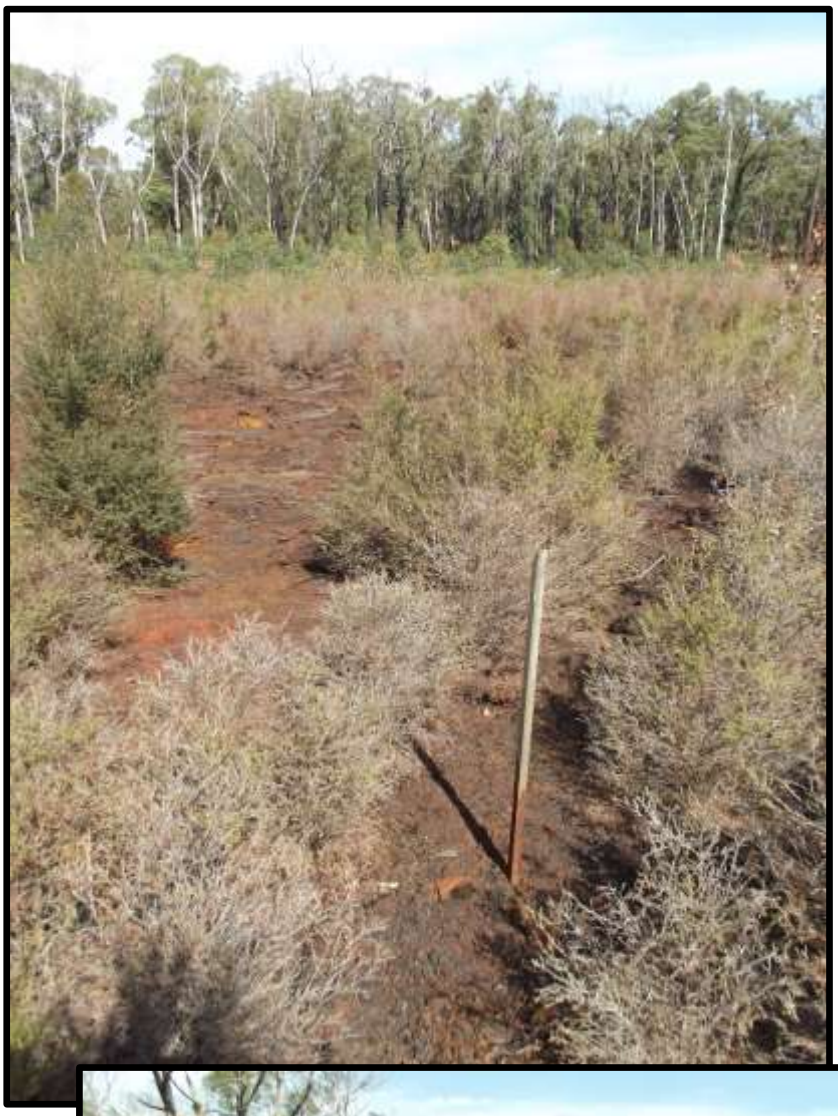
Then, along came the 2010 fire. It burnt intensively, fuelled by the large volumes of dead and dying vegetation.

Photo 2011 showing the depth of peat burnt in this particular area of the wetlands.

Photographs taken in 2011. Massive changes.

Photo 2011. ewetting to the north in the Big Swamp close to the Boundary Creek streambed.

2011. Vegetation attempting to re-establish. Looking north towards Boundary Creek.



Once again the Tea Tree attempted to re-establish but failed. This photograph was taken in 2013. The galvanised dropper was placed here during the 2008 vegetation survey. It had been lifted up showing the acid impact.

The photograph below shows eucalyptus trees that germinated and survived for a short period before dying off. The roots of these trees spread out across the top of the soil in a similar way to strawberry runners.

This is the result of an effort by plants to re-establish but as seasons came and went the plants were unable to flourish.

It took quite a few efforts over several seasons before the vegetation was able to re-establish in some areas of the Big Swamp.

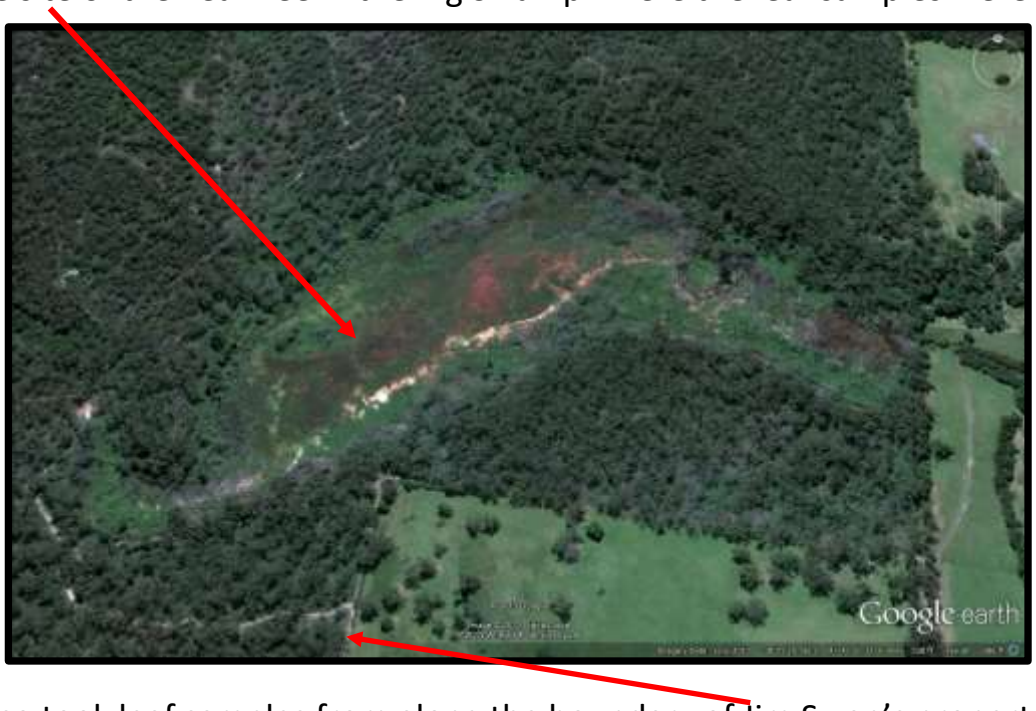
Having seen plants attempting to survive in what was once the Big Swamp Wetlands I was interested to take leaf samples for analysis. In 2017 I chose a next generation of Tea Tree that was surviving and looking healthy, just like its predecessors had, to see what could be learnt from the make up of the leaves before and after the shrub died. My thinking at the time was that the vegetation died either when the water table with any acid and heavy metals rose, or when the plants sent roots down looking for moisture into this toxic mix during drier periods. Or, a combination of both. I thought maybe leaf analysis might throw some light on what was happening.

The first sampling was done in 2017 but was never followed up due to so many other important groundwater issues happening during this period. Also, from regular visits and observations, the Tea Tree in the swamp did not appear to be under any stress as time went by. Not like earlier growings. As the plants were not dying thoughts and work on leaf analysis was put aside.

This photo shows the generation of vegetation from 2014.

As it turned out it looked like the plants were maintaining a relatively healthy existence. They continued to survive and even when there was a very high acid and heavy metal discharge in 2016 from the area, the plants seemd to suffer little, if at all.

