Possible causes and measures to prevent excessive leaf abscission in the avocado (*Persea americana* Mill.) cultivar Ryan

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ABSTRACT

Excessive leaf abscission occurs in the avocado cultivar Ryan during flowering. Severity of leaf abscission varies between areas and orchards and could be influenced by stress factors, such as drought, climate and nutrient deficiencies. Natural aging might also play a role in excessive leaf abscission. The possible causes of excessive leaf abscission were investigated, and chemicals applied for its prevention. The largest percentage of leaves dropped from flowering time until two weeks after flowering, although leaf drop was not statistically significant for the two 'Ryan' blocks under investigation. Macro- and micro-element leaf analysis of 'Ryan', 'Fuerte' and 'Hass' trees showed a significant decrease in leaf N and P content for 'Ryan' over the monitoring time (six weeks before flowering to six weeks after flowering) to values lower than the recommended norms. Iron levels increased markedly at flowering after which it again declined to the previous levels, which might be an indication of earlier water stress. Further leaf samples were taken for 'Ryan' at five different stages of discoloration up to leaf abscission and again a significant decrease in N and P content was observed over the different stages. Nitrogen and P levels, as well as irrigation may therefore play a role in the leaf abscission problem of 'Ryan'. Bark starch content of 'Ryan' trees decreased significantly after flowering, as found by measuring on a monthly basis from June to October 2005. The early morning determinations of RWC of 'Ryan', 'Fuerte' and 'Hass' showed no significant difference in trend for 'Ryan'. The midday stem water potential and midday RWC of 'Ryan' showed no obvious deviation in trend over six different stages from flower bud break to fruit set. No definite leaf abscission layer was observed in any of the different stages of 'Ryan' leaf discoloration as determined by light microscopy. Scanning electron microscopy (SEM) studies of 'Ryan' and 'Fuerte' leaf samples of the different coloration stages showed that immature leaves had a thinner cuticle as compared to the other leaf stages. In an attempt to reduce leaf abscission, the following chemical applications were made to 'Ryan' trees: 2 kg/tree dolomitic lime, 50 g/tree solubor and a combination thereof, as well as 30 mg/ℓ NAA, 10 mg/ℓ BA, 5 ml/ℓ Kelpak®, 20 mg/ℓ sodium nitroprusside and 30 g/ℓ (3%) kaolin, at the stage of flower bud break. Leaf abscission and fruit set were recorded. None of the chemical treatments reduced leaf abscission. Chemical treatments also had a negative effect on fruit set.

Abbreviations: *NAA = Naphthalene acetic acid, BA = 6-benzylamino purine, RWC = Relative water content.*

INTRODUCTION

Trees of the avocado cultivar Ryan is known for their excessive leaf abscission in early spring when the inflorescences emerge. Apparently leaf drop occurs mostly from the previous spring and summer flush and it is thus the over-wintered leaves which are abscised. No scientific study could be found in which these observations were investigated in detail.

The efficiency of over-wintered leaves is of high importance for the production of photoassimilates, which are needed for flowering, fruit set and early fruit development. In addition to photoassimilates produced by over-wintered leaves, stored carbohydrates are also important to fuel fruit set and early fruit development (Wolstenholme, 2001). The spring flush that emerges at the time of fruit set may take up to 40 days before the leaves become net exporters of carbohydrates thereby competing with developing fruit for food reserves and photoassimilates (Whiley & Schaffer, 1994). Consequently the developing spring flush increases the demand for food reserves and photoassimilates, and fruit set and production may thus be negatively affected when the abscission of photosynthetically active leaves is high and stem reserve levels are low.

Leaf abscission in avocado and other plants in general can be caused by various stress factors (Aharoni, 1978; Prince & Cunningham, 1988; Nitzsche *et al*., 1991; Whiley & Schaffer, 1994; Temple & Riechers, 1995). Stress factors include water, nutrient and climatic stress factors. It is, however, not known what percentage of 'Ryan' leaf drop is the result of the natural aging of leaves.

Abscission is a process that occurs naturally in all plant species and is genetically programmed into different plant organs and tissues of individual plant species (Salisbury & Ross, 1992). Leaf drop in avocado naturally occurs during spring time, which is the time when flowering and fruit set occur. Avocado leaves are thus short-lived (9 to 12 months), as expected from the abscission pattern (Whiley & Schaffer, 1994; Wolstenholme, 2001).

Plant growth regulators (PGR's) can be used to delay leaf abscission. Leaf abscission of avocado cuttings was delayed by applying a combination of the synthetic auxin, NAA, and the synthetic cytokinin, BA (Raviv & Reuveni, 1984). NAA also delayed abscission of the cotyledons of 'Red Kidney' beans (Abeles & Rubinstein, 1964) and improved fruit retention in litchi (Kift *et al.*, 2002). However, the time of application and the concentration of synthetic PGR's to be applied is crucial in getting the desired effect (Abeles & Rubinstein, 1964; Sachs & Hackett, 1972; Kitsaki *et al.*, 1999; Griffin & Schroeder, 2004). It is therefore important to determine the stage and optimum concentration for application of PGR's for the avocado cultivar 'Ryan' to improve leaf retention. However, the serverity of abscission varies with area and orchard and it would thus appear that the occurrence of stress factors may play a major role in leaf abscission.

Climatic stress factors that might promote leaf abscission in avocado is a lowering in temperature in relation to irradiance during the winter. A lowering in temperature during the onset of winter may cause a drop in the photosynthetic rate because of photo-inhibition. Photo-inhibition is mainly caused by an excess of light that cannot be used under cold conditions. Photo-inhibition leads to chlorophyll damage and destruction with the consequent abscission of leaves (Wolstenholme, 2001). Reflective substances were used in a number of crops to reduce solar radiation and minimize damage caused by solar radiation. The reflective substance, Kaolin, was used especially to minimize solar injury on apples by reflecting solar radiation (Glenn *et al.*, 2002; Shupp *et al.*, 2002; Gindaba & Wand, 2005). Kaolin also reduced leaf temperature by the reflection of heat (Glenn *et al.*, 2001). Kaolin application might therefore be effective in reducing low temperature – high irradiance stress, with subsequent improvement in leaf retention.

It is well known that drought leads to leaf abscission (Munne-Bosch & Alegre, 2004). Water deficits also have a negative effect on a range of physiological processes in plants, which include growth, photosynthesis, protein synthesis, solute accumulation, leaf anatomy and gas exchange, and negatively affected avocado trees (Chartzoulakis *et al.*, 2002). During the development of water stress various physiological changes occur in plants, including changes in hormone and amino acid levels (Evans, 1984; Rabe, 1990). Hormonal changes include an increase in abscisic acid and ethylene levels as well as a decrease in cytokinin levels (Evans, 1984; Morgan, 1984; Brown, 1997). Reactions due to these changes in hormone levels include stomatal closure and in more severe cases leaf abscission (Morgan, 1984). A change in the irrigation scheduling to prevent a water stress situation could therefore reduce leaf abscission.

Nutrient stress might also cause leaf abscission. Reproductive organs (e.g. flowers and fruit) compete with leaves for nutrients (Salisbury & Ross, 1992) and heavy flowering and low nutrient levels may cause an insufficient movement of nutrients to the leaves. In addition, mobile elements, such as nitrogen, phosphorus and potassium, are transported out of the leaves to developing organs (Mengel & Kirkby, 1978) and the demand will be more for these moveable elements under nutrient stress conditions. Nutrient stressed leaves also show similar physiological changes than leaves subjected to other forms of stress, such as drought (Rabe, 1990), and leaf abscission may therefore occur under nutrient stress situations. It is therefore important to keep leaf nutrient levels at the recommended levels. Avocado leaf nitrogen levels drop during the winter (Wolstenholme, 2001) and nitrogen application during the summer compensates for nitrogen loss during the winter (Whiley & Schaffer, 1994).

As the main factors that can cause leaf abscission in the avocado cultivar Ryan are unknown, the aim of this study was to identify them and to determine whether leaf abscission can be prevented by the application of chemicals without affecting fruit set negatively.

MATERIALS AND METHODS

The trial was conducted on the farm of Mr Leon Weirich in the Hazyview (Kiepersol) area of Mpumalanga where the cultivars Ryan, Hass and Fuerte were situated in adjacent blocks. For the cultivar Ryan two blocks were selected: one that was reported to have high leaf abscission and another that was reported to have low leaf abscission. For all four blocks, the irrigation scheduling was the same. The following experiments were conducted:

Determination of the time of leaf abscission

In each of the two 'Ryan' blocks eight trees were randomly selected. On each tree eight branches (four on the eastern and four on the western side of the tree) were randomly selected for measurements. Leaf counts were done on a weekly basis from six weeks before flowering (bud swell stage) to six weeks after flowering (after fruit set).

The relationship between tree nutrient status and leaf abscission

For each of the cultivars Ryan, Fuerte and Hass eight trees were again randomly selected in each block. Leaf samples were taken at three week intervals from six weeks before flowering till six weeks after flowering for each tree. The first full grown leaf, taken from the back of the branch, on the latest or summer flush of each branch was selected for sampling. A sample consisted of 20 leaves. Leaves were analyzed for N, P, K, Ca, Mg, Zn, Cu, Mn, Fe and B and the N:K-ratio at the ARC-ITSC.

Determination of nutrient loss by old leaves and nutrient status of flowers

Leaves of 'Ryan' trees were sampled at five different stages: 1) green hardened-off leaves, 2) slightly yellow coloration (<50%), 3) predominately yellow coloration (more than 50% but less than 100%), 4) completely (100%) yellow coloration, and 5) completely yellow coloration with browning of the veins. Flower samples were also taken for all cultivars at full bloom. The flowers were analysed for macro- and micronutrients as above.

Effect of reserve food resources on leaf abscission

Eight trees in each of the two 'Ryan' blocks were randomly selected and bark discs of four scaffold branches per tree were taken for starch analysis by using a 25 mm bell punch. The bark discs were oven-dried for 72 hours at 60°C and ground through a 0.5 mm sieve. Starch was then determined using the modified enzyme-chromogen method of Davie (1997).

Effect of plant water status on leaf abscission

For each of the three cultivars, Ryan, Fuerte and Hass, five trees were randomly selected. Leaf samples (consisting of 15 leaves per tree) were taken in the mornings around 08:30 and at weekly intervals from six weeks before flowering untill six weeks after flowering for the latest summer flush. The fresh mass of the individual leaves were obtained whereafter they were allowed to absorb water at low light intensity for three hours in a water bath (Hall *et al.*, 1993). The saturated or turgid mass was determined and the leaves were then oven-dried at 60°C for 72 hours, to determine the dry mass. From the fresh mass, turgid mass and dry mass the relative water content of the leaves was calculated with the equation:

$$RWC = (W_f - W_d)/(W_t - W_d) \text{ X } 100\%$$

where W_f is the fresh weight after sampling, W_t the turgid weight after three hours in the water bath and W_d the dry weight

(Hall *et al.*, 1993).

