SEASONAL VARIATIONS IN LEAF MINERAL CONTENT

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ABSTRACT

Fertilizer programmes for avocados in New Zealand are designed to both replace the minerals in fruit removed from the orchard and to support growth. A fertilizer programme requires leaves to be sampled at a time that best describes the tree's overall mineral content and takes into account how the levels of minerals change over a year. Healthy, mature leaves of the spring flush are commonly sampled in autumn, as this is the time when leaf mineral content has been considered to be the most stable following extrapolation from sampling times in California and South Africa. An improved knowledge of the pattern of mineral changes in the leaves over a year would help better refine fertilizer programmes. In this study leaf mineral content was monitored monthly over three years to identify periods when the leaf mineral content was most stable and to show patterns of changes in leaf mineral content. The amount of minerals in 'Hass' avocado leaves did not remain constant throughout the year nor was there a single time of year when all leaf minerals were stable. The greatest changes in concentration each year were in: nitrogen, sulphur, calcium, manganese and boron. There were periods within each year when individual minerals in the leaves was rising, falling or remaining the same as the previous month. The best time of year to sample leaves when individual minerals were most stable would be in: February/March for sulphur, phosphorus and boron; May for nitrogen, potassium and iron; and October or December for calcium. April/May was not found to be an ideal time to sample leaves for mineral content in this study. A good compromise time for taking leaf samples would be in February/March when spring flush leaves were 4 to 5 months old rather than April/May. The leaf mineral content in May did not show a clear difference between years despite the trees carrying a very light crop in 2006. In contrast, leaf minerals in February/March showed clear differences between 2006 and the years 2005 and 2007. It is suggested that changing the leaf sampling period used in New Zealand to February/March be considered.

Keywords: *leaf samples, timing, nutrients*

INTRODUCTION

'Hass' avocado yields can be irregular on New Zealand avocado orchards. Fertilizer programmes for avocados in New Zealand are designed to both replace the minerals lost due to fruit removal and to support sufficient growth of the shoots and roots such that regular cropping can occur. The amount of fertilizer required is determined by a number of factors with an important component being the results of mineral analysis of the leaves and soil. The mineral levels can then be compared to target levels in the leaves where deficiency or excess can be identified and corrected. To make a fertilizer programme that will support regular crops leaves must be sampled at a time that best reflects the trees nutrient status. In addition there needs to be a good understanding of what changes in the leaf nutrient levels may occur over the course of a year.

The uptake and utilization of mineral nutrients is reflected in the tissue nutrient concentration. Through chemical analysis of leaves useful information about the nutrient status of a tree can be determined. While the soil is the most common source of minerals, analysis of the total amounts of nutrients in the soil can partially describe what is plant available but not how much is taken up. In many cases the mineral content of leaves do not show a direct relationship with yield but analysis of leaves continues to be the best means of assessing the nutrient status of avocado trees (Lahav, 1995).

There are a number of factors that influence leaf mineral content of avocado leaves that include: leaf age, position within the tree, fruit load, cultivar, soil type and cultural practice (Lahav *et al*., 1989). The most common recommendation is to sample healthy, mature leaves of the first growth in the current year, *i.e.* leaves of the spring flush. It is easy nonetheless to sample the wrong leaves as leaf age is difficult to determine visually due to the leaves from different flushes having the same shape, size and colour. Not only is it important to sample similar leaves each time leaves are collected for mineral analysis but to have an understanding of how the minerals in leaves may change during the year. Leaf samples are usually taken in autumn as this is the time where leaf mineral content is considered to be the most stable (Lahav *et al.*, 1989).

To our knowledge there are no studies published on the changes in leaf mineral content during the year for 'Hass' avocado leaves in New Zealand. Sampling of avocado leaves for mineral analysis in New Zealand has been extrapolated from overseas recommendations, most notably from California and South Africa. Late April to late May has been selected as the mineral content of the leaves are thought to be most stable and best reflect the nutrient status of the trees (Abercrombie, 2001; Lahav and Kadman, 1980). This assumption has never been validated in New Zealand through empirical measurements of leaf mineral content throughout the year. Knowledge of the pattern of mineral changes in the leaves during the year would help both avocado growers and consultants to better refine their fertilizer programmes to meet desirable leaf target levels of nutrients. This study was conducted to identify periods when the leaf mineral content was most stable and therefore the best time to sample leaves for nutrient analysis and to show patterns of changes in leaf mineral content throughout the year.

MATERIALS AND METHODS

Leaves were collected from a commercial 'Hass' avocado orchard in the Western Bay of Plenty once a month on about the 15th (± 2-3 days) of the month by the same person for three years. A total of 120 leaves per sample, consisting of 30 leaves from each tree were taken from the same four trees each month. The leaves were collected at random, evenly spaced around each tree. The sample included leaves from both sunny and shaded areas at shoulder height (1.5-2.0m). The age of the leaves was not determined.

From February to August the sample consisted of healthy, fully mature, expanded leaves (approximately 4-10 months old) from the number 2 to 4 leaf position below the apical buds of branches that were not flushing or fruiting. During the months of September to January, when the flower inflorescences or new flush was developing, healthy, fully mature, expanded leaves from 30cm behind the flowers or flush were included in the sample to ensure the sample consisted of 120 leaves. Approximately 25% of the leaves collected in September to January were from flowering or flushing branches.

The sample was then analyzed by a commercial testing facility, Hill Laboratories, Hamilton, for mineral content. The leaves were washed with an acid detergent solution prior to drying (oven dried at 62°C overnight) and grinding (to pass through a 1.0mm screen). Nitrogen content was determined by Near Infrared Spectroscopy (NIR). Nitric acid/Hydrogen peroxide digestion followed by Inductively Coupled Plasma – Optical Emission Spectroscopy (ICP-OES) was used to determine phosphorus, potassium, sulphur, calcium, magnesium, sodium, iron, manganese, zinc, and boron content. Copper levels are not reported as fungicide sprays containing copper were applied that may have contaminated the sample.