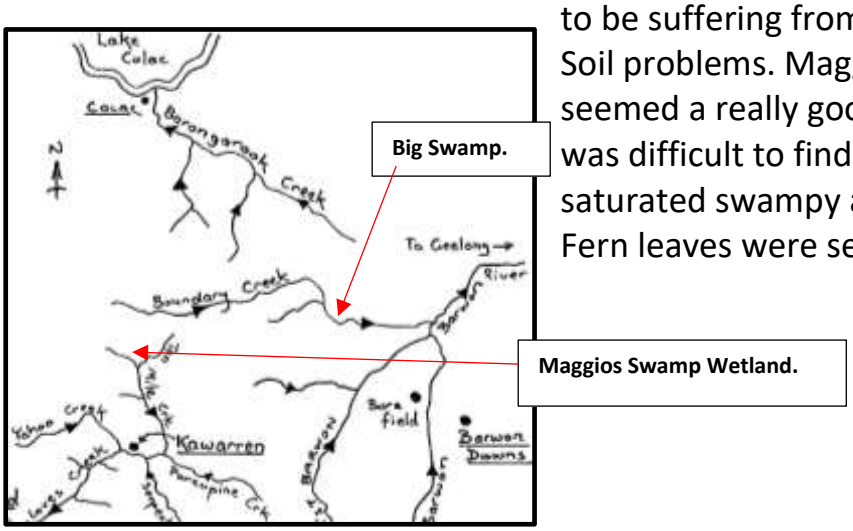
The site of the Tea Tree in the Big Swamp where the leaf samples were taken.



I also took leaf samples from along the boundary of Jim Swan's property and samples from Maggio's Swamp in the Ten Mile Creek Catchment. Perhaps the analysis results would help clarify part of the reason why the plants were dying off each season.

The Maggio's Swamp Wetland Sample.

I wanted to take Tea Tree samples from a swamp wetland that did not appear to be suffering from Actual Acid Sulfate Soil problems. Maggio's Swamp Wetland seemed a really good choice. However, it was difficult to find any Tea Tree in the saturated swampy area. This is why Elf Fern leaves were selected.



Maggio’s Swamp Wetland.

Maggio's Swamp Wetland.

New Thoughts - 2021.

Several factors came together in 2021 that prompted taking another look at the leaf sampling results.

- The vegetation in the Big Swamp was surviving and outwardly appearing quite healthy prompting thoughts that maybe nature was capable of remediating the Big Swamp problems.
- Thoughts prompted the idea that rather than the pollutants killing off each successive generation of plants being a bad thing, was it possible

that each year the plants were slowly taking up and somehow neutralising the toxicity?

- Perhaps humans could take a back seat and let nature do its thing.
- Given the cessation of groundwater extraction and the Lower Tertiary Aquifer hydraulic pressure heads recovering, why spend millions more dollars on "artificial" human induced remediation measures? Were natural processes doing the job? At the very best, effort to work with nature and natural processes might be the way to go.
- It was also worthy of consideration that perhaps local communities impacted by the past poor management of the Barwon Downs Borefield were prepared to…
    - accept the assurances from authorities that the aquifers of the area would no longer be under any pressure from extraction by Barwon Water. Iron clad assurances that is. And,
    - as a result were prepared to bear some pain that could be tolerated as nature took it's course.
- Was it possible that certain plant species could be used to help speed up the processes of the Big Swamp Wetlands recovery?
- Re-examining the leaf analysis results taken in 2017 seemed a good idea.

While waiting for the test results in 2017.

In 2017 when waiting for the first test results to come through an attempt was made to see how things such as aluminium, iron, arsenic etc. interact with one another and as a consequence impact on plants. Lots has been written on agricultural crops and garden plants regarding imbalances and how to possibly correct these. However, knowing exactly what is going on and how the various elements inter-react is a bit of a mine field. There appeared to be very little on the interpretation of leaf analysis results in Actual Acid Sulfate Soil site situations. Not that it is surprising. Appendix Four gives an insight into the inherent problems understanding the interactions and processes that plants adopt and highlights the fact that there appears to be very little work done on the subject of how plants can assist in rehabilitation of polluted sites.

Acid Loving Plants?

It would appear that Australian native plants are acid loving plants demanding a good supply of iron. But, I couldn't find any reference to what iron levels the acid loving plants desired. Another comment gave some degree of comfort, that Australian plants may be useful in neutralising toxins, stating Tea Tree plants naturally accumulate fluoride and other heavy metals including lead,

aluminium and arsenic. The Web has these throw away lines but with no follow up data, reports or investigations how vegetation…

- is impacted by polluted sites, or
- how plants may be able to assist in the re-habitation of polluted sites, still requires some considerable work.

Appendix Four contains an extract from a text from "Plant Analysis, an Interpretation Manual" that illustrates the complexities of plant analysis. It also typifies that the emphasis of most plant analysis text targets a different type of situation to the re-habitation of an Actual Acid Sulfate Soil wetlands site.

The Following Table is a Summary of the Analysis Results found in the Appendices.

Nutrient Tested for…	**Sample One. Big Swamp. Tea Tree leaves. 28/04/2017.**	**Sample Three. West Boundary Swan's Property. Tea Tree. 14/07/2017.**	**Sample Two. Maggios Swamp. Elf Fern. 28/04/2017.**
Nitrogen % units	0.88	0.98	1.51
Phosphorus %	0.05	0.05	0.06
Potassium % units	0.44	0.32	1.38
Sulfur %	0.13	0.11	0.21
Carbon %	50.08	52.6	44.8
Calcium %	0.82	0.94	0.50
Magnesium %	0.17	0.27	0.54
Sodium %	0.17	0.16	0.26
Copper mg/kg	3.0	3.8	7.9
Zinc mg/kg	10	17	10
Manganese mg/kg	111	177	135
Iron mg/kg	623	87	106
Boron mg/kg	25	23	56
Molybdenum mg/kg	<0.2	<0.2	<0.2
Cobalt mg/kg	0.1	0.2	<0.1
Silicon mg/kg	352	350	803
Nitrogen:Sulfur ratio units	6.9	9.2	7.2
Nitrogen:Phosporus ratio units	16.2	21.5	23.3

Nitrogen:Potassium ratio units	2.0	3.1	1.1
Carbon:Nitrogen	57.2	53.5	29.7
Crude Protein ratio units	5.5	6.2	9.4
Aliminium mg/kg	126	65	99
Selenium mg/kg	<0.5	<0.5	<0.5
Cadmium mg/kg	<0.1	<0.1	<0.1
Lead mg/kg	<0.5	<0.5	<0.5
Arsenic mg/kg	0.4	0.1	<0.1
Chromium mg/kg	<1	<1	<1
Nickel mg/kg	29	4.4	8.3
Mercury mg/kg	<0.1	<0.1	<0.1
Silver mg/kg	<0.1	<0.1	<0.1