Another five trees of the cultivar Ryan were selected randomly and the RWC was determined at midday (11:00 to 12:00) for the six different bud stages. In addition, midday stem water potential was measured with a pressure bomb and soil moisture was monitored at 300 and 600 mm depth with tensiometers. The six different bud stages were: 1) Dormant bud stage, 2) Developing inflorescence, 3) Extended inflorescence, 4) Full bloom, 5) Flower petal drop, and 6) Fruit set.

Anatomical work

Leaf samples were taken at the same stages as for the nutrient study to investigate the development of the abscission layer in the leaf petiole by means of a light microscope.

Similarly, leaf samples of the same stages were also taken to investigate the development of the cuticle on the leaf surface, but in addition samples of immature leaves and leaves from the cultivar Fuerte were studied by scanning electron microscopy (SEM).

Chemical applications to retard excessive leaf abscission

In each of the two 'Ryan' blocks, eight trees for each treatment were selected in a complete random design. Applications of fertilizers were made during the middle of July 2005, and plant growth regulators as well as the reflective substance, kaolin, were applied at the end of July 2005 (**Table 1**) when the tree buds just started to swell. Leaf abscission and fruit set were recorded.

Table 1. Fertilizers and chemicals used to prevent excessive leaf abscission for the avocado cultivar Ryan.

Chemical applied	Concentration
Control	No application
Dolomitic lime	2 kg/tree
Solubor	50 g/tree
Dolomitic lime & Solubor in combination	2 kg/tree & 50 g/tree
NAA	30 mg/ℓ
BA	10 mg/ℓ
*Kelpak®	5 ml/ℓ
Sodium nitroprusside	20 mg/ℓ
Kaolin	30 g/ℓ (3%)

**Extract from seaweed containing 0.031 mg/ℓ cytokinins and 11 mg/ℓ auxins*

RESULTS AND DISCUSSION

Determination of the time of leaf abscission

The leaf abscission pattern is given as the cumulative percentage of leaves that dropped from six weeks before flowering till six weeks after flowering (**Figure 1**). Leaf abscission started from two weeks before flowering till approximately three weeks after flowering, which was the stage of fruit set. The two 'Ryan' blocks did not differ significantly and overall leaf abscission for both blocks was not found to be significant.

Effect of tree nutrient status on leaf abscission

Leaf nitrogen levels decreased significantly

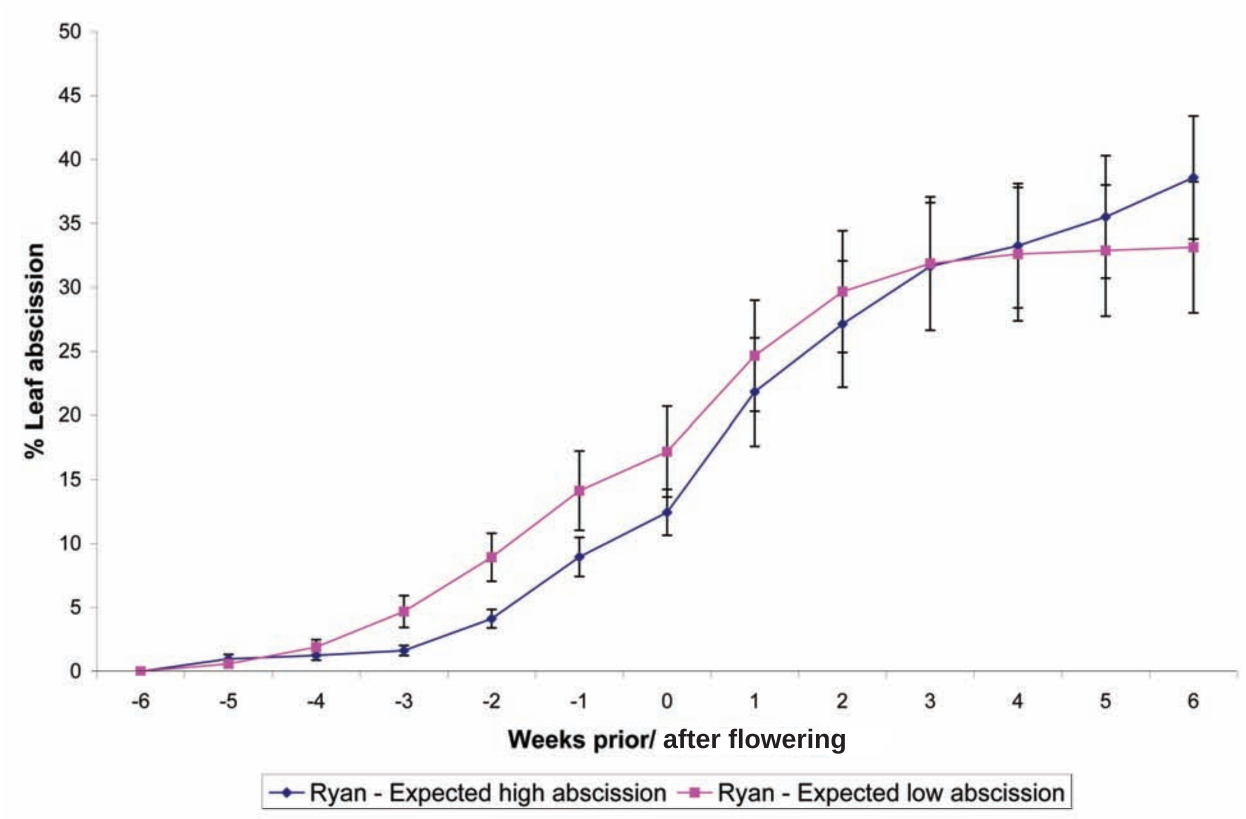

Figure 1. Leaf abscission pattern for the avocado cultivar Ryan.

for all cultivars, except 'Fuerte' (**Figure 2**). Leaf nitrogen levels were also significantly lower for both 'Ryan' blocks when compared with 'Hass'. Further, leaf nitrogen levels were lower than the norm of 2.0-2.3%. Leaf phosphorus levels also decreased significantly prior to flowering (**Figure 3**) and these values were also much lower than for the norm of 0.17-0.25%. Leaf potassium, calcium and magnesium levels did not change significantly during the monitoring period (**Figure 4**, **5** & **6**). The levels of potassium, calcium and magnesium were also within the recommended range.

Leaf zinc levels (**Figure 7**), leaf manganese levels (**Figure 8**) and leaf boron levels (**Figure 9**) did not show any significant

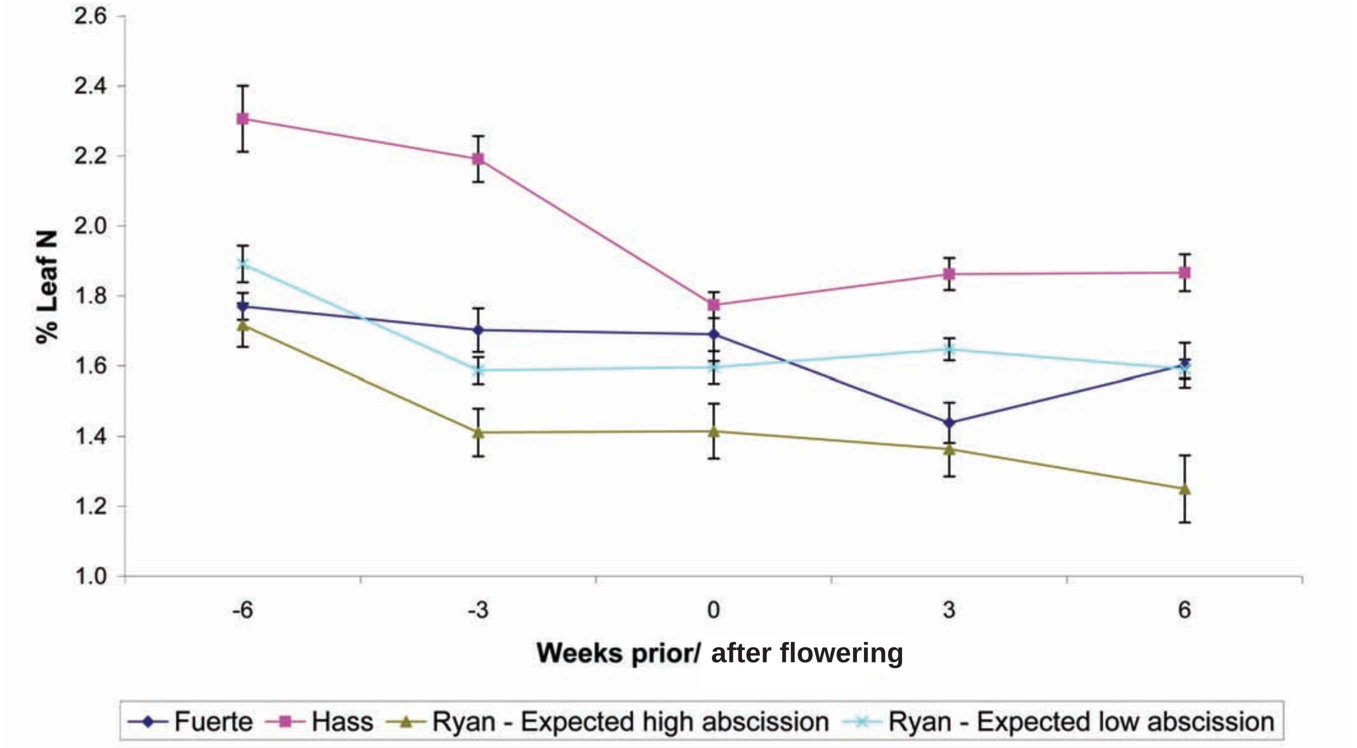

Figure 2. Comparison of leaf nitrogen levels for three avocado cultivars.