The fertilizer applied to the trees was recorded in a diary during the three years of the trial. Fertilizer application times are presented in Table 1.

The leaf minerals were correlated with one another data and where the correlation co-efficient was significant to at least $p = 0.05$ were analyzed using linear regression analysis as either a linear function or a quadratic function by MINITAB version 13.31.

Table 1. Time when individual mineral elements were added to 'Hass' on 'Zutano' rootstock avocado trees from 2004 to 2007.

	2004					2005												2006												2007								
Mineral	**A**	**S**	**O**	**N**	**D**	**J**	**F**	**M**	**A**	**M**	**J**	**J**	**A**	**S**	**O**	**N**	**D**	**J**	**F**	**M**	**A**	**M**	**J**	**J**	**A**	**S**	**O**	**N**	**D**	**J**	**F**	**M**	**A**	**M**	**J**	**J**	**A**	**S**
Nitrogen	■	■	■		■	■	■	■		■	■	■	■	■	■		■	■	■	■		■				■	■	■		■				■			■	
Phosphorus		■	■		■	■	■	■						■	■		■	■	■	■						■	■	■		■				■				
Potassium	■	■	■		■	■	■	■		■			■	■	■		■	■	■	■		■				■	■	■		■				■				
Sulphur		■	■		■	■	■	■						■	■		■	■	■	■						■	■	■		■				■				
Calcium		■	■		■	■	■	■						■	■		■	■	■	■						■	■	■		■				■				
Magnesium	■	■	■		■	■	■	■			■	■	■	■	■		■	■	■	■						■	■	■		■				■			■	
Sodium																																						
Iron		■	■		■	■	■	■						■	■		■	■	■	■						■	■	■		■				■				
Manganese		■	■		■	■	■	■						■	■		■	■	■	■						■	■	■		■				■				
Zinc		■	■		■	■	■	■						■	■		■	■	■	■						■	■	■		■				■				
Boron	■	■	■		■	■	■	■					■	■	■		■	■	■	■						■	■	■		■				■				

RESULTS

The amount of minerals in 'Hass' avocado leaves did not remain constant throughout the year. The minerals with the greatest changes each year were: nitrogen, sulphur, calcium, manganese and boron (Table 2). The amount of minerals present in the leaves each year was within the range of values measured for the entire orchard previously in 2001 to 2004. The fertilizer programme for the trees changed in the spring of 2006 with a much reduced application of fertilizer in general (Table 1). The only boron applied from September 2006 was in the compound fertilizer used while from August 2004 to March 2005 and August 2005 to March 2006 a supplementary application each year of 450g boric acid and 150g Borax per tree was applied.

The trees used in this study were in a strong alternate bearing cycle. The cropping history of the trees was: a heavy fruit set in the spring of 2004; no fruit set in the spring of 2005; a heavy fruit set in the spring of 2006. The mineral content of leaves sampled at the commonly recommended time of late April to mid May is presented in Table 2. The year the trees were without fruit the leaves had lower sulphur, calcium, magnesium, manganese and boron in 2006 compared to the mineral content of leaves when there was large amounts of fruit in

Table 2. Average mineral content of avocado leaves collected on the 16-18 of May each year.

		Year		
Mineral	**Unit**	**2005**	**2006**	**2007**
Nitrogen	%	2.3	2.4	2.8
Phosphorus	%	0.16	0.17	0.17
Potassium	%	1.1	1.0	1.0
Sulphur	%	0.30	0.25	0.28
Calcium	%	1.87	1.26	1.49
Magnesium	%	0.36	0.31	0.35
Sodium	%	0.01	0.01	0.01
Iron	mg/kg	42	43	51
Manganese	mg/kg	160	140	270
Zinc	mg/kg	33	31	31
Boron	mg/kg	44	22	18

Table 3. Average mineral content of avocado leaves collected at about the 15th of the month for each season within the years 2005 to 2007.

		Season			
Mineral	**Unit**	**Spring**	**Summer**	**Autumn**	**Winter**
Nitrogen	%	2.3	2.3	2.5	2.4
Phosphorus	%	0.14	0.16	0.15	0.17
Potassium	%	1.0	1.1	1.0	1.1
Sulphur	%	0.26	0.26	0.27	0.28
Calcium	%	1.8	1.6	1.7	1.7
Magnesium	%	0.40	0.37	0.39	0.38
Sodium	%	0.01	0.01	0.01	0.01
Iron	mg/kg	55.9	57.6	56.1	50.1
Manganese	mg/kg	320.0	255.6	245.6	203.3
Zinc	mg/kg	31.9	34.3	34.6	35.4
Boron	mg/kg	18.7	30.9	21.9	30.2