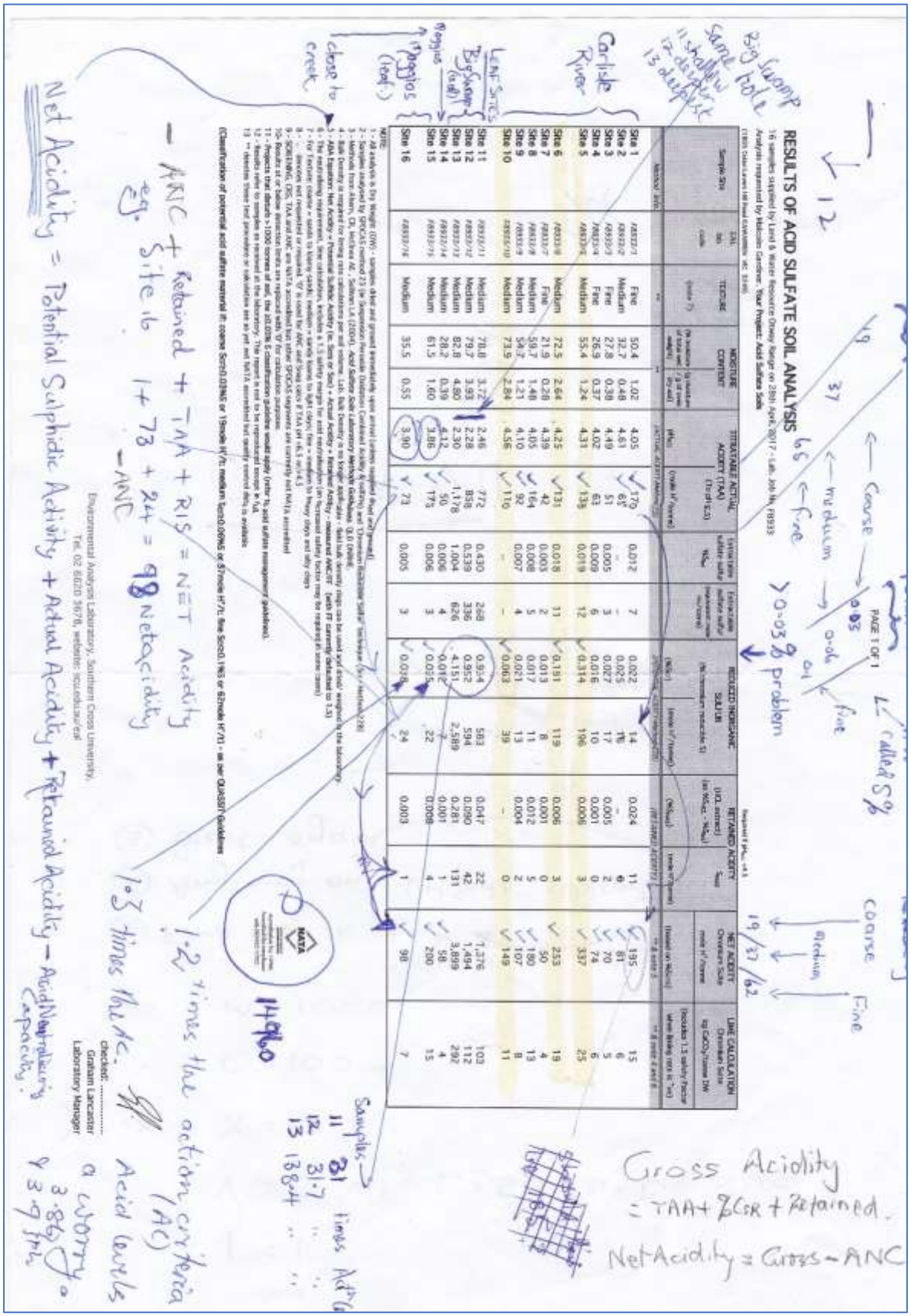
A Story to Tell.

It would appear that Zinc, Manganese, Iron, Silicon, Aluminium, Arsenic and Nickel levels in the Big Swamp Tea Tree samples have a story to tell.

CONCLUSION.

When looking at these very limited leaf analysis results, there would appear to be enough indications that “experts” in the field of leaf analysis should be involved in the future development of acid and heavy metal soil rehabilitation in the Big Swamp with an emphasis on the use of native vegetation.

APPENDIX ONE.

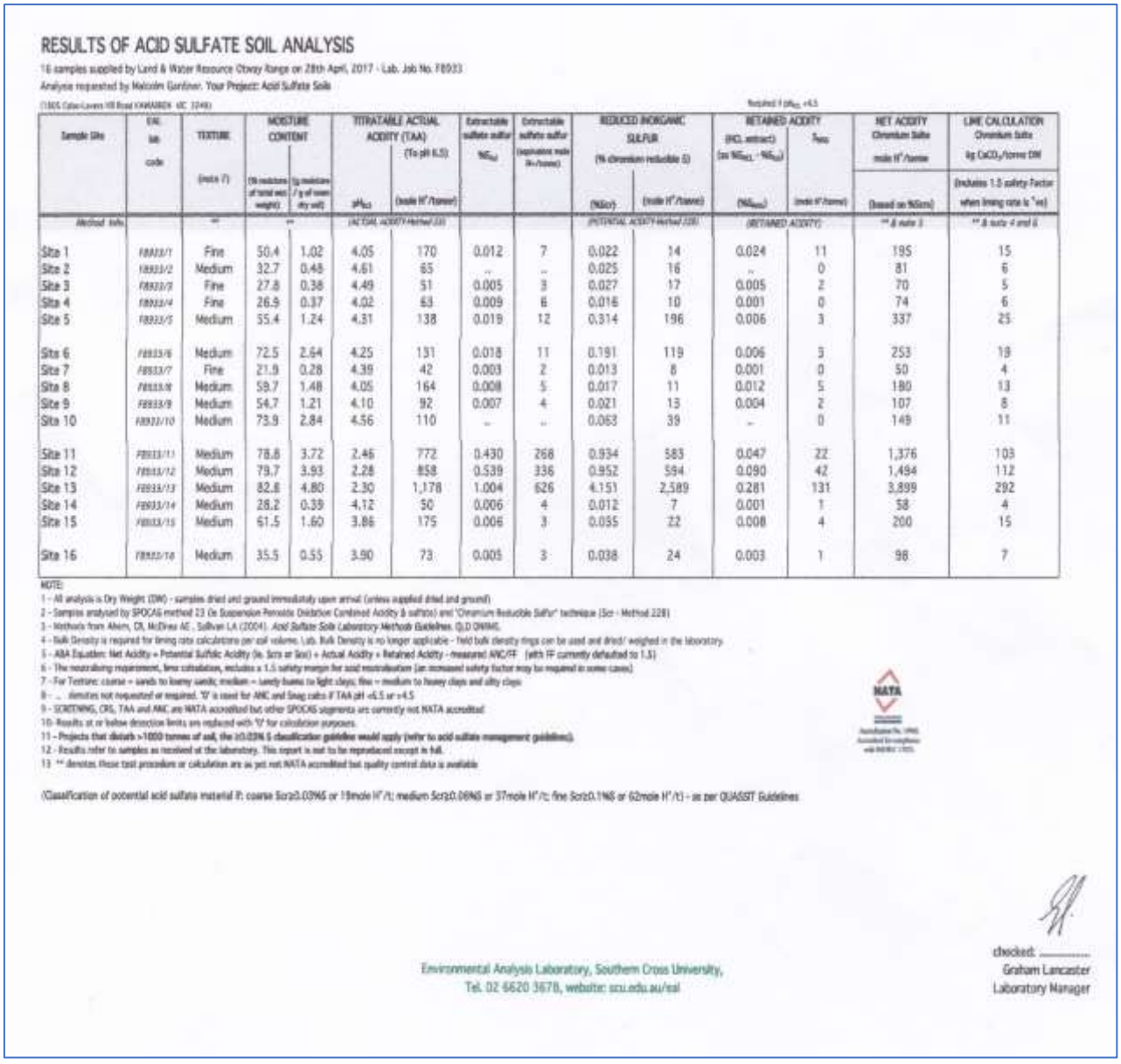


RESULTS OF ACID SULFATE SOIL ANALYSIS

Acid Sulfate Soil test results with my scribblings.