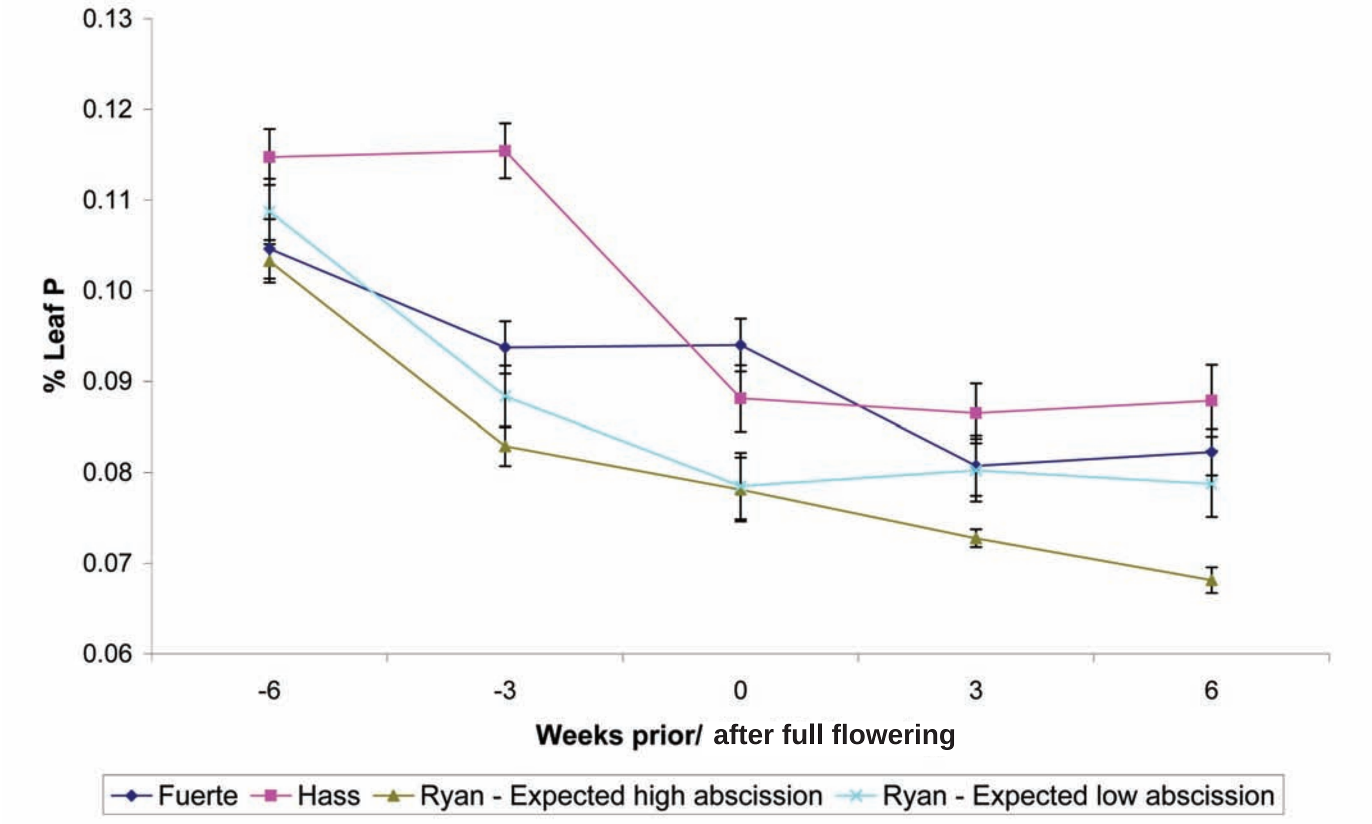

Figure 3. Comparison of leaf phosphorus levels for three avocado cultivars.

change or trend during the monitoring period. Manganese levels were higher for both 'Ryan' blocks when compared with 'Hass' and 'Fuerte'. Leaf manganese (**Figure 8**) and leaf copper (**Figure 10**) levels were also extremely high and far more than the recommended levels. Leaf zinc levels were slightly lower than the norms, while iron levels were normal. Boron levels were higher in the 'Ryan' block that was expected to have high leaf abscission. The levels of these micro-nutrients obtained were therefore mostly outside the recommended levels for all the cultivars. Leaf copper levels (**Figure 10**) and leaf iron levels (**Figure 11**) in 'Ryan' showed a significant increase at flowering, followed by a significant decrease to levels similar to the

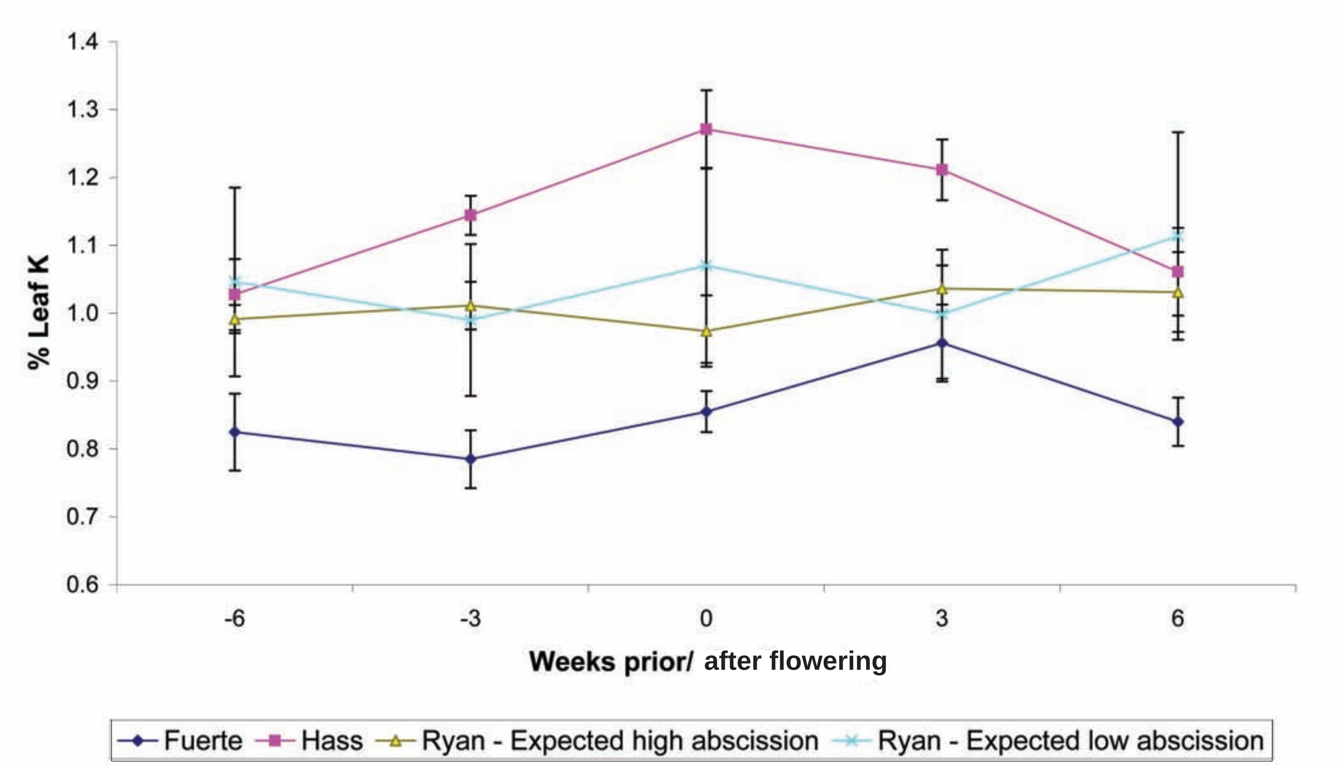

Figure 4. Comparison of leaf potassium levels for three avocado cultivars.

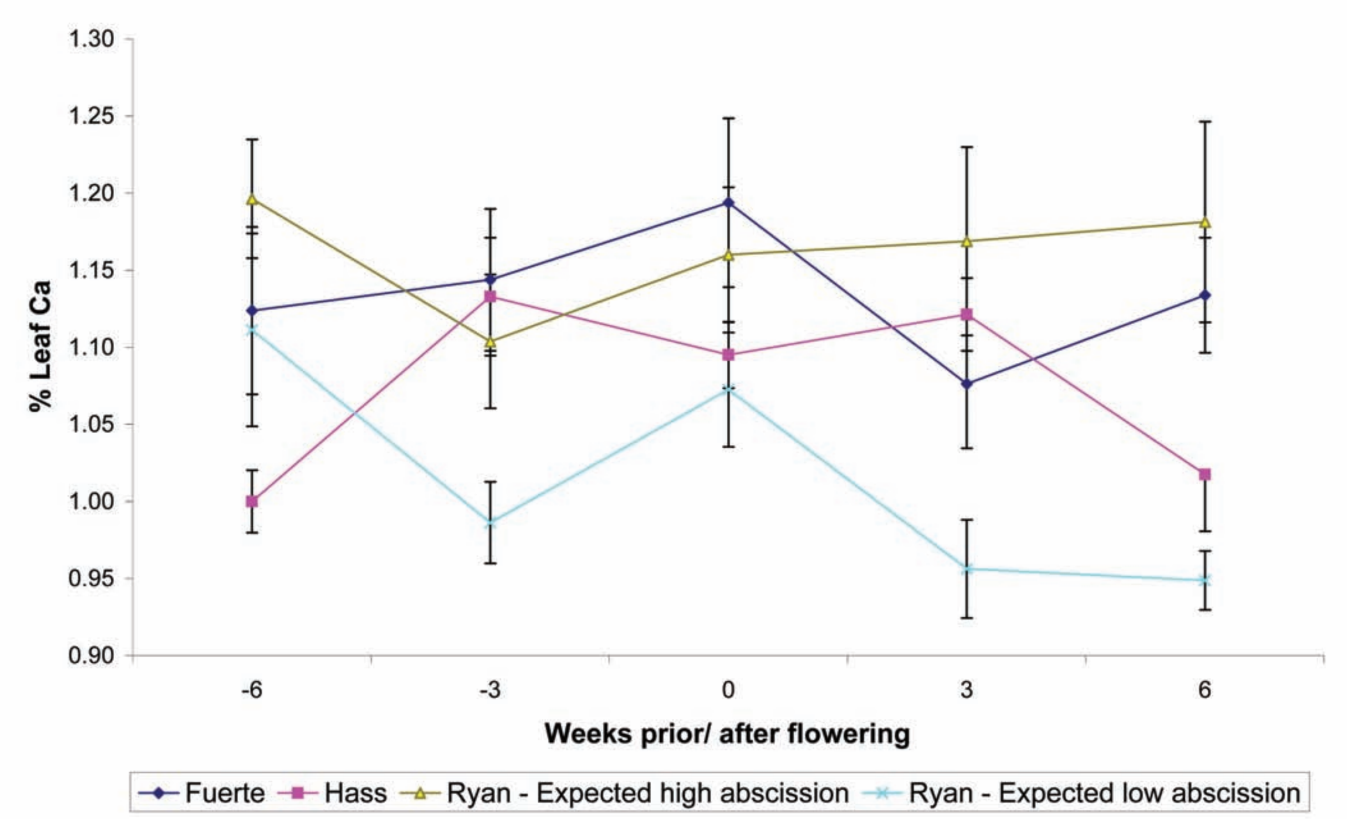

Figure 5. Comparison of leaf calcium levels for three avocado cultivars.