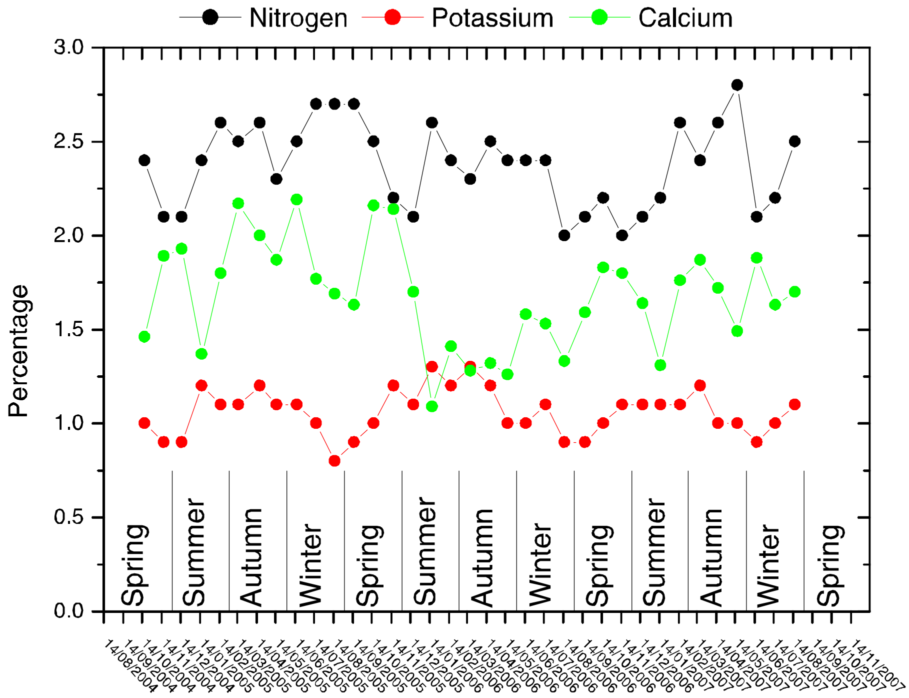

Figure 1. Percentage nitrogen, potassium and calcium of 'Hass' avocado leaves sampled monthly from spring 2004 until spring 2007.

2005 and 2007 (Table 2). The mineral content was lower despite a similar fertilizer programme in 2006 when there was no crop and 2005 when there was a very heavy crop.

Within each year the amount of the macro elements: nitrogen, phosphorus, potassium, sulphur, calcium and magnesium were similar (Table 3). The iron content of the leaves tended to be lower when sampled in winter than in the other seasons while the manganese content decreased from spring through to winter. The boron content fluctuated from a low value in spring to a higher value in summer, was lower again in autumn and then higher in winter (Table 3).

The graphs of each mineral analyzed within the leaves were examined to identify periods with in each year when the mineral content was most stable. There were periods in the year when individual minerals in the leaves was rising, falling or remaining the same as the previous month. Overall there was not a single time of year when all minerals had the same change in mineral content as one another.

Nitrogen: leaf content varied throughout the year with no well-defined stable period each year (Figure 1). In the winters of 2005 (June, July, August) and 2006 (May, June, July) nitrogen content of the leaf sample was similar. There was no stable period in 2007.

Calcium: the months of October to December were periods where the leaf calcium content rose or decreased only a small amount (Figure 1). The pronounced decrease in leaf calcium in January 2006 marks the beginning of a change in irrigation practice on the orchard.

Potassium: the amount of potassium in the leaves tended to follow a pattern of increase from the spring through to the end of autumn and decline through winter (Figure 1). The lowest amounts of potassium were found in leaves sampled in August and September with the highest amounts tending to present in late summer (February to March).

Magnesium: there was only one stable period in the summer and autumn of 2006 when there was no fruit on the trees (Figure 2).

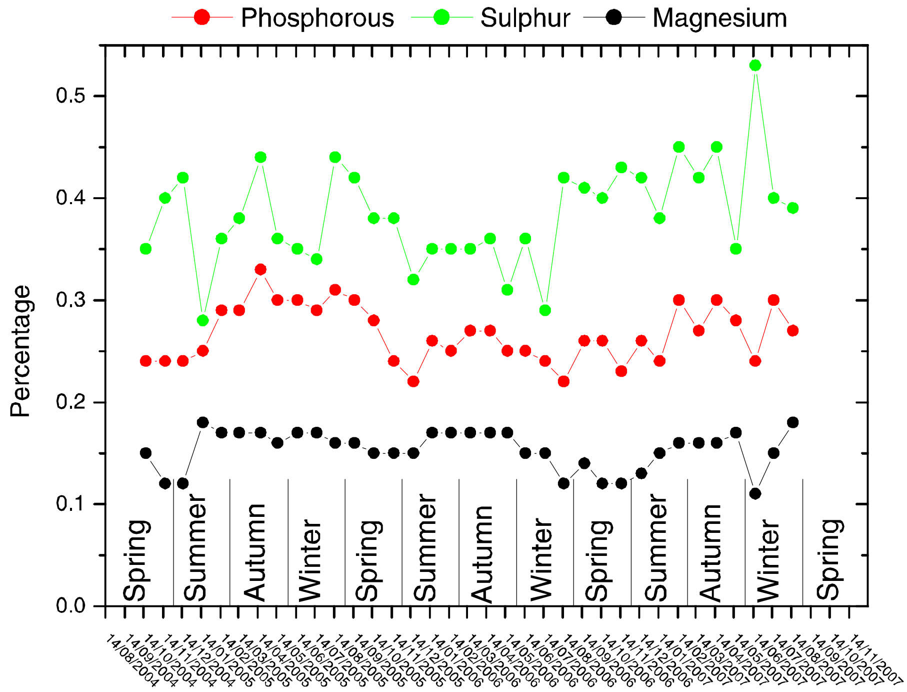

Figure 2. Percentage phosphorus, sulphur and magnesium of 'Hass' avocado leaves sampled monthly from spring 2004 until spring 2007.

Sulphur: there were periods of stable sulphur levels in the leaves sampled in the months February to June and March to June in 2005 and 2006, respectively (Figure 2). There was no similar stable period in 2007.

Phosphorus: in general stable leaf levels were found from February to April each year (Figure 2).

Iron: there were two peaks each year in the amount of iron in the leaves in the summer/autumn and