Same document as above.

Site 11, 12 and 13 below, where soil samples were taken at different levels at the Tea Tree site (Sample 1) in the Big Swamp.



RESULTS OF ACID SULFATE SOIL ANALYSIS

16 samples supplied by Land & Water Resource Otway Range on 28th April, 2017 - Lab. Job No. F8933

Analysis requested by Malcolm Gardiner. Your Project: Acid Sulfate Soils

Sample Site	ESAL lab code	TEXTURE (note 1)	MOISTURE CONTENT (% moisture of total wet weight)	MOISTURE CONTENT (g moisture / g of oven dry soil)	TITRATABLE ACTUAL ACIDITY (TAA) pHKCl	TITRATABLE ACTUAL ACIDITY (TAA) (To pH 6.5) (moles H+/tonne)	Extractable sulfate sulfur %SKCl	Extractable sulfate sulfur (equivalent mole H+/tonne)	REDUCED INORGANIC SULFUR (% chromium reducible S) (%Scr)	REDUCED INORGANIC SULFUR (% chromium reducible S) (moles H+/tonne)	RETAINED ACIDITY (HCl extract) (as %SHCl - %SKCl)	RETAINED ACIDITY fhcl (moles H+/tonne)	NET ACIDITY Chromium Suite mole H+/tonne (based on %Scrs)	LIME CALCULATION Chromium Suite kg CaCO3/tonne DM (includes 1.5 safety Factor when liming rate is "+ve")
Method Info		**	**		(ACTUAL ACIDITY Method 23)				(POTENTIAL ACIDITY Method 22B)		(RETAINED ACIDITY)		** & note 3	** & note 4 and 8
Site 1	F8933/1	Fine	50.4	1.02	4.05	170	0.012	7	0.022	14	0.024	11	195	15
Site 2	F8933/2	Medium	32.7	0.48	4.61	65	..	..	0.025	16	..	0	81	6
Site 3	F8933/3	Fine	27.8	0.38	4.49	51	0.005	3	0.027	17	0.005	2	70	5
Site 4	F8933/4	Fine	26.9	0.37	4.02	63	0.009	6	0.016	10	0.001	0	74	6
Site 5	F8933/5	Medium	55.4	1.24	4.31	138	0.019	12	0.314	196	0.006	3	337	25
Site 6	F8933/6	Medium	72.5	2.64	4.25	131	0.018	11	0.191	119	0.006	3	253	19
Site 7	F8933/7	Fine	21.9	0.28	4.39	42	0.003	2	0.013	8	0.001	0	50	4
Site 8	F8933/8	Medium	59.7	1.48	4.05	164	0.008	5	0.017	11	0.012	5	180	13
Site 9	F8933/9	Medium	54.7	1.21	4.10	92	0.007	4	0.021	13	0.004	2	107	8
Site 10	F8933/10	Medium	73.9	2.84	4.56	110	..	..	0.063	39	..	0	149	11
Site 11	F8933/11	Medium	78.8	3.72	2.46	772	0.430	268	0.934	583	0.047	22	1,376	103
Site 12	F8933/12	Medium	79.7	3.93	2.28	858	0.539	336	0.952	594	0.090	42	1,494	112
Site 13	F8933/13	Medium	82.8	4.80	2.30	1,178	1.004	626	4.151	2,589	0.281	131	3,899	292
Site 14	F8933/14	Medium	28.2	0.39	4.12	50	0.006	4	0.012	7	0.001	1	58	4
Site 15	F8933/15	Medium	61.5	1.60	3.86	175	0.006	3	0.055	22	0.008	4	200	15
Site 16	F8933/16	Medium	35.5	0.55	3.90	73	0.005	3	0.038	24	0.003	1	98	7

NOTE:

1 - All analysis is Dry Weight (DW) - samples dried and ground immediately upon arrival (unless supplied dried and ground)

2 - Samples analysed by SPOCAS method 23 (Ie Suspension Peroxide Oxidation Combined Acidity & sulfate) and 'Chromium Reducible Sulfur' technique (Scr - Method 22B)

3 - Methods from Ahern, CR, McElnea AE , Sullivan LA (2004). *Acid Sulfate Soils Laboratory Methods Guidelines*. QLD DNRME.

4 - Bulk Density is required for liming rate calculations per soil volume. Lab. Bulk Density is no longer applicable - field bulk density rings can be used and dried/ weighed in the laboratory.

5 - ABA Equation: Net Acidity = Potential Sulfidic Acidity (ie. Scrs or Scr) + Actual Acidity + Retained Acidity - measured ANC/FF (with FF currently defaulted to 1.5)

6 - The neutralising requirement, lime calculation, includes a 1.5 safety margin for acid neutralisation (an increased safety factor may be required in some cases).

7 - For Texture: coarse = sands to loamy sands; medium = sandy loams to light clays; fine = medium to heavy clays and silty clays

8 - '..' denotes not requested or required. '0' is used for ANC and Snag cells if TAA pH >6.5 or >4.5

9 - SCRTWING, CRS, TAA and ANC are NATA accredited but other SPOCAS segments are currently not NATA accredited

10 - Results at or below detection limits are replaced with '0' for calculation purposes.

11 - Projects that disturb >1000 tonnes of soil, the ≥0.03% S classification guideline would apply (refer to acid sulfate management guidelines).

12 - Results refer to samples as received at the laboratory. This report is not to be reproduced except in full.

13 ** denotes those test procedure or calculation are as yet not NATA accredited but quality control data is available

(Classification of potential acid sulfate material P: coarse Scr≥0.03%S or 18mole H+/t; medium Scr≥0.06%S or 37mole H+/t; fine Scr≥0.1%S or 62mole H+/t) - as per QUASSIT Guidelines

NATA

Environmental Analysis Laboratory, Southern Cross University,
Tel. 02 6620 3678, website: scu.edu.au/eal

checked: .....
Graham Lancaster
Laboratory Manager

Sites 15 and 16 were taken at the leaf sampling site (Sample 2) in Maggios Swamp.

Soil sample taken at the leaf sampling site (Sample 3) along the west boundary of the Swan property south of the Big Swamp.