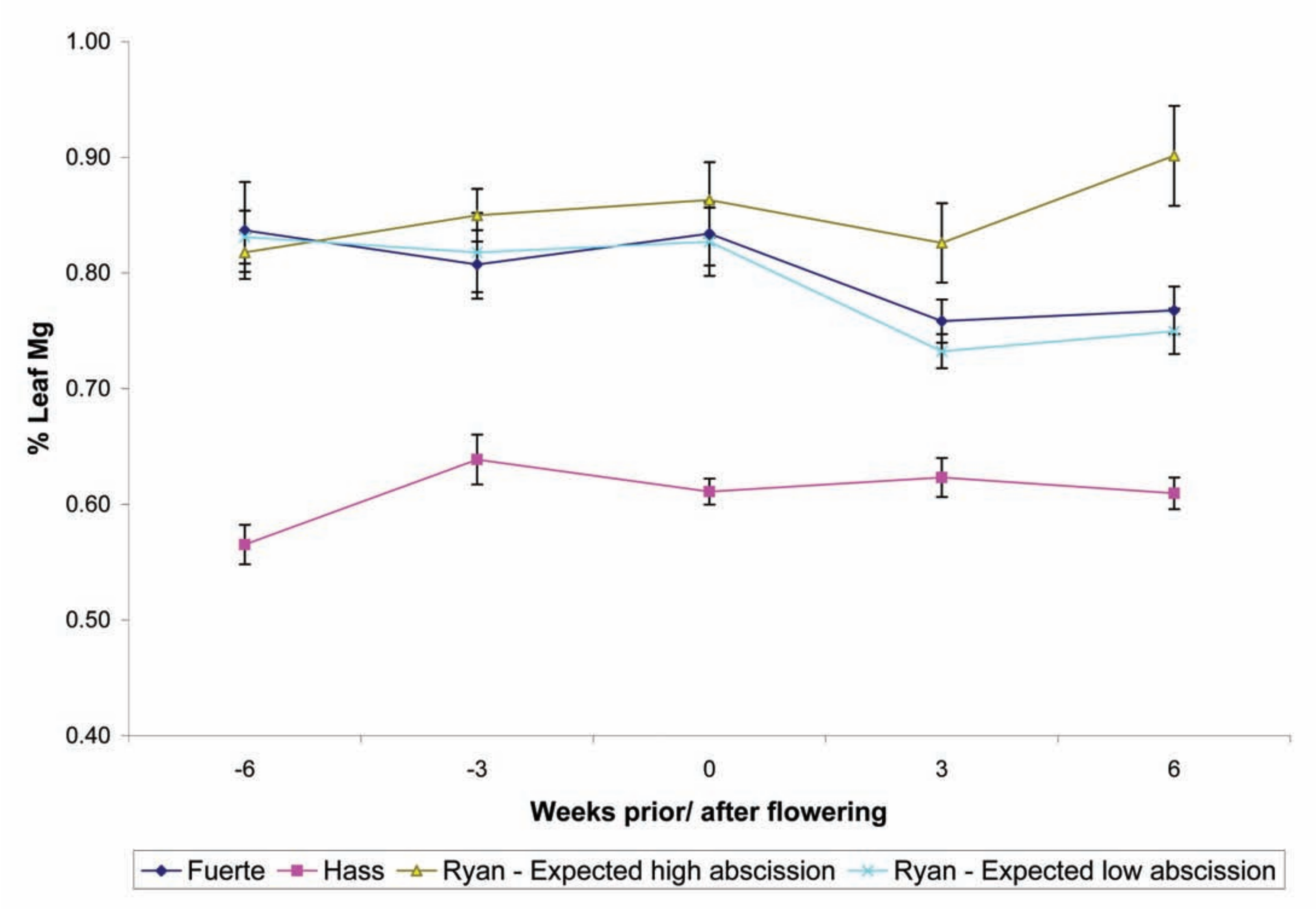

Figure 6. Comparison of leaf magnesium levels for three avocado cultivars.

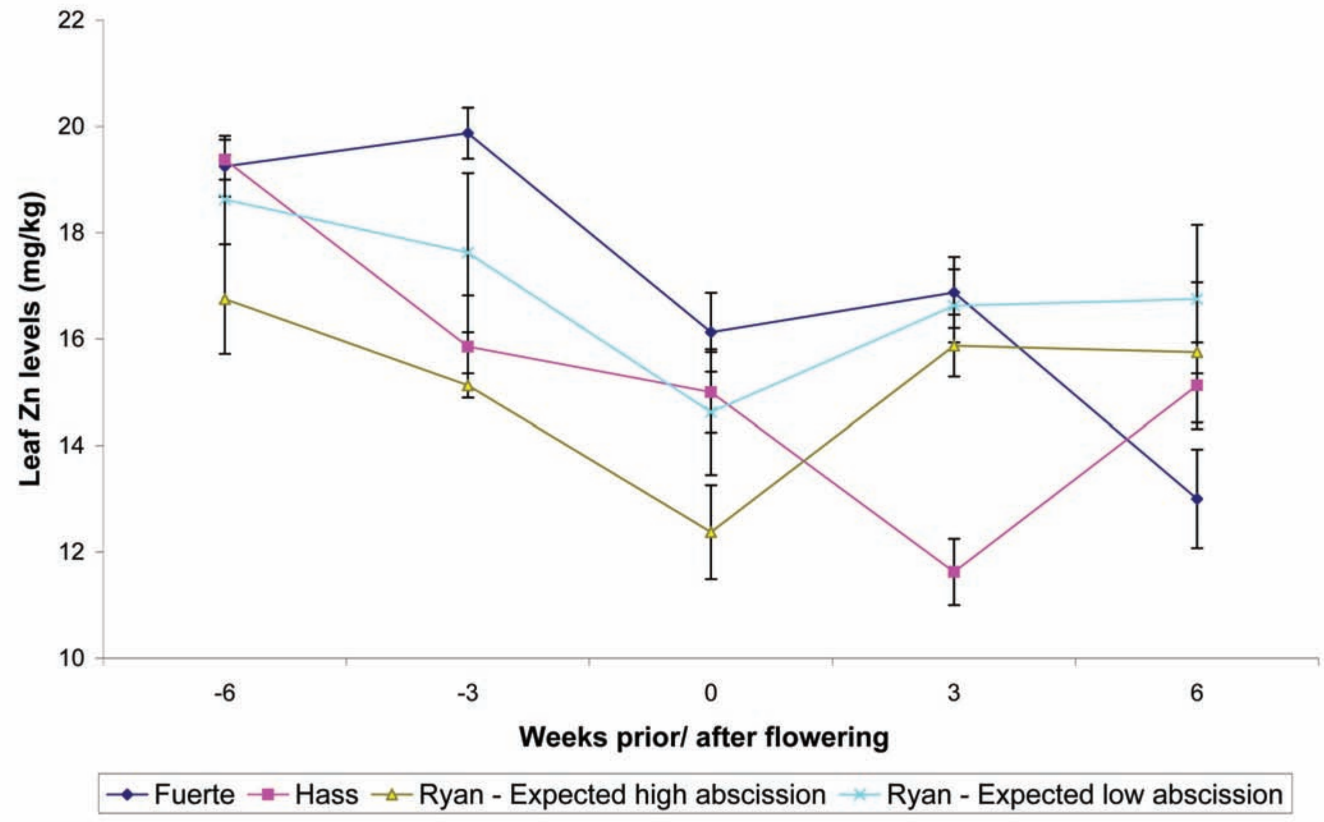

Figure 7. Comparison of leaf zinc levels for three avocado cultivars.

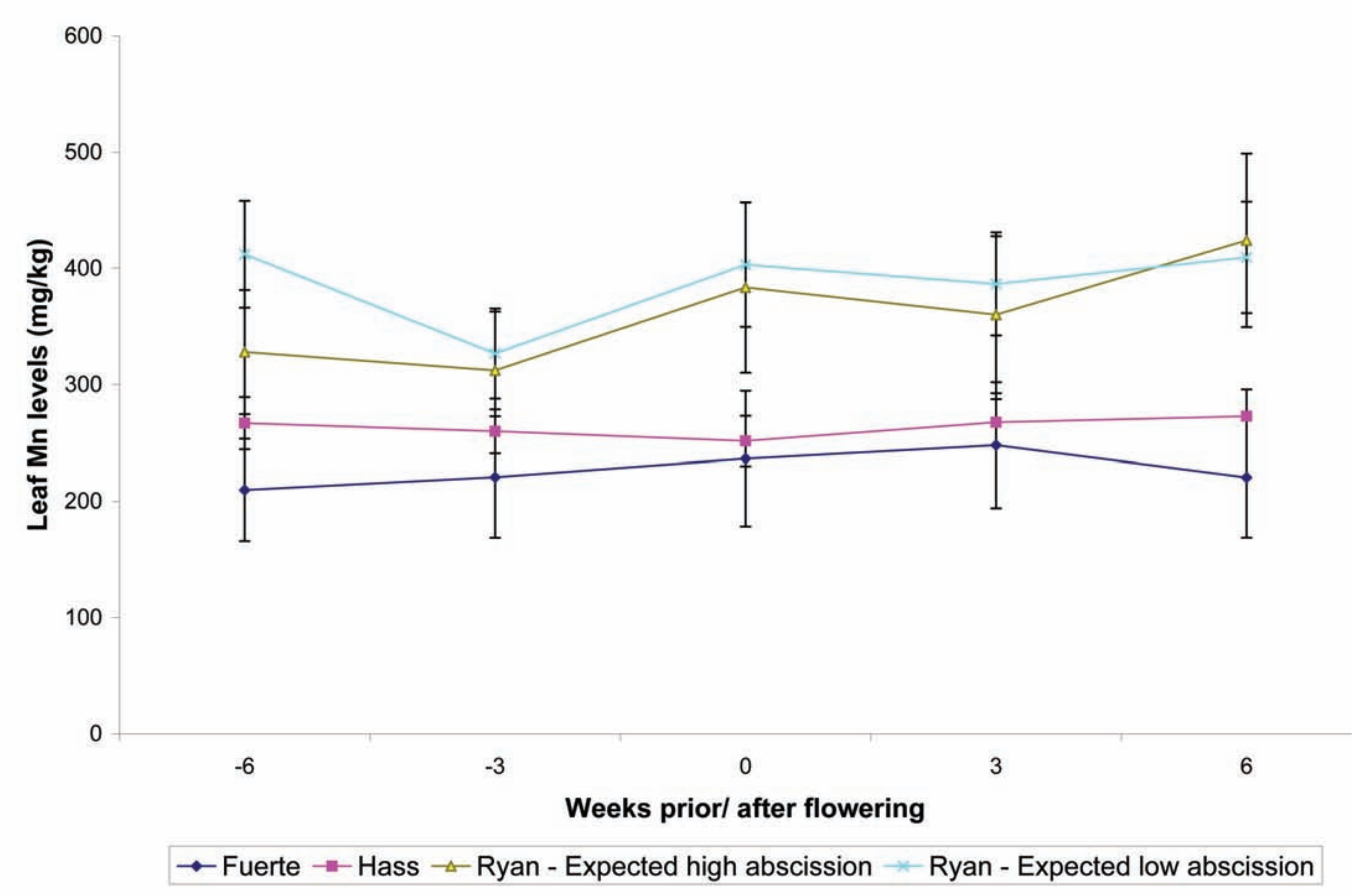

Figure 8. Comparison of leaf manganese levels for three avocado cultivars.