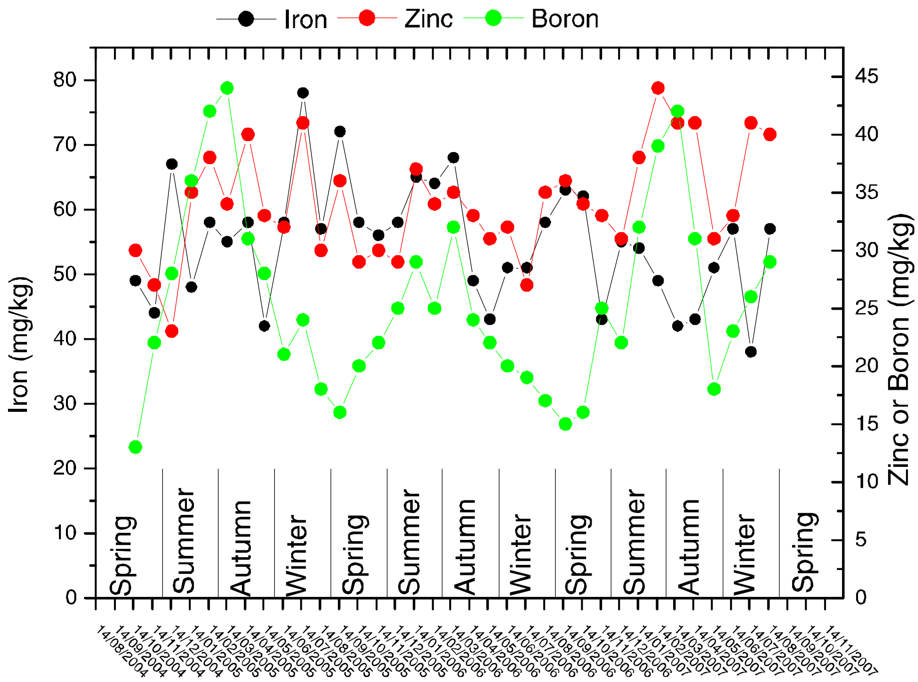

Figure 3. Concentration of iron, zinc and boron in 'Hass' avocado leaves sampled monthly from spring 2004 to spring 2007.

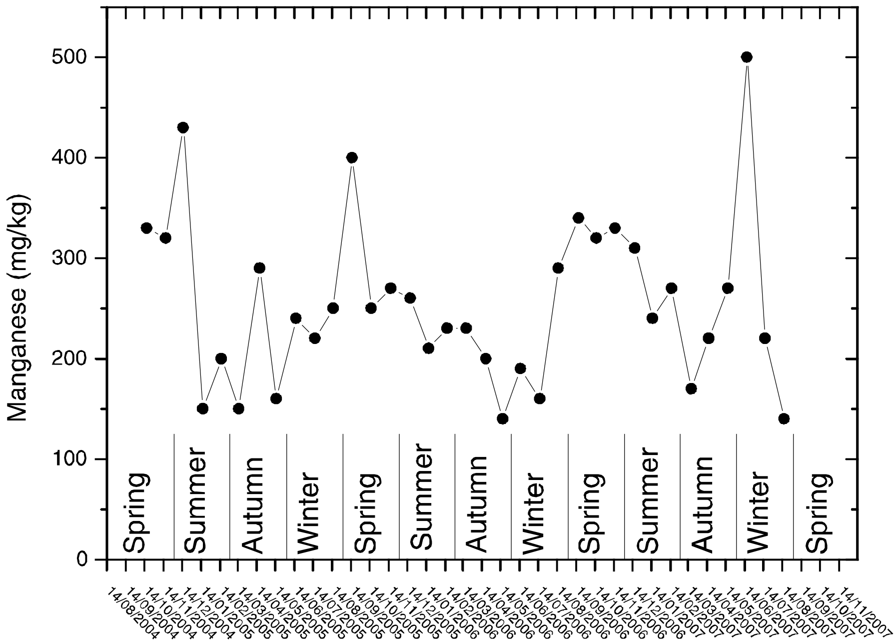

Figure 4. Concentration of manganese in 'Hass' avocado leaves sampled monthly from spring 2004 to spring 2007.

winter/spring (Figure 3). Leaves sampled in the months of April and May had the lowest iron content each year.

Zinc: tended to follow the same pattern of increase and decrease as iron in the years 2005 and 2006, but not in 2007 (Figure 3).

Boron: there was no stable period but a clear pattern of boron increasing to reach a maximum value in February/March each year and the lowest value in September/October harvested leaves (Figure 3). The lowest amount of boron in the leaves each year was around 15 mg/kg with the greatest amount of boron was about 45 mg/kg in years when the trees were carrying a heavy crop. In the years 2005 and 2006 the amount of boron measured in the leaf sample in April/May was about twice the leaf boron content of leaves sampled at the lowest point in September/October.

Manganese: leaf content varied each month with an increase in the leaf that generally was associated with addition of fertilizer (Figure 4 and Table 1).

The leaf nitrogen content of leaves was positively related to the leaf phosphorus sulphur and zinc (Figure 5). Leaf nitrogen content had a quadratic relationship to manganese content with minimum manganese content at about 2.4% leaf nitrogen (Figure 5).