RESULTS OF ACID SULFATE SOIL ANALYSIS

1 sample supplied by Land & Water Resource Otway Ranges on 14th July, 2017 - Lab. Job No. 61223
Analysis requested by Malcolm Gardiner. **Your Project: A/S Soil**

1803 Colac – Lavers Hill Road KANARGEN VIC 3249

Sample Site	EAL lab code	TEXTURE (note 7)	MOISTURE CONTENT (% moisture of total wet weight)	MOISTURE CONTENT (g moisture / g of oven dry soil)	TITRATABLE ACTUAL ACIDITY (TAA) pH$_{KCl}$	TITRATABLE ACTUAL ACIDITY (TAA) (To pH 6.5) (mole H$^+$/tonne)	REDUCED INORGANIC SULFUR (% chromium reducible S) (%Scr)	REDUCED INORGANIC SULFUR (% chromium reducible S) (mole H$^+$/tonne)	NET ACIDITY Chromium Suite mole H$^+$/tonne (based on %Scrs)	LIME CALCULATION Chromium Suite kg $CaCO_3$/tonne DW (includes 1.5 safety Factor when liming rate is ^{t}/ML)
Method refs.		**	**		*(ACTUAL ACIDITY-Method 23)*		*(POTENTIAL ACIDITY-Method 22B)*		*** & note 5*	*** & note 4 and 8*
Soil	61225/1	..	18.1	0.19	4.54	20	<0.005	0	20	1.5

NOTE:
1 - All analysis is Dry Weight (DW) - samples dried and ground immediately upon arrival (unless supplied dried and ground)
2 - Samples analysed by SPOCAS method 23 (ie Suspension Peroxide Oxidation Combined Acidity & sulfate) and 'Chromium Reducible Sulfur' technique (Scr - Method 22B)
3 - Methods from Ahern, CR, McElnea AE , Sullivan LA (2004). ***Acid Sulfate Soils Laboratory Methods Guidelines.*** QLD DNRME.
4 - Bulk Density is required for liming rate calculations per soil volume. Lab. Bulk Density is no longer applicable - field bulk density rings can be used and dried/ weighed in the laboratory.
5 - ABA Equation: Net Acidity = Potential Sulfidic Acidity (ie. Scrs or Scs) + Actual Acidity + Retained Acidity - measured ANC/FF (with FF currently defaulted to 1.5)
6 - The neutralising requirement, lime calculation, includes a 1.5 safety margin for acid neutralisation (an increased safety factor may be required in some cases)
7 - For Texture: coarse = sands to loamy sands; medium = sandy loams to light clays; fine = medium to heavy clays and silty clays
8 - .. denotes not requested or required. '0' is used for ANC and Snag calcs if TAA pH >6.5 or >4.5
9 - SCREENING, CRS, TAA and ANC are NATA accredited but other SPOCAS segments are currently not NATA accredited
10- Results at or below detection limits are replaced with '0' for calculation purposes.
11 - Projects that disturb >1000 tonnes of soil, the ≥0.03% S classification guideline would apply (refer to acid sulfate management guidelines).
12 - Results refer to samples as received at the laboratory. This report is not to be reproduced except in full.
13 - ** denotes these test procedure or calculation are as yet not NATA accredited but quality control data is available

(Classification of potential acid sulfate material if: coarse Scr≥0.03%S or 19mole H$^+$/t; medium Scr≥0.06%S or 37mole H$^+$/t; fine Scr≥0.1%S or 62mole H$^+$/t) - as per QUASSIT Guidelines

NATA

checked: .....
Graham Lancaster
Laboratory Manager

Environmental Analysis Laboratory, Southern Cross University,
Tel. 02 6620 3678, website: scu.edu.au/eal

APPENDIX TWO.

Test Results for Samples 1 and 2.

PLANT TISSUE ANALYSIS REPORT

2 samples supplied by Land & Water Resource Otway Ranges on 28th April, 2017 - Lab Job No. F8934.

Analysis requested by Malcolm Gardiner.

				Sample ID:	Sample 1 Site 11,12,13	Sample 2 Site 15,16	Guideline TEA TREE	
				Crop:	T-Tree	Elf Fern	(Leptospermum lanigerum)	(Leptospermum rotundifolium)
				Client:	LAWROC	LAWROC		
	Nutrient		Units		F89343	F89342	Adequate	Adequate
Macronutrients	Nitrogen	N	%		0.88	1.51	1.11-1.29	1.29-1.79
	Phosphorus	P	%		0.05	0.06	0.09-0.23	0.48-0.85
	Potassium	K	%		0.44	1.38	0.69-0.89	0.98-1.59
	Sulfur	S	%		0.13	0.21	-	-
	Carbon	C	%		50.8	44.8	-	-
	Calcium	Ca	%		0.82	0.50	0.76-1.04	0.90-1.52
	Magnesium	Mg	%		0.17	0.54	0.14-0.32	0.13-0.33
	Sodium	Na	%		0.17	0.26	-	-
							-	-
Micronutrients	Copper	Cu	mg/kg		3.0	7.9	-	-
	Zinc	Zn	mg/kg		10	10	-	-
	Manganese	Mn	mg/kg		111	135	-	-
	Iron	Fe	mg/kg		623	106	-	-
	Boron	B	mg/kg		25	56	-	-
	Molybdenum	Mo	mg/kg		<0.2	<0.2	-	-
	Cobalt	Co	mg/kg		0.1	<0.1	-	-
	Silicon	Si	mg/kg		352	803	-	-
Calculations	Nitrogen : Sulfur	ratio	units		6.9	7.2	-	-
	Nitrogen : Phosphorus	ratio	units		16.2	23.3	-	-
	Nitrogen : Potassium	ratio	units		2.0	1.1	-	-
	Carbon : Nitrogen	ratio	units		57.5	29.7	-	-
	Crude Protein		%		5.5	9.4	-	-
Heavy Metals	Aluminium	Al	mg/kg		129	99	-	-
	Selenium	Se	mg/kg		<0.5	<0.5	-	-
	Cadmium	Cd	mg/kg		<0.1	<0.1	-	-
	Lead	Pb	mg/kg		<0.5	<0.5	-	-
	Arsenic	As	mg/kg		0.4	<0.1	-	-
	Chromium	Cr	mg/kg		<1	<1	-	-
	Nickel	Ni	mg/kg		29	8.3	-	-
	Mercury	Hg	mg/kg		<0.1	<0.1	-	-
	Silver	Ag	mg/kg		<0.1	<0.1	-	-

Agricultural laboratory testing notes:

1. All analysis is dry weight - Samples dried at 70°C for 24 hours prior to fine grinding
2. Unless requested, leaf samples are NOT washed to remove soil spray or liquid fertilisers prior to analysis
3. Samples are hotblock digested with nitric acid and read on the ICP-MS
4. Carbon / Nitrogen / Sulfur measured using a LECO CNS2000 Analyser
5. mg/kg = ppm
6. By Calculation - Crude Protein = %N x 6.25
7. Nitrate / Ammonium / Chloride measured on a water extract.
8. Moisture based on sample dried at 105°C

Quality Checked: Kris Saville
Manager, Agricultural testing division

APPENDIX THREE.

Test Results for Sample 3.

Environmental Analysis Laboratory
Southern Cross University

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PO Box 157 Lismore NSW 2480

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E: eal@scu.edu.au
W: scu.edu.au/eal
ABN: 41 995 651 524

PLANT TISSUE ANALYSIS REPORT

1 sample supplied by L.W.R.O.R on the 14th July 2017 - Lab Job No. G1226.

Analysis requested by Malcolm Gardiner.