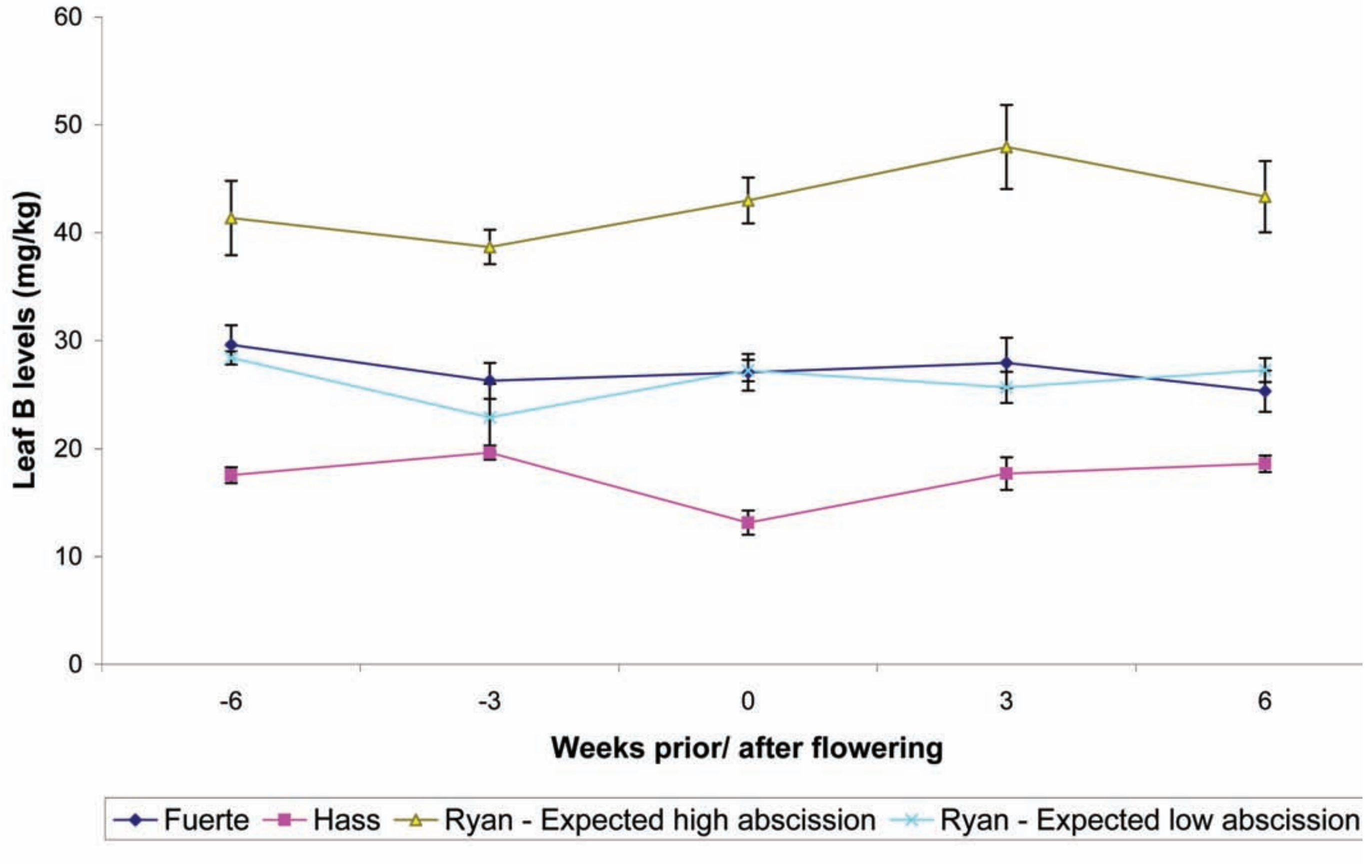

Figure 9. Comparison of leaf boron for three avocado cultivars.

period before flowering. The leaf iron pattern (**Figure 11**) could be an indication of drought stress. Stolzy *et al*., 1971 and Ponnamperuma, 1972 found that irrigation after a period of drought increased the solubility of iron, and increased uptake by the roots (Whiley & Schaffer, 1994). Increased iron transport to the leaves and increased iron uptake by the roots could be the explanation for the observed leaf iron levels (**Figure 11**) while continued irrigation would restore iron levels to the initial levels. Indications are that the orchard might have been subjected to drought and that 'Ryan' could be more sensitive to drought than 'Fuerte' and

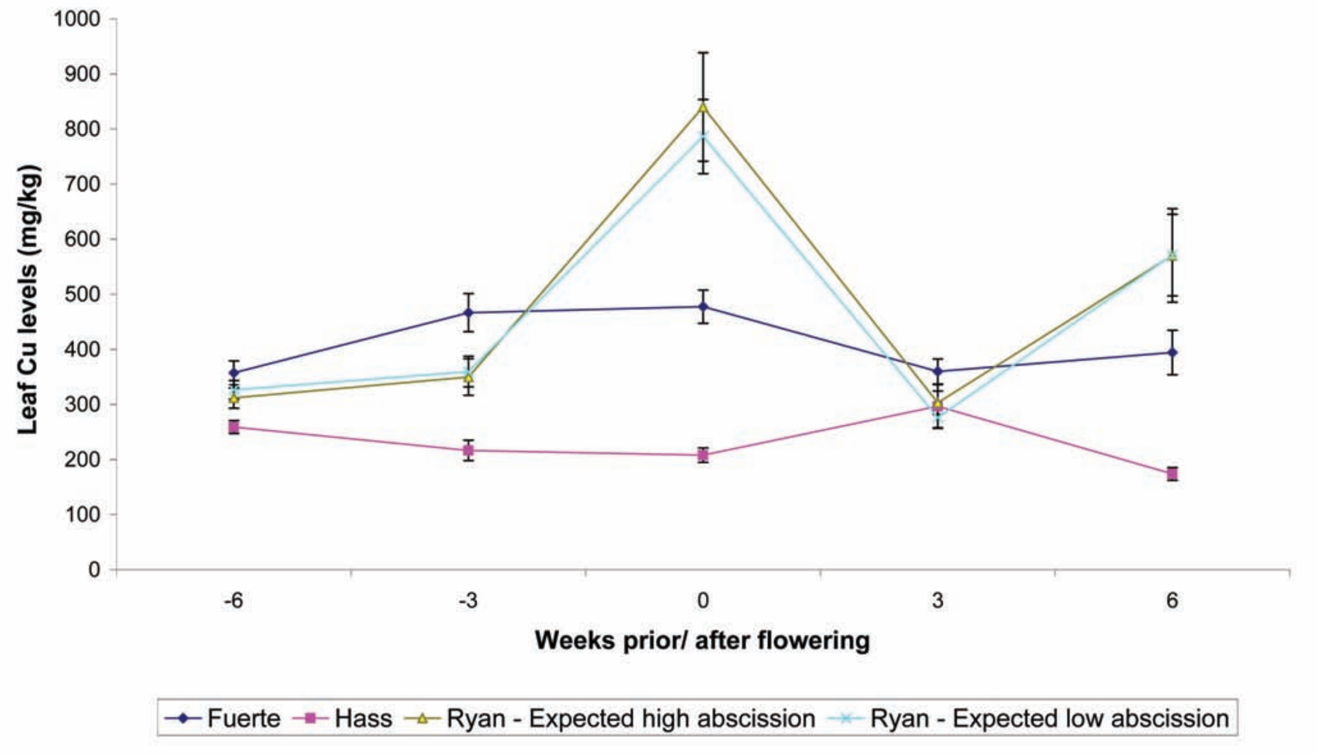

Figure 10. Comparison of leaf copper levels for three avocado cultivars.

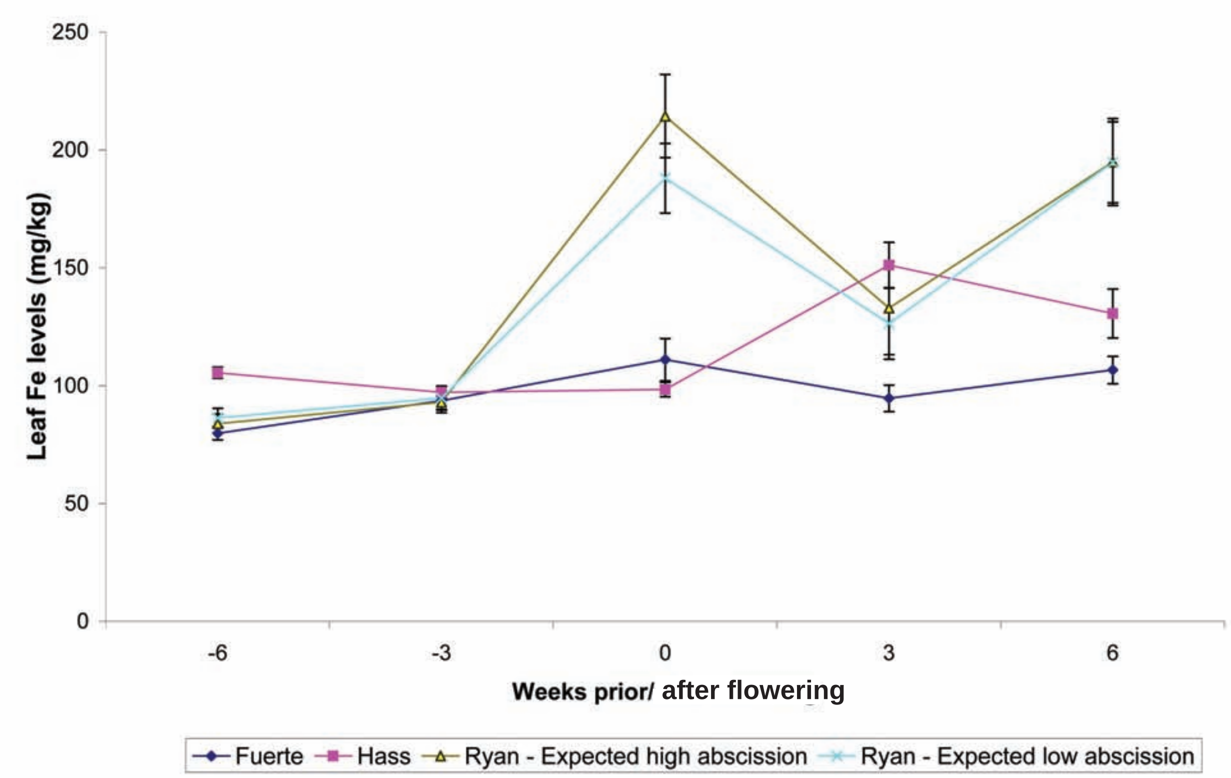

Figure 11. Comparison of leaf iron levels for three avocado cultivars.