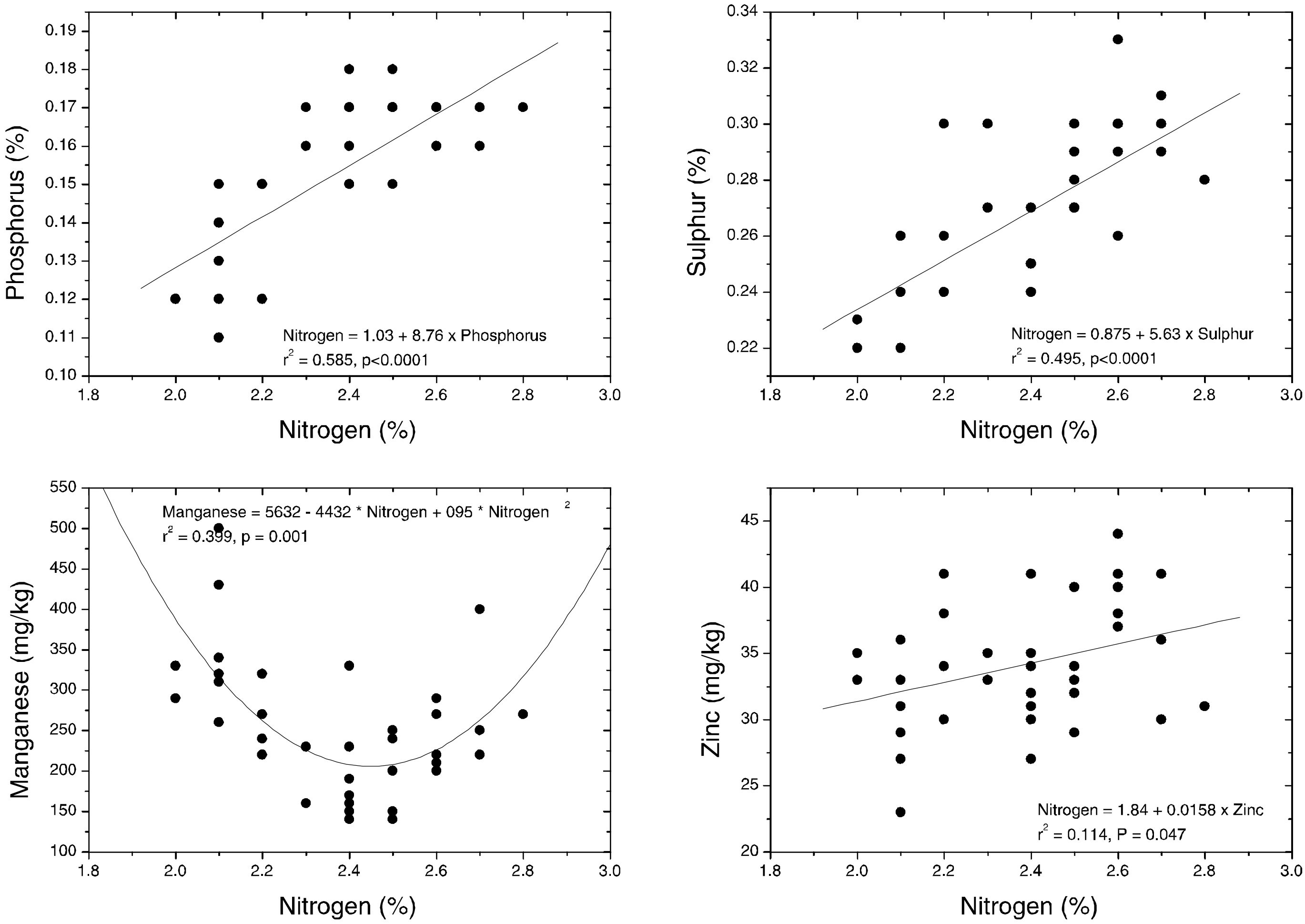

Figure 5. Relationships between leaf nitrogen content and leaf phosphorus, sulphur, manganese and zinc content.

Leaf phosphorus content was positively related to leaf potassium, sulphur, zinc and boron and negatively related to leaf magnesium and manganese content (Figure 6).

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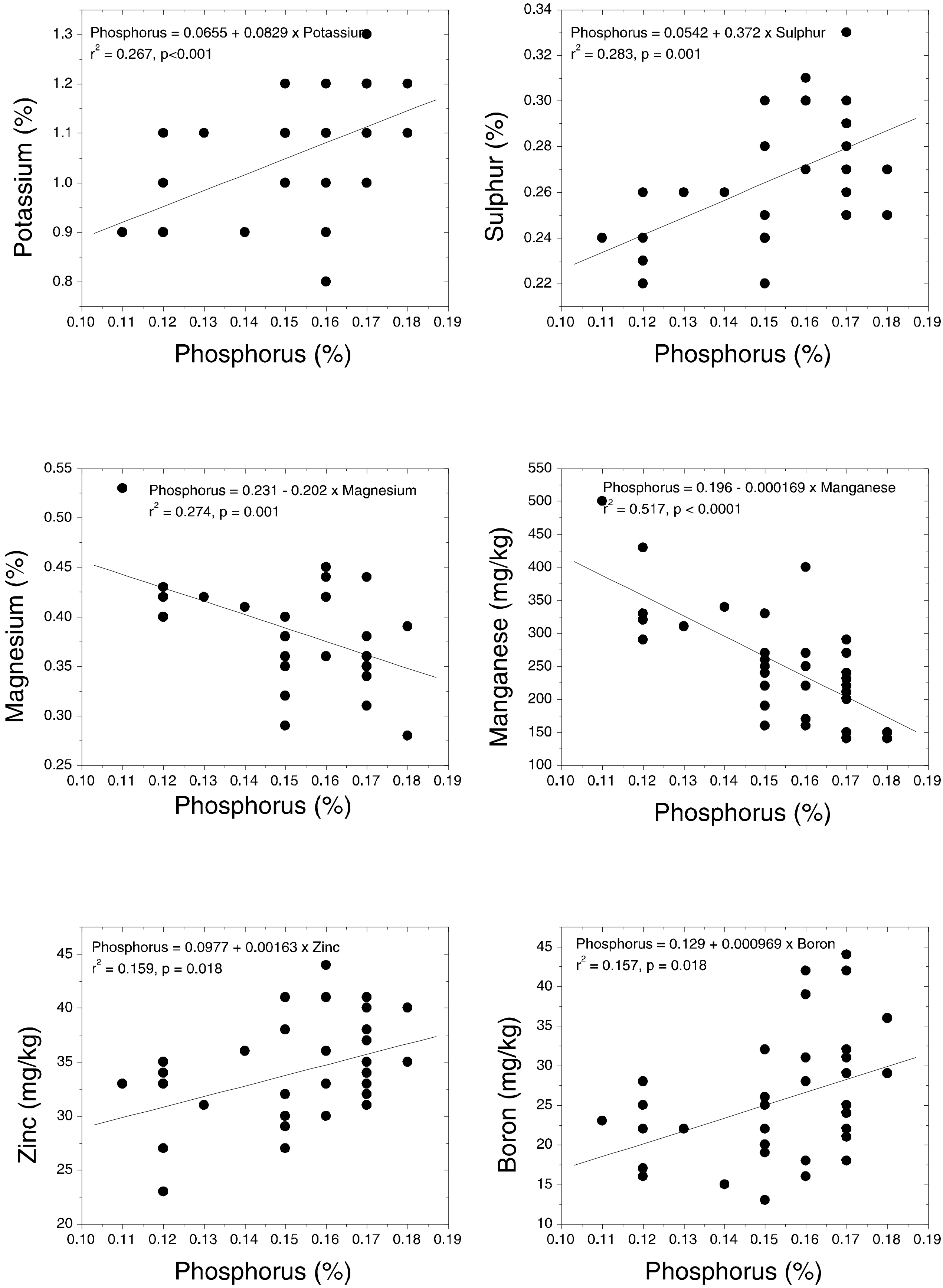

Figure 6. Relationship between leaf phosphorus content and leaf potassium, sulphur, magnesium, manganese, zinc and boron.