				Sample 1
		Sample ID:		T-Tree
		Crop:		T-Tree
		Client:		M. Gardiner
	Nutrient		Units	G1226/1
Macronutrients	Nitrogen	N	%	0.98
	Phosphorus	P	%	0.05
	Potassium	K	%	0.32
	Sulfur	S	%	0.11
	Carbon	C	%	52.6
	Calcium	Ca	%	0.94
	Magnesium	Mg	%	0.27
	Sodium	Na	%	0.16
Micronutrients	Copper	Cu	mg/kg	3.6
	Zinc	Zn	mg/kg	17
	Manganese	Mn	mg/kg	177
	Iron	Fe	mg/kg	87
	Boron	B	mg/kg	23
	Molybdenum	Mo	mg/kg	<0.2
	Cobalt	Co	mg/kg	0.2
	Silicon	Si	mg/kg	350
Calculations	Nitrogen : Sulfur	ratio	units	9.2
	Nitrogen : Phosphorus	ratio	units	21.5
	Nitrogen : Potassium	ratio	units	3.1
	Carbon : Nitrogen	ratio	units	53.5
	Crude Protein		%	6.2

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eal Environmental Analysis Laboratory
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				Sample 1
			Sample ID:	T-Tree
			Crop:	T-Tree
			Client:	M. Gardiner
	Nutrient		Units	G1226/1
Heavy Metals	Aluminium	Al	mg/kg	65
	Selenium	Se	mg/kg	<0.5
	Cadmium	Cd	mg/kg	<0.1
	Lead	Pb	mg/kg	<0.5
	Arsenic	As	mg/kg	0.1
	Chromium	Cr	mg/kg	<1
	Nickel	Ni	mg/kg	4.4
	Mercury	Hg	mg/kg	<0.1
	Silver	Ag	mg/kg	<0.1

Agricultural laboratory testing notes:

1. All analysis is dry weight - Samples dried at 70°C for 24 hours prior to fine grinding
2. Unless requested, leaf samples are NOT washed to remove salt spray or liquid fertilizers prior to analysis
3. Samples are hotblock digested with nitric acid and read on the ICP-MS
4. Carbon / Nitrogen / Sulfur measured using a LECO CNS2000 Analyser
5. mg/kg = ppm
6. By Calculation:- Crude Protein = %N x 6.25
7. Nitrate / Ammonium / Chloride measured on a water extract.
8. Moisture based on sample dried at 105°C

Quality Checked: Kris Saville
Manager, Agricultural testing division

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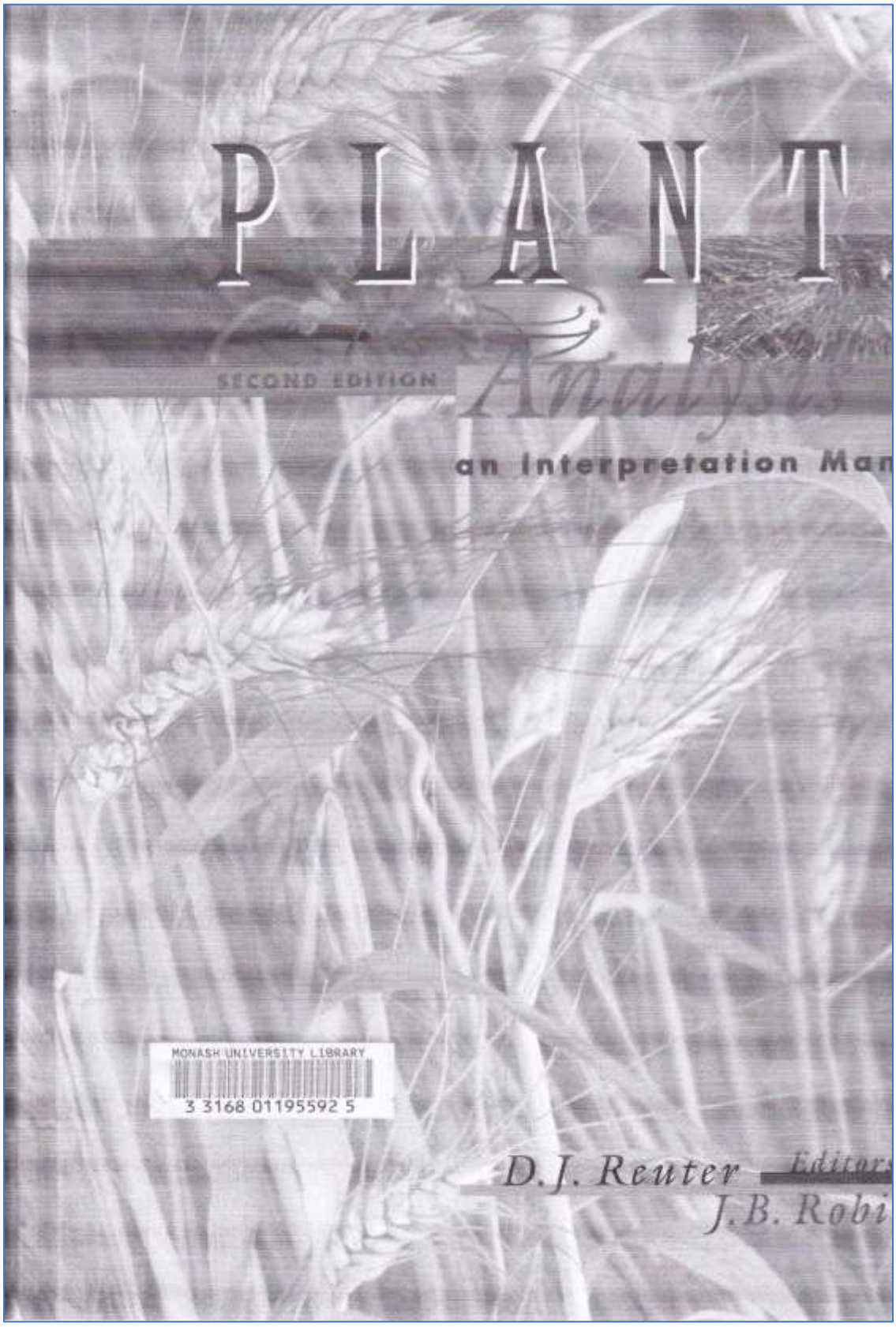
APPENDIX FOUR.



2. Nutrient Deficiency and Toxicity Symptoms

N.J. Grundon, A.D. Robson, M.J. Lambert and K. Snowball

A. Introduction

Nutrient deficiencies and toxicities cause impaired metabolism within plants resulting in the appearance of visible symptoms. Many symptoms are sufficiently characteristic to permit identification of the disorder causing the impaired metabolism and reduced growth. Other symptoms are less characteristic, and their presence could indicate one of several possible stresses. For example, a general paleness of shoots with reddening and premature senescence of old leaves could indicate nitrogen, sulfur or molybdenum deficiency in legumes (Andrew and Pieters 1972; Smith and Pieters 1983; Smith *et al.* 1983; Snowball and Robson 1983), while in pines (Turner *et al.* 1979) and eucalypts (Will 1985), these symptoms could indicate nitrogen or sulfur deficiency. Failure of terminal spikelets in cereals to produce grain can be a symptom of moisture stress or frost damage at anthesis, root disease, mouse or insect attack, or copper deficiency (Graham 1975; Grundon 1987; King 1974; Snowball and Robson 1983). By contrast, a number of distinct symptoms may be produced by the one disorder. For example, boron deficiency generally causes death of growing points, but in pines it results also in buds failing to flush, and the main stem forking and becoming deformed (Turner *et al.* 1979).

The use of visual symptoms to diagnose nutrient disorders has distinct advantages for the extension worker, agronomist, farmer, or forest manager. Two great advantages are: (i) that the technique can be applied in the field; and (ii) that it is not dependent on laboratory support services.

However, there are a number of disadvantages in relying on visible symptoms as the sole diagnostic tool. One major disadvantage is that a disorder is diagnosed only after severe stress has occurred and yield may have been severely depressed. Another major disadvantage is that, by the time distinctive symptoms become evident, it is often too late to correct the problem in that growing season.