'Hass'. Since 'Ryan' leaf drop was not statistically significant, no definite conclusions can be made as to whether partial drought conditions or sub-optimal nutrient levels contributed to leaf abscission. It is recommended that the study be repeated to further investigate whether drought and nutrient stress could contribute to leaf abscission of 'Ryan' trees.

Determination of nutrient loss by old leaves and nutrient status of flowers

The aging of green leaves leading to yellow leaves with brown

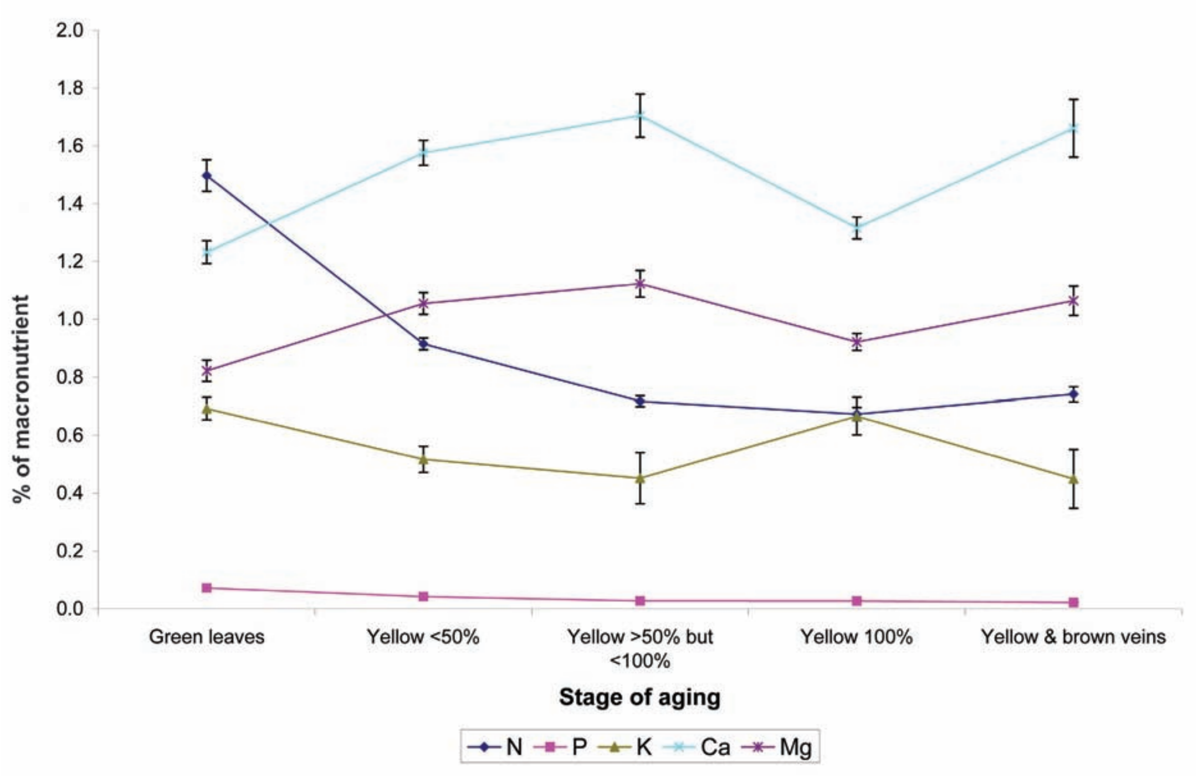

Figure 12. Macronutrient levels for 'Ryan' leaves at different stages of aging.

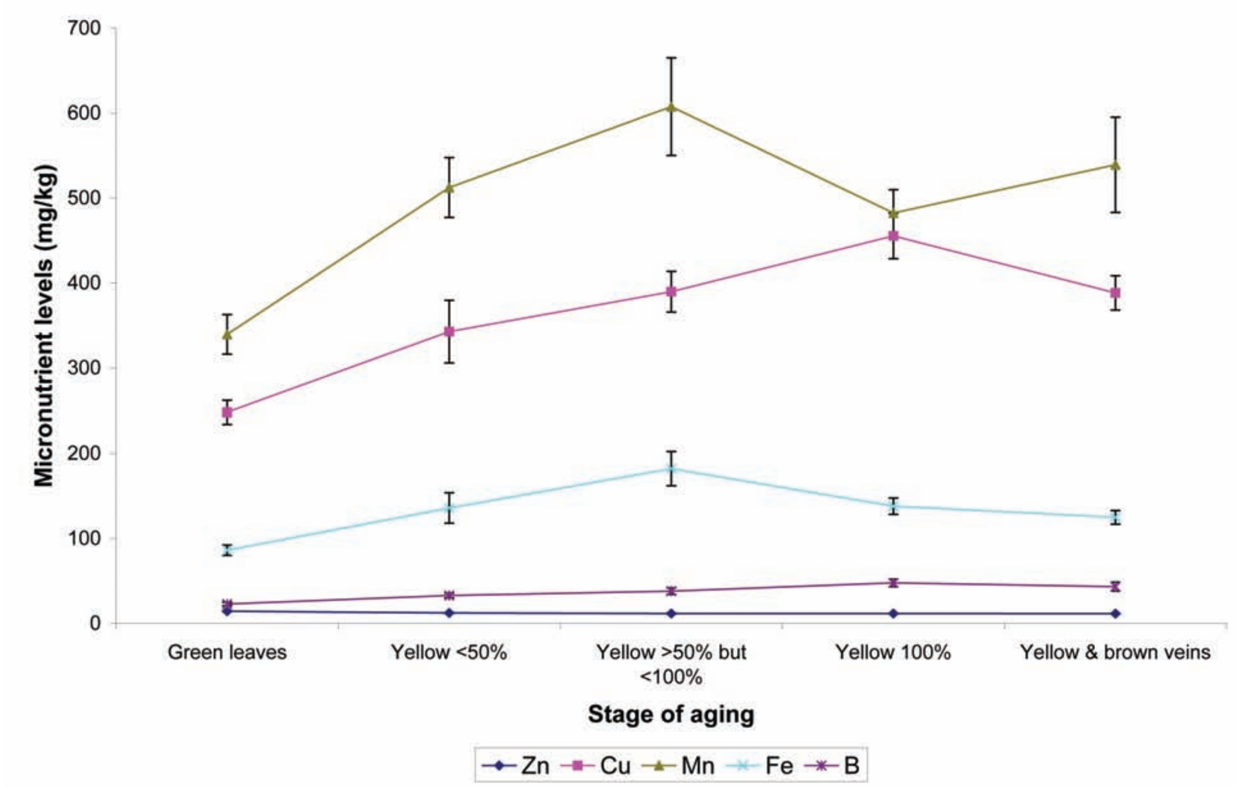

Figure 13. Micronutrient levels for 'Ryan' leaves at different stages of aging.

veins coincides with a significant decrease in nitrogen and phosphorus levels. None of the other macro- or micronutrients changed significantly (**Figure 12** and **13**). Nitrogen, phosphorus and potassium are easily transported from older leaves to newly active growing organs, while leaf calcium and micronutrients are relatively immobile (Mengel & Kirkby, 1978). Possibly, the mobilised nitrogen and phosphorus could be transported to the developing inflorescences. 'Ryan' flowered heavily during the 2005 season and the demand for nutrients that easily moved out of the leaves could have been high, if soil nutrient levels were suboptimal.

The macronutrients analysis of the flowers showed that phosphorus and potassium levels did not differ significantly between the cultivars (**Figure 14** and **15**).

Calcium and magnesium levels were higher for both 'Ryan' blocks when compared with 'Fuerte' and 'Hass' (**Figure 14**). Floral nitrogen content was higher for 'Hass' and the "low leaf abscission" 'Ryan' block when compared with 'Fuerte' and the "high leaf abscission" 'Ryan' block (**Figure 15**). The micronutrient levels of flowers differed for the different cultivars (**Figure 16**). Zinc and copper levels were significantly higher in both 'Ryan' blocks when compared with 'Fuerte' and 'Hass'. There were no significant differences in zinc and copper levels between the two 'Ryan' blocks and between 'Fuerte' and 'Hass' respectively. Both 'Ryan' blocks and 'Hass' had significantly higher manganese levels than 'Fuerte', but the differences between 'Hass' and the two 'Ryan' blocks were not significant. Boron levels did not differ between the cultivars (**Figure 16**). Of all the mobile elements it was only the magnesium content of the flowers that was significantly higher for 'Ryan' when compared to the other cultivars (**Figure**

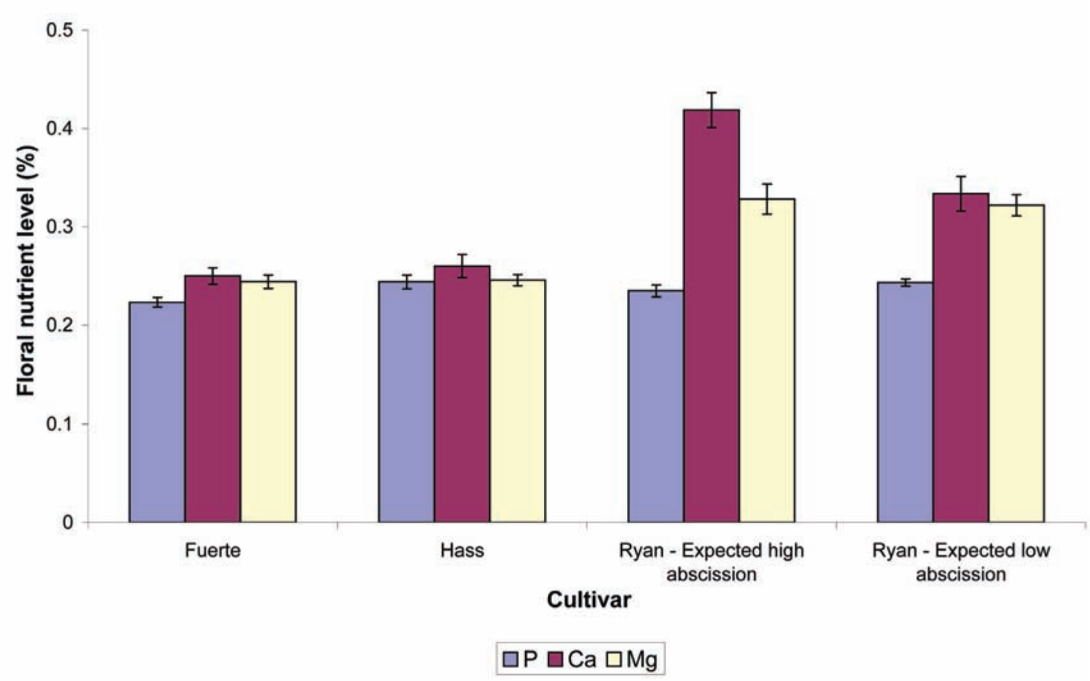

Figure 14. Comparison of floral P, Ca and Mg levels for three avocado cultivars.