Leaf potassium content was positively related to leaf boron content but negatively related to leaf magnesium and manganese content (Figure 7). Leaf sulphur content was positively related to leaf zinc content (Figure 7).

Leaf magnesium content was positively related to leaf calcium and manganese content (Figure 8). Leaf boron content was positively related to leaf zinc content but was negatively related to leaf manganese content (Figure 8).

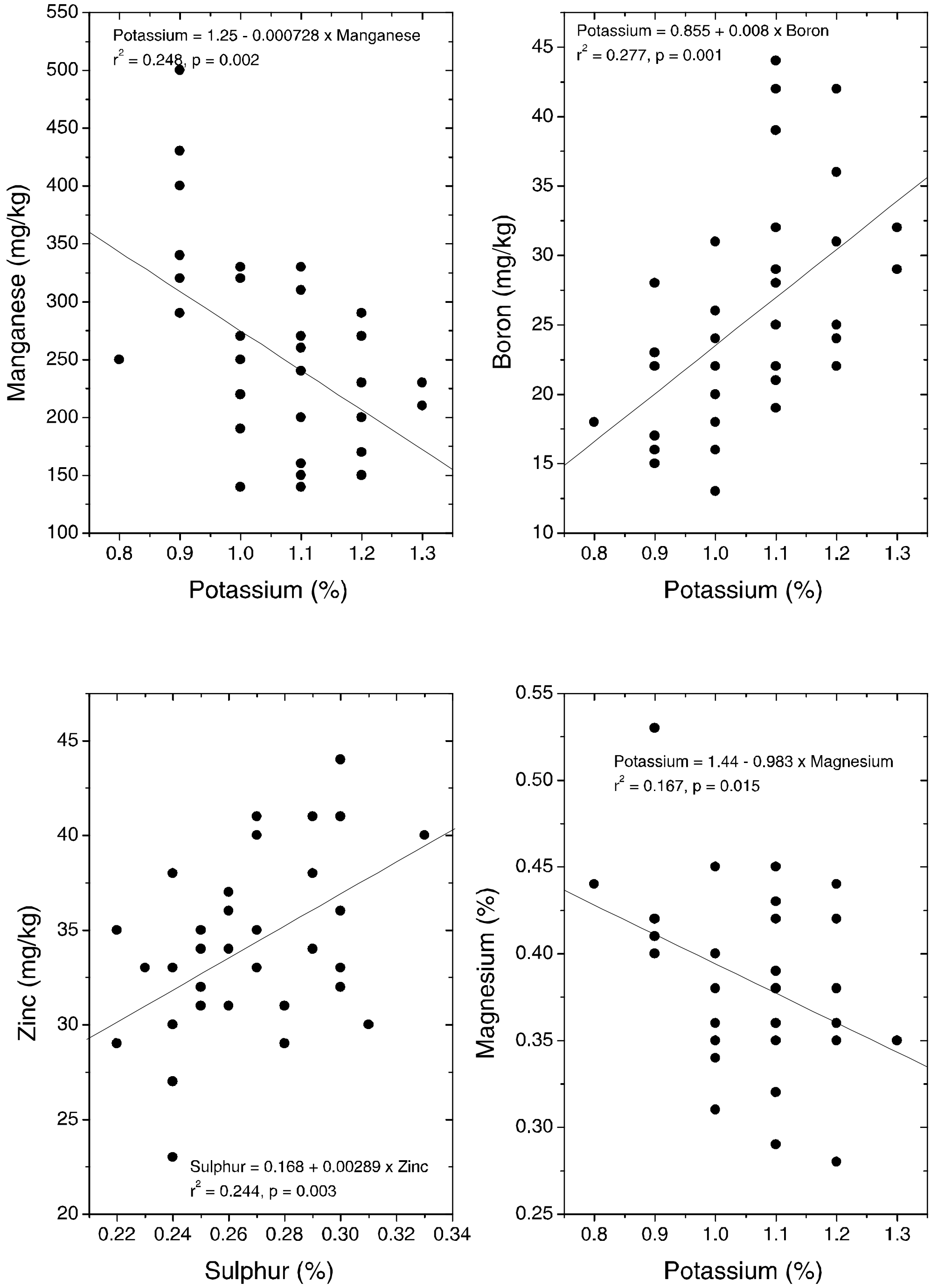

Figure 7. Relationship between leaf potassium content and leaf magnesium, manganese and boron and leaf sulphur to leaf zinc.