Even in the hands of experienced workers, the use of foliar symptoms as a diagnostic tool must be undertaken with care because a number of factors affect the form and appearance of visual symptoms and their usefulness as a diagnostic tool for nutrient disorders:

- Under certain conditions, different nutrient disorders can produce rather similar symptoms. At other times, the effects of insect pests or diseases may produce symptoms similar to those of specific nutrient disorders. Practitioners need to be able to distinguish between symptoms caused by nutrient disorders and those caused by pathological infection, insect damage, senescence, and management practices. As these facts have been recognized, some authors have included specific comments, sections or keys referring to the similarity of symptoms produced by different nutrient disorders, insect attack, or disease infection (Turner *et al.* 1979; Blamey *et al.* 1987; Grundon *et al.* 1987; O'Sullivan *et al.* 1995).
- When the disorder is mild or transient in nature, no foliar symptoms may be produced but seed production may still fail completely, as in copper deficiency in wheat (Graham 1975; Grundon 1987).
- With some crop species, there are considerable differences between cultivars in the form of expression of the symptoms; examples include magnesium deficiency in grain sorghum (Grundon *et al.* 1987), and nitrogen, phosphorus and magnesium deficiencies in sweet potato (O'Sullivan *et al.* 1995).
- For some micronutrient disorders, environmental factors such as light, temperature and soil moisture conditions may have a profound effect on the appearance or severity of the symptoms (Moraghan and Mascagni 1991). For example, in most field crops, iron, manganese and zinc deficiencies are increased in severity by a combination of low temperature and high soil moisture, while hot, dry summers intensify the severity of iron deficiency. However, in forest trees, zinc deficiency is more acute in summer than in winter (Boardman and McGuire 1990), and in both field crops and trees symptoms of zinc deficiency are more severe at high light intensities than in partial shade (e.g. Cakmak *et al.* 1995);
- There are significant differences between species, and even between cultivars or varieties within species, in their sensitivity to nutrient disorders. For example, Honduras Caribbean pine (*Pinus*

caribaea var. *hondurensis*) is much more sensitive to copper deficiency than slashpine (*Pinus elliotti*), and similar differences occur between individual trees of their F1 hybrids (Simpson and Osborne 1993). Then again, different provenances or families of Honduras Caribbean pine respond differently to fertilizers and have different sensitivity to copper deficiency in the presence or absence of nitrogen (Simpson *et al.* 1996).

- Plants may suffer also from multiple nutrient disorders on sites where more than one element may be deficient and/or toxic. However, the symptoms may not always be characteristic of one disorder, and may represent a combination of the more strongly expressed symptoms of individual disorders. In these instances, very complex symptoms can develop, and considerable skill and experience is required for a correct diagnosis. Then again, where multiple deficiencies exist, symptoms of the most limiting deficiency may be exhibited; when this nutrient is applied to the plant or soil, new symptoms develop that are characteristic of the second nutrient limiting growth. Specific symptoms may also occur where excessive levels of other nutrients depress either the uptake or transport of another nutrient to the shoots. For example, symptoms of iron deficiency often develop in the presence of high levels of manganese, zinc, aluminium, phosphorus and some non-essential heavy metals in the growing medium (Clark *et al.* 1981; Blamey *et al.* 1987; Grundon *et al.* 1987).

Despite any limitations to their usefulness, plant symptoms remain a valuable diagnostic tool, especially when applied by an experienced practitioner. However, few protocols for the successful application of the technique are given in the literature. Grundon (1987) lists some of the items to be considered, which include recording the history of the problem and the pattern of development of the symptoms. For forest species, Dell *et al.* (1995) list the distinctive characteristics of nutrient disorders that can be used to distinguish them from symptoms caused by other factors.

It is important to recognize that the use of visible symptoms provides a preliminary diagnosis. Confirmation by other methods such as plant and soil analyses, pot culture assays, field experiments, or test strips is an essential second step.

In this chapter, symptoms of nutrient stresses are considered in relation to function and distribution of nutrients within plants. A list of published descriptions of symptoms of nutrient deficiencies and toxicities is given in Appendix 1 to this Chapter.

B. Symptoms in Relation to the Function of Nutrients

There are several levels of knowledge of nutrient function. In ascending order these are: (i) the element is essential; (ii) the element plays a role in a physiological process; (iii) the element activates an enzyme or regulates the rate of an enzyme-mediated process; and (iv) the element is an integral constituent of an essential metabolite, complex or macromolecular assembly. Our knowledge of nutrient function varies for the essential nutrients (Table 2.1)

Essential elements may have non-specific roles in plant growth which may be additional to the specific functions listed. For example, many ions are important for the establishment of osmotic potentials within plants and for the maintenance of electrical neutrality. Another example is in maintenance of quality aspects of the harvested products: in fruits and vegetables, calcium deficiency often leads to reduced quality of storage tissues such as fruit (e.g. bitter pit of apples; blossom-end rot in tomato fruit).

It is clear that one nutrient may have several functions within the plant. The symptoms which are most characteristic for a particular nutrient are those in which one specific function dominates, causing a more visible symptom than those produced by the other functions. For example, zinc has been shown to be associated with auxin metabolism, nucleotide synthesis, and membrane integrity (Boardman and McGuire 1990; see Table 2.1), but it is impairment of auxin metabolism in zinc-deficient plants that leads to the characteristic symptoms of leaf distortion and internode shortening (rosetting).

When one nutrient is involved in the assimilation or metabolism of another nutrient, the symptoms may not clearly differentiate the causal element. For example, nitrogen and sulfur are biochemically related in plant proteins. Because there is no inorganic nitrogen in tree foliage, there is a constant ratio between organic (and total) nitrogen and organic sulfur, and the rate of nitrogen uptake is limited by the rate of sulfur accumulation (Turner and Lambert 1986). Thus, in many forest and horticultural tree species, protein formation is limited by the amount of sulfur available, and the symptoms of nitrogen and sulfur deficiency are very similar (Sprague 1964; Turner *et al.* 1979). Another example is in legumes, where molybdenum deficiency is indistinguishable from nitrogen deficiency because molybdenum is required most as a constituent of the enzyme involved in nitrogen fixation (nitrogenase). In plants other than legumes, symptoms of molybdenum deficiency depend on the level of nitrate supplied (see Snowball and Robson (1983) for wheat). At adequate but not luxury levels of nitrate, leaves of wheat plants deficient in molybdenum are paler and more flaccid than those on plants supplied with adequate molybdenum. When very high rates of nitrate are applied, tip scorch of old leaves may occur on molybdenum-deficient plants as a result of an excessive accumulation of nitrate. Molybdenum is a constituent of nitrate reductase, the enzyme involved in the first step of nitrate assimilation in leaf cells (see Table 2.1); nitrate reductase activity decreases the build-up of nitrate in leaves.