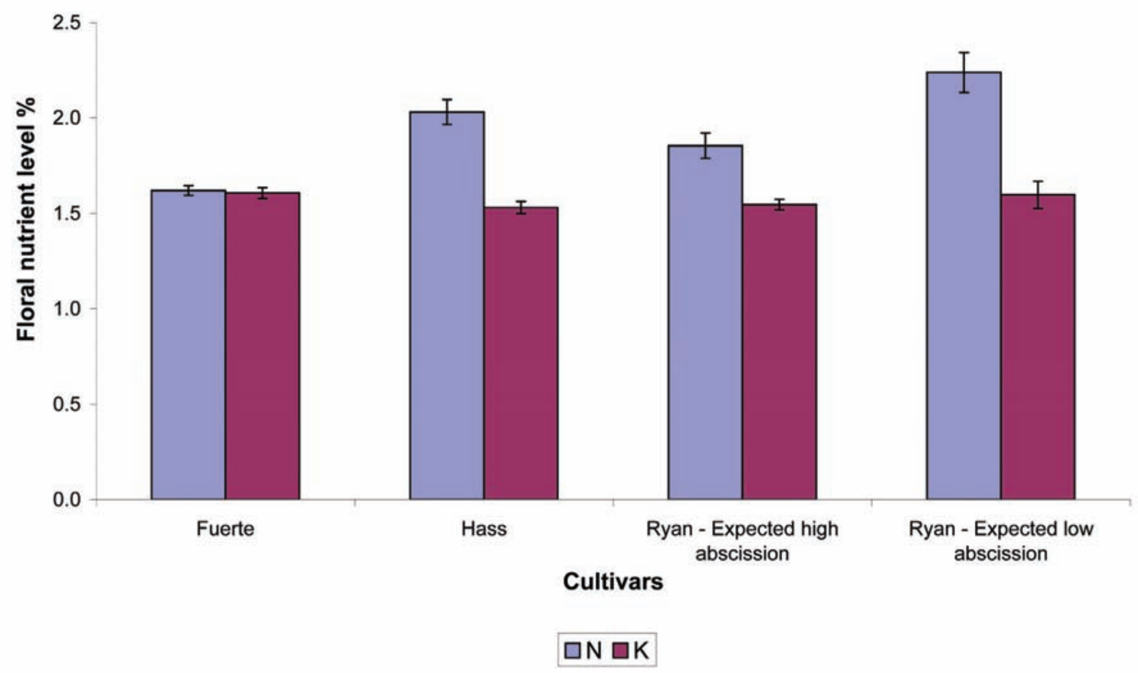

Figure 15. Comparison of floral N and K levels for three avocado cultivars.

14), but there was no significant change in the 'Ryan' magnesium leaf content during flowering (**Figure 6**). It would therefore appear as if flowering did not have a significant effect on leaf nutrient status.

Effect of reserve food resources on leaf abscission

Bark starch levels were measured to determine the fluctuation in reserve foods (Kaiser & Wolstenholme, 1994) before flowering, and during flowering and fruit set. From June to September 2005 (before flowering) no significant change was observed in the starch levels of the two 'Ryan' blocks (**Figure 17**). During flowering time and fruit set (September to October 2005) significant decreases in starch levels, to almost 50% of the original levels, were recorded for both 'Ryan' blocks (**Figure 17**) and can it be concluded that 'Ryan' depends to a large extent on food reserves for flowering and fruit set. Since flowering and fruit set

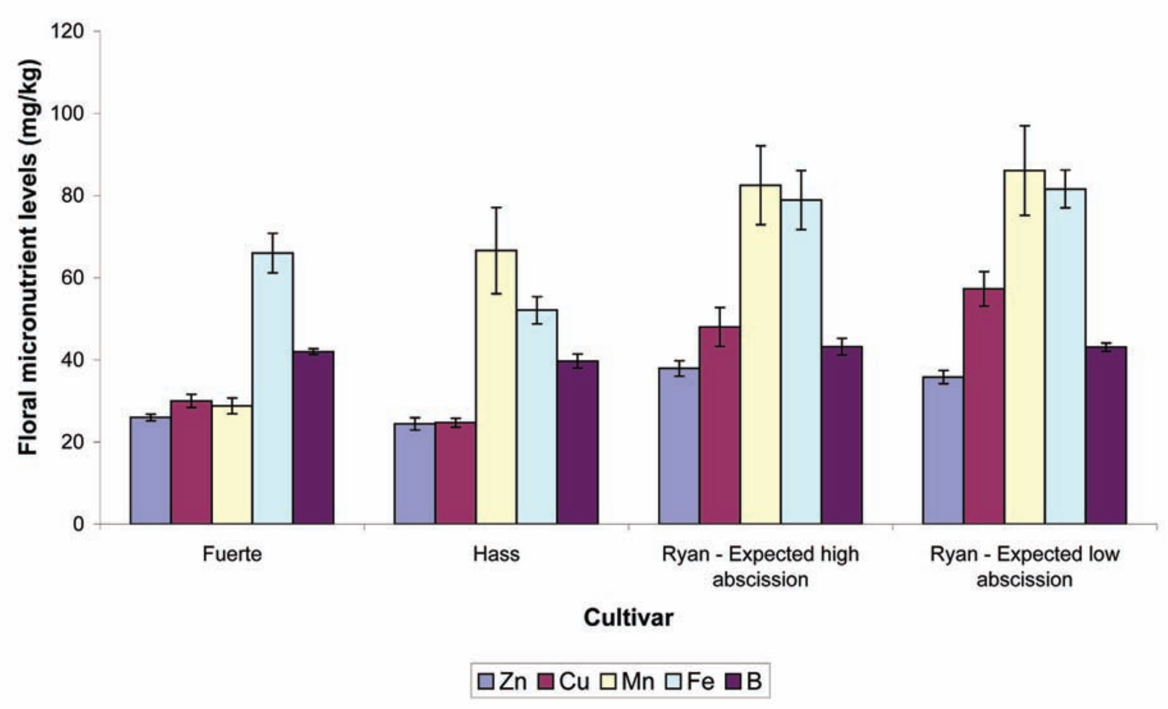

Figure 16. Comparison of floral micronutrient levels for three avocado cultivars.

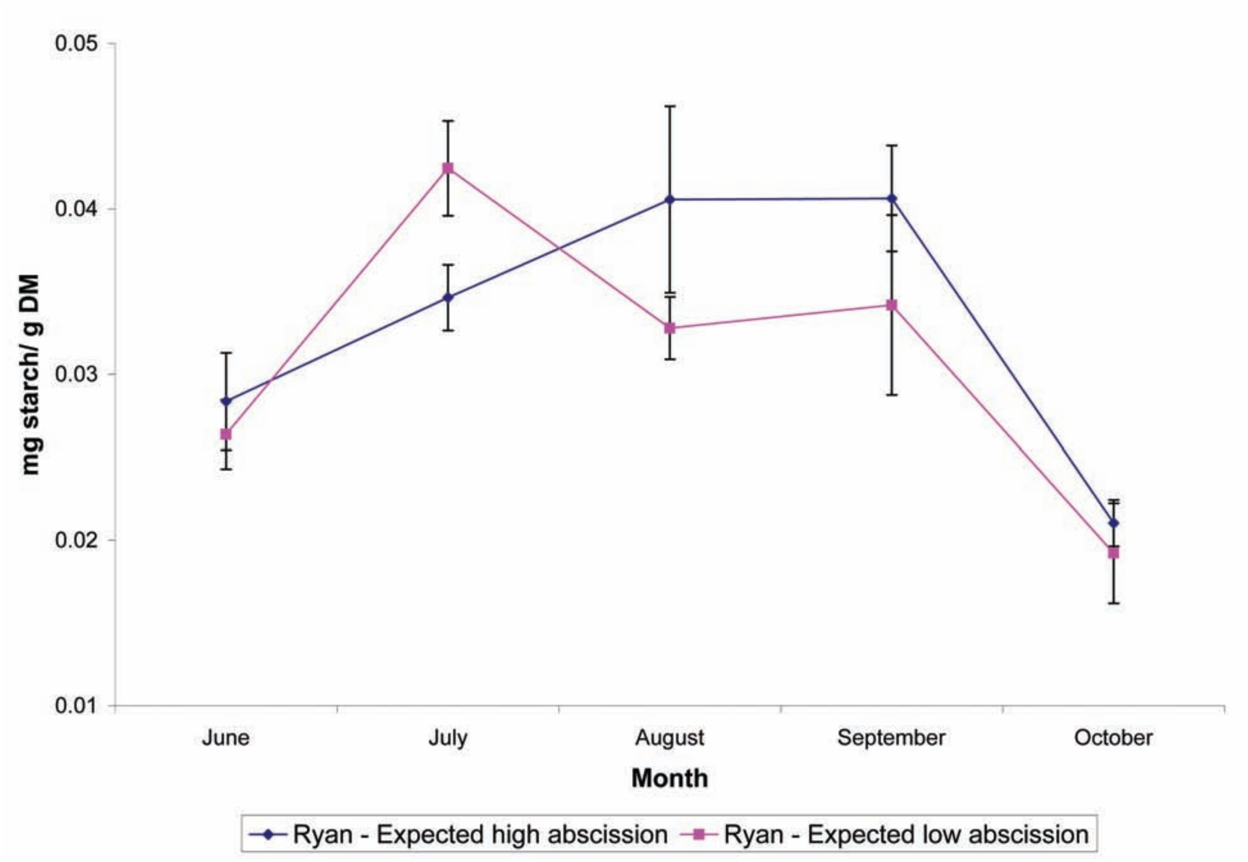

Figure 17. Changes in bark starch levels for 'Ryan' avocado.

also depend on photoassimilates from over-wintered leaves, it will be important to determine leaf photosynthetic rates studies in future research.