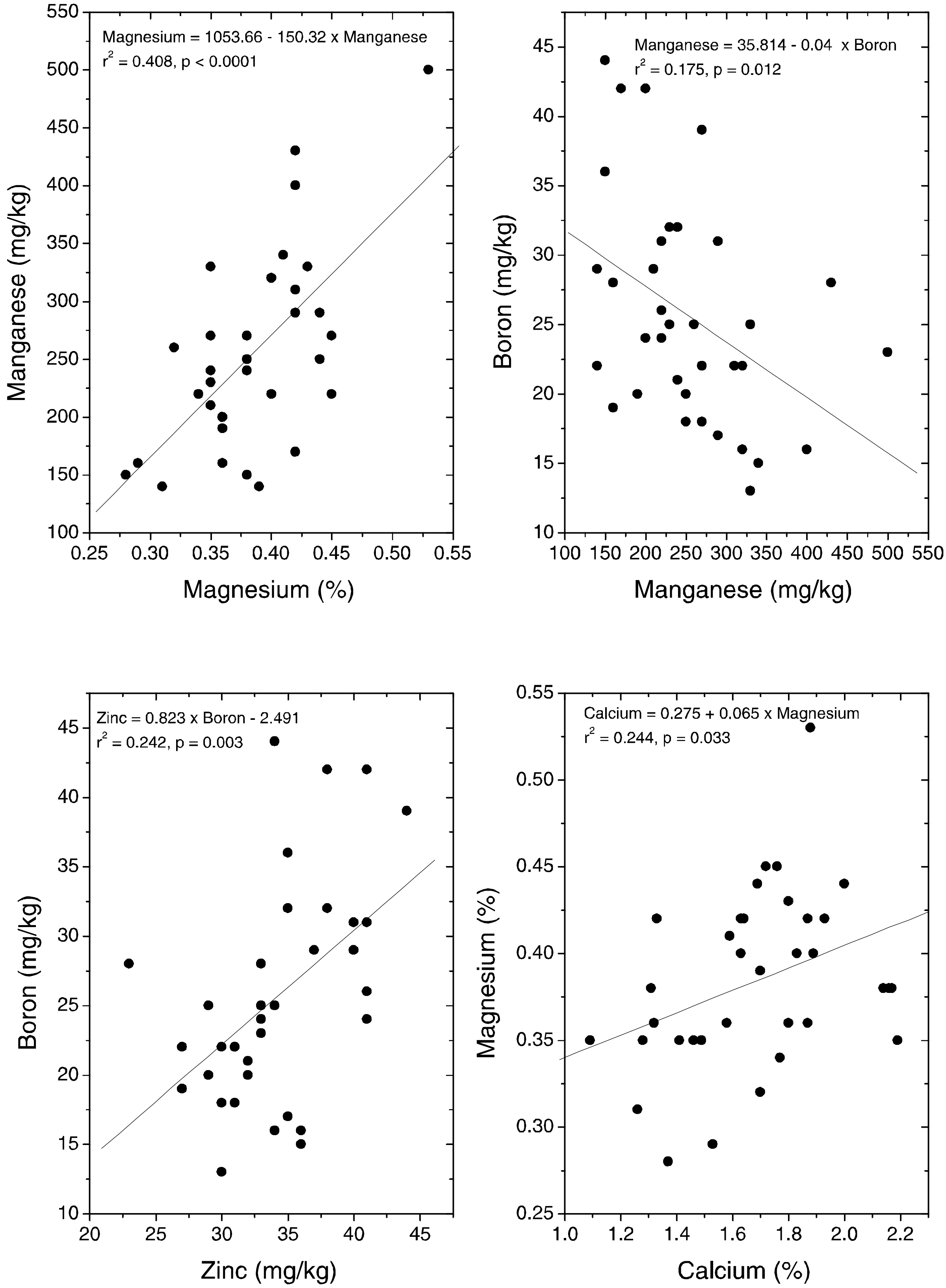

Figure 8. Relationships between leaf manganese content and leaf magnesium and boron content. Relationship between leaf zinc content and leaf boron content. Relationship between leaf calcium and magnesium content.

DISCUSSION

The results of this study indicate that there was not a single time of the year where the content of all the minerals in the leaf could be considered to be stable over the three years the leaves were sampled. In New Zealand, avocado leaves are recommended to be sampled in late April to early May when the spring flush leaves are about 5 to 7 months old (Cutting and Dixon, 2004). The time of year when leaves are collected is similar to the recommendation for avocado trees in South Africa (Abercrombie, 2001) and Israel (Lahav and Kadman, 1980). In Australia the time of leaf collection is similar to New Zealand but the leaves collected are recommended to come from the summer flush at about 8 weeks old (Vock, 2001). The leaf sampling times have been selected to give the most accurate representation of the mineral content of the tree where the mineral levels are the most stable. Based on an assessment of times of the year when the leaf mineral content could be considered most stable the results in this study would indicate that leaf sampling times for each mineral would be different. The minerals sulphur, phosphorus and boron could be measured in February/March; nitrogen, potassium and iron in May; calcium in October or December; zinc, manganese and magnesium have no stable periods. These findings are similar to those reported in California where the measurement of most macronutrients in Hass avocado leaves was proposed to be in August to December (January to February in New Zealand) although the calcium and micronutrient levels would not be at stable levels (Bingham, 1961). This contrasts with a study on 'Tonnage' avocados in Florida where the leaf mineral content was considered to be stable at the beginning of September to October (March to April in New Zealand) (Koo and Young, 1977). In Israel 'Hass' and 'Fuerte' avocados spring flush leaves have been reported to have the most stable leaf mineral content in autumn (Lahav *et al*, 1989; Lahav, 1995).

The results of this study would suggest that April/May may not have been the ideal time to sample leaves for mineral content as April/May was not when the minerals were at stable levels or that the minerals were in the mid-cycle of a well defined pattern of change. The ideal time of year to sample leaves for mineral content depends on which minerals are considered to be of most importance. The concentration of the following minerals: nitrogen, phosphorus, potassium, calcium, magnesium, zinc and boron are considered to be the most important under New Zealand conditions (Cutting and Dixon, 2004). The leaf mineral contents presented in Figures 1 to 3 imply that a good compromise time for taking leaf samples would be in February/March rather than April/May. Leaf samples taken in February/March would be during the most consistently stable period over three years for leaf phosphorus and the high point for boron and zinc content. The leaf nitrogen content in February/March was close to the May levels while the leaf potassium and magnesium content was relatively stable compared to other times of the year. Only leaf calcium levels were variable and may not represent well values in the leaves sampled in February/March. Sampling in February/March also has the advantage that only spring flush leaves need be collected and that the leaves are about 4-5 months old and likely to have their highest leaf mineral content (Lahav and Whiley, 2002).