TABLE 2.1 Functions of essential elements in higher plants; for more detail see Epstein (1972), Mengel and Kirkby (1982), Marschner (1988), Asher (1991) and Tisdale *et al.* (1993)

Element	Physiological process	Activator of enzyme	Constituent of metabolite or cell component
Nitrogen			Amino acids, proteins, nucleic acids, nucleotides, chlorophyll
Phosphorus	Energy storage and transfer, membrane integrity		ATP, nucleotides, nucleic acids, phospholipids
Potassium	Translocation, water relations, energy relations, stomatal opening, regulation of cellular pH, osmoregulation, cation–anion balance	+	
Sulfur	Protein synthesis and function, energy transfer, structure		Amino acids, co-enzymes, ferredoxins, sulfolipids, proteins
Calcium	Membrane maintenance, cell division and elongation, cell wall stabilization, cation–anion balance, osmoregulation, second messenger in environmental signals	+	Calcium pectates
Magnesium	CO_2 assimilation, regulation of cellular pH, cation–anion balance, protein synthesis, carbohydrate partitioning	+	Chlorophyll, ribosomes
Chlorine	Maintenance of electrical neutrality, internal turgor		
Copper	Lignin synthesis, terminal oxidation in redox reactions, pollen formation and fertilization		Ascorbate oxidase, phenol oxidases, cytochrome oxidase, plastocyanin, CuZn superoxide dismutase
Zinc	Auxin metabolism, nucleotide synthesis, membrane integrity	+	Dehydrogenases, CuZn superoxide dismutase, carbonic anhydrase, RNA polymerase,alkaline phosphatase, phospholipase, carboxypeptidase
Manganese	Oxidation–reduction in electron transport, O_2 evolution in photosynthesis	+	Mn superoxide dismutase
Iron	Oxidation–reduction in electron transport		Iron porphyrins (leaves), ferredoxins
Boron	Nucleotide synthesis, assimilate translocation, cell wall biosynthesis and structure, plasma membrane integrity		
Nickel	Urea metabolism via urease	+	
Sodium	Conversion of pyruvate to phosphoenolpyruvate in C_4 photosynthetic pathway		
Molybdenum	Nitrogen fixation, nitrate reduction		Nitrogenase, nitrate reductase, xanthine oxidase/dehydrogenase

C. SYMPTOMS IN RELATION TO THE MOBILITY OF NUTRIENTS

The location of symptoms of nutrient deficiencies within plants depends on the extent and rate of retranslocation of nutrients within the plant. Nutrients differ markedly in their mobility within the plant (Table 2.2). Some nutrients, such as nitrogen, phosphorus and potassium, are readily retranslocated from old leaves to new growth. For these nutrients, the symptoms occur initially in the older leaves. Other nutrients, such as calcium and boron, have little or minimal mobility and do not appear to be retranslocated from old leaves to new growth. Hence, for these nutrients, deficiency symptoms occur generally in young growing areas of the plant.

Manganese does not appear to be retranslocated out of old leaves (Hill *et al.* 1979; Radjagukguk 1981; Nable and Loneragan 1984a, 1984b). Nevertheless, symptoms of manganese deficiency frequently occur in fully expanded young leaves rather than in new growth. This may reflect a greater internal requirement in these leaves than in new growth. Symptoms of manganese deficiency do not occur on old leaves.

TABLE 2.2 Mobility of nutrients within plants

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

Magnesium is considered to be a mobile nutrient, and thus it is expected that the symptoms of magnesium deficiency would appear first in older leaves. However, under certain conditions, the symptoms of magnesium deficiency occur initially in young leaves. For example, when magnesium supply to subterranean clover was constant but inadequate, symptoms of magnesium deficiency appeared, as expected, firstly in older leaves (Scott and Robson 1990). However, when magnesium supply to the roots was interrupted suddenly, the symptoms appeared first in young tissue. In young wheat seedlings, symptoms of magnesium deficiency appeared first in young leaves when the supply to the roots was interrupted suddenly, or when the supply was constant but inadequate (Scott and Robson 1991). In both species, the symptoms of yellowing and necrosis that characterize magnesium deficiency occurred when leaf concentrations of magnesium fell below 'critical' values. Although magnesium was retranslocated from older leaves when the supply was limiting, the symptoms presumably appeared first in young tissue when their concentrations were the first to fall below the 'critical' levels. Apparently, the rate of movement of magnesium from old leaves to new growth was not sufficient to meet the requirements of the new growth.

For many nutrients, the extent of retranslocation is variable and depends upon the degree of deficiency, the plant species, and either the nitrogen or phosphorus status of the plant. For example, there is little or no movement of copper, zinc and molybdenum out of old leaves of plants deficient in these elements, and symptoms occur mainly in young tissues. However, should nitrogen deficiency or some other physiological or environmental factor cause the older leaves to senesce, copper, zinc and molybdenum can be remobilized from older to younger leaves along with the retranslocation of nitrogen (Hill *et al.* 1978). For nutrients where nitrogen supply affects the movement of other nutrients from old leaves to new growth, the location of symptoms may vary with the supply of nitrogen. An example of this is the effect of nitrogen supply on the location of sulfur-deficiency symptoms. For example, in cashew seedlings receiving a luxury supply of nitrogen, symptoms of sulfur deficiency occur initially in young leaves, but in plants with a marginal nitrogen status, sulfur deficiency produce a general paleness with symptoms occurring initial in older leaves (N. Grundon, unpublished data).

Symptoms of nutrient toxicity occur more commonly in old leaves, at least partly because the elements have accumulated progressively in older leaves over a longer period of time than in younger tissues. Many toxicities result in chlorosis and necrosis of the margins of older leaves. For exampl in dicotyledons such as subterranean clover, phosphorus toxicity causes a marginal necrosis of older leaves (Rossiter 1952). Similar symptoms of marginal chlorosis and necrosis in older leaves are seen in boron toxicity in sunflower (Blamey *et al.* 1987) and in salt (sodium chloride) injury in sunflower, navybean, soybean, and cotton (Grundo 1987). In gymnosperms and mono-cotyledons, the tips of the leaves are usually the first to display symptoms of toxicity. Thus, the symptoms of phosphorus toxicity in wheat (Bhatti and Loneraga 1970), salinity effects and toxicity of sodium, chlorid and sulfate in sorghum (Grundon *et al.* 1987), and o boron toxicity in wheat and barley (Paull *et al.* 1988 Nable *et al.* 1990; Riley and Robson 1994) develop first and are more severe on the tips and distal margins of the leaves. In these instances, the locatio of the symptoms reflects the localized accumulation of the nutrients in those parts of the leaf most affected by evapotranspiration conditions. For example, increased water use by barley increased boron accumulation, with the boron being concentrated in the leaf tips where the symptoms o toxicity were the most severe (Nable *et al.* 1990).

For aluminium toxicity, seedling emergence, growth and survival are affected, resulting in variable shoot growth and stand density (Blamey *et al.* 1987). On individual shoots, the symptoms are frequently similar to those of phosphorus deficiency, reflecting an impairment of phosphorus absorption and metabolism by high concentrations of aluminium (see Robson and Pitman (1983) for an account). However, the characteristic symptoms of aluminium toxicity are expressed more clearly on the roots than on the shoots: root growth is restricted, and roots are thick and stubby with many laterals, commonly with brown tips (Blamey *et al.* 1987; Grundon *et al.* 1987; O'Sullivan *et al.* 1995).

If the toxic nutrient interacts with the metabolism of a second nutrient, thereby inducing a deficiency of the second nutrient, symptoms of the toxicity occur on older leaves while symptoms of deficiency occur on those leaves characteristic for that nutrient. For example, on acidic soils, excessive levels of soluble manganese can induce iron deficiency in some plants, thereby causing the development of manganese toxicity symptoms on older leaves and iron deficiency symptoms on younger leaves (Grundon *et al.* 1987; O'Sullivan *et al.* 1995).

CONTENTS.