The differences in leaf mineral content from month to month appear to also show the difference in mineral content due to the leaves getting older (Lahav and Whiley, 2002) as well as possible reallocation of minerals to other parts of the plant (Cameron *et al.*, 1952; Lahav and Whiley, 2002). The leaves sampled from September to January had a proportion of leaves that were from flowering or flushing branches. The mixture of leaves did not appear to greatly change the amount of minerals in the leaf or the pattern of change in leaf mineral content. The youngest leaves were those sampled in February/March as these leaves were taken from the newly developed spring flush. Leaf Boron levels were highest in February sampled leaves in agreement with previous research (Labanauskas *et al.*, 1961; Robbertse and Coetzer, 1992).

Increases in leaf mineral content did not necessarily follow the application of fertilizer (compare Table 1 with Figures 1 to 4). Leaf manganese content appeared to be closely related to when manganese containing fertilizer was applied but leaf nitrogen levels did not appear to respond as readily to nitrogen application unlike that reported by Bar *et al.* (1987) in Israel. Having an understanding of how leaf age affects leaf mineral content is helpful when designing fertilizer programmes to know how much the concentration of a mineral may change outside of the time when the leaves were sampled. The mineral changes can also be related to phenological events. For example, the leaf boron content in February 2005 and 2006 was about 40 mg/kg which were "on" crop years but only reached a maximum of about 32 mg/kg in an "off" crop year but the leaf boron content declined to similar low points of about 15 mg/kg each year.

At present, the most common time for taking leaf samples for mineral content is in late April to late May (Cutting and Dixon, 2004). The results of this study indicate that leaves collected in February or March rather than April/May each year would be a more appropriate for establishing the mineral content of Hass avocado trees in the Western Bay of Plenty, New Zealand. The leaf mineral content in May (Table 2) does not show a large difference between the years despite the trees in 2006 carrying a very light crop. In contrast, the leaf mineral content in February or March show low values in 2006 compared to values in 2005 and 2007 (Tables 4 and 5). The reason for lower leaf mineral content is not known but may have been due to reduced mineral uptake due to a lack of fruit or to dilution of minerals throughout the tree due to increased vegetative growth. The amounts of minerals in February and March 2005 and 2007 more closely match the leaf target levels in the Growers' Manual (Cutting and Dixon, 2004) than do the leaf minerals in 2006. The difference in leaf mineral content between years correlates well to the difference in the amount of fruit the trees were carrying in 2005 and 2007. Such findings indicate that the time at which the leaves are sampled for mineral analysis should be reviewed.

There appeared to be a number of minerals interacting with one another were when the concentration of one mineral increased several other minerals concentration was also increased (Figures 5 to 8). The minerals magnesium and manganese decreased when phosphorus and potassium were increased (Figures 6 and 7).

Table 4. Average mineral content of avocado leaves collected on the 16-17 of February each year.

		Year		
Mineral	**Unit**	**2005**	**2006**	**2007**
Nitrogen	%	2.6	2.4	2.6
Phosphorus	%	0.17	0.17	0.16
Potassium	%	1.1	1.2	1.1
Sulphur	%	0.29	0.25	0.30
Calcium	%	1.80	1.41	1.76
Magnesium	%	0.36	0.35	0.45
Sodium	%	0.01	0.01	0.01
Iron	mg/kg	58	64	49
Manganese	mg/kg	200	230	270
Zinc	mg/kg	38	34	44
Boron	mg/kg	42	25	39

Table 5. Average mineral content of avocado leaves collected on the 16-20 of March each year.

		Year		
Mineral	**Unit**	**2005**	**2006**	**2007**
Nitrogen	%	2.5	2.3	2.4
Phosphorus	%	0.17	0.17	0.16
Potassium	%	1.1	1.3	1.2
Sulphur	%	0.29	0.27	0.27
Calcium	%	2.17	1.28	1.87
Magnesium	%	0.38	0.35	0.42
Sodium	%	0.01	0.01	0.01
Iron	mg/kg	55	68	42
Manganese	mg/kg	150	230	170
Zinc	mg/kg	34	35	41
Boron	mg/kg	44	32	42

Manganese had a quadratic relationship with leaf nitrogen content with the least manganese at a leaf nitrogen content of about 2.4% (Figure 5). Most of the relationships of one mineral to another while statistically significant were weak (regression coefficients < 0.3). The strongest relationships (regression coefficients > 0.4) were between leaf nitrogen and phosphorus, sulphur and manganese. Leaf manganese was also related to leaf phosphorus and magnesium. These interactions should be taken into account when interpreting the results of a leaf mineral test and in any fertilizer programmes. Based on the results presented here the leaf content of magnesium and manganese are likely to be difficult to maintain when addition of fertilizer is seeking to increase the leaf content of other minerals.

Changing the time when leaves are sampled for mineral analysis from late April/late May to February/March could alter how a leaf mineral test is interpreted and what fertilizer programme may be recommended. For example for some minerals February/March leaf values would represent their highest values making it important that the values obtained reached target levels. This study did not examine target leaf levels of minerals but the timing of leaf sampling. To determine if leaf mineral target levels need to be altered further research would be required. The time when leaf samples are collected also needs to be considered in the context the trees phenological cycle. Important events such as the irreversible commitment to flowering, spring flush initiation and root flush will need to be considered along with the patterns of change in leaf mineral content over the year. By looking at all the factors associated with the leaf mineral content it should be possible to refine when fertilizer is applied in the orchard currently.

CONCLUSIONS

Monthly monitoring of leaf mineral content shows that the most stable and peak period, overall, for leaf minerals was in February and March rather than late April/late May each year. The leaf mineral content in February or March appeared to describe well the difference in fruit load on the trees in the study. There were a number of interactions between different minerals identified with leaf magnesium and manganese content being decreased when other minerals are increased in the leaf. It is suggested that changing the leaf sampling period used in New Zealand to February/March be considered.

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