Effect of plant water status on leaf abscission

The plant water status was compared for 'Ryan', 'Fuerte' and 'Hass' by the determination of the early morning RWC (**Figure 18**). On the whole, no significant change in the overall morning RWC was evident over monitoring period for any of the cultivars. However, the RWC for 'Hass' and 'Fuerte' was lower one week before flowering when compared with 'Ryan'. The "high leaf abscission" 'Ryan' block had a lower RWC, not only during flowering when compared to the other blocks, but its RWC significantly decreased from two weeks before flowering until flowering time,

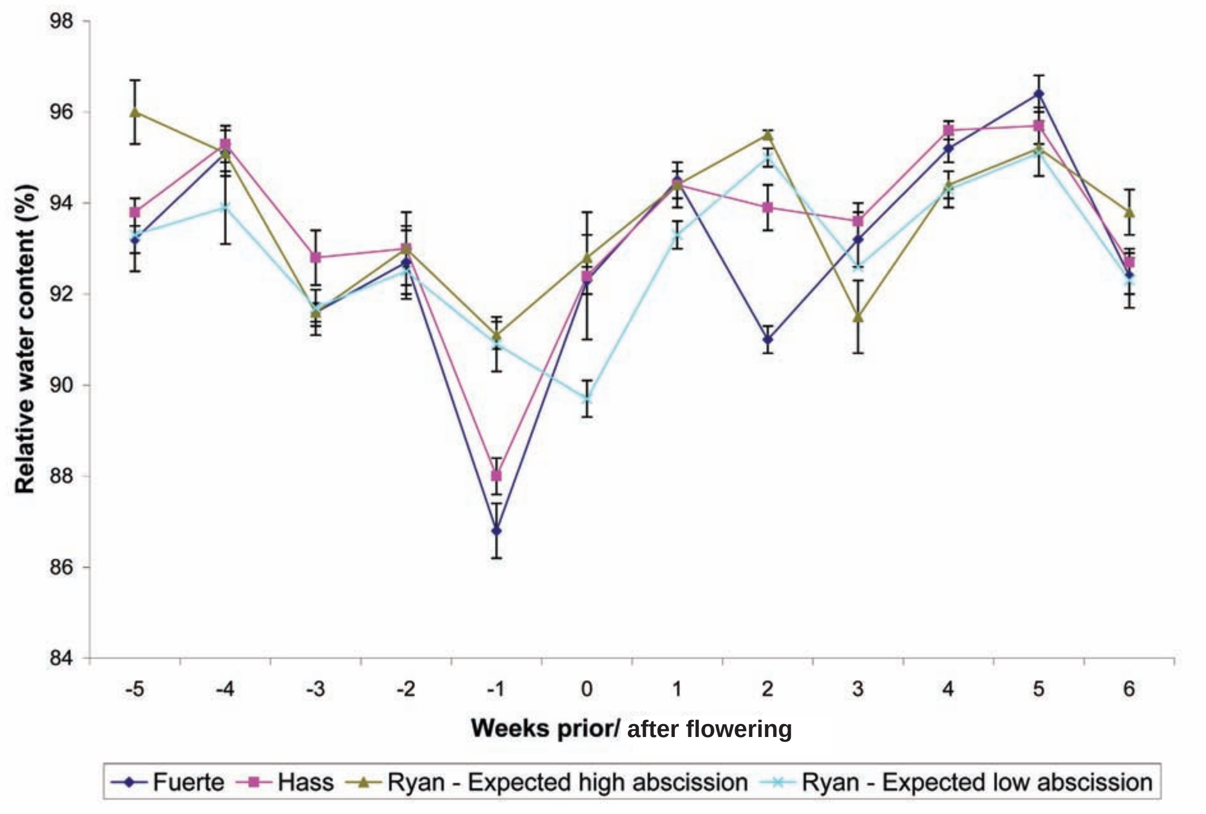

Figure 18. Relative leaf water status compared for three avocado cultivars.

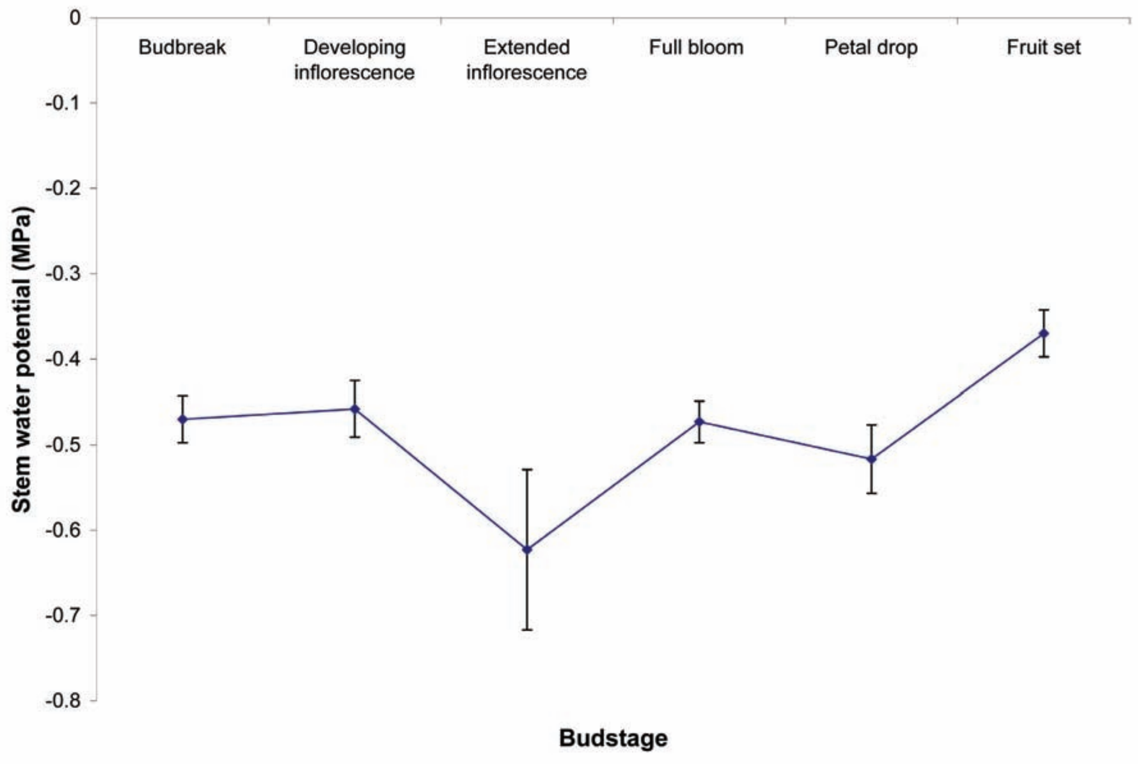

Figure 19. Midday stem water potential for 'Ryan' at six different stages.

followed by an increase in RWC after the flowering period.

With regard to the midday leaf RWC, a significant decrease for the "high leaf abscission" 'Ryan' block occurred from the extended inflorescence stage to the flowering stage (**Figure 20**), and having a trend similar (**Figure 18**) to that of the early morning leaf RWC for the trees of this block. The midday stem water potential (**Figure 19**) showed no significant changes in the tree water status of 'Ryan', from the time of budbreak to the time of fruit set. The trend in midday leaf RWC of the "high leaf abscission" 'Ryan' block (**Figure 20**) is similar to the trend in midday stem water potential though shifted one week earlier (**Figure 19**), both trends indicating that the trees experienced a water deficiency at the time of full bloom when leaf abscission for 'Ryan' was high, though not statistically significant.

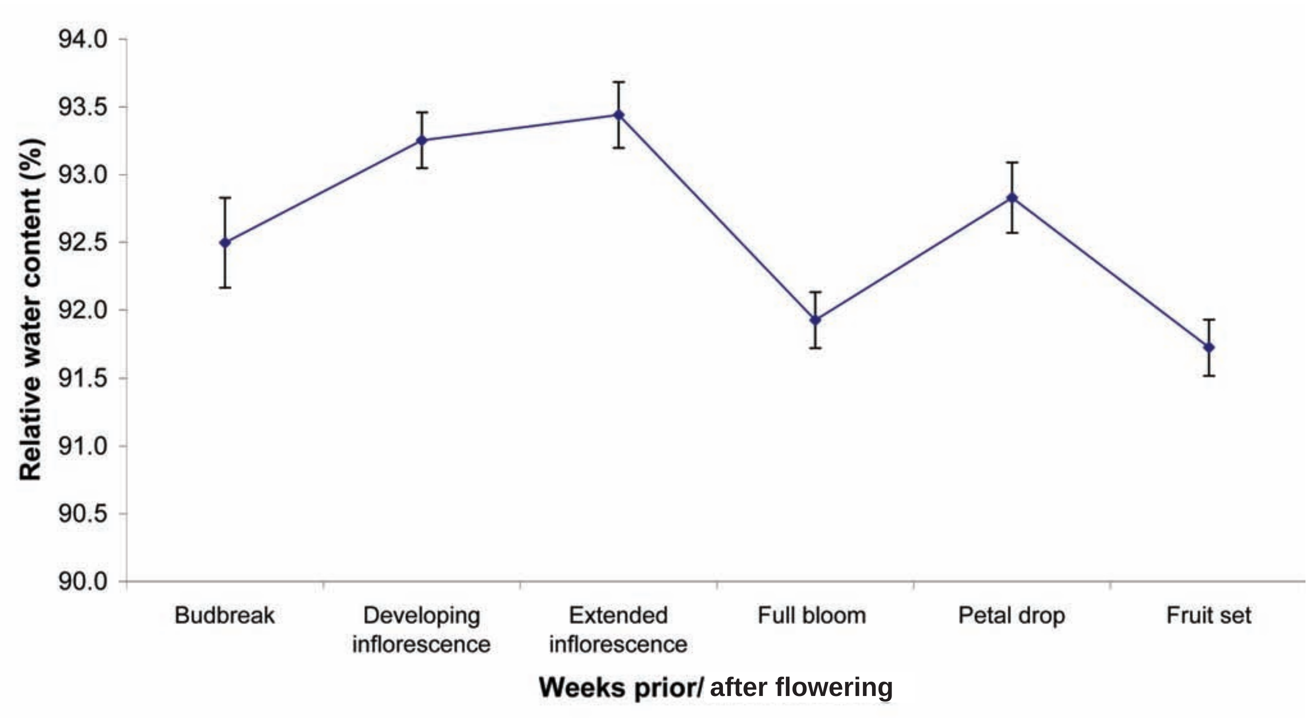

Figure 20. Relative leaf water status compared for three different avocado cultivars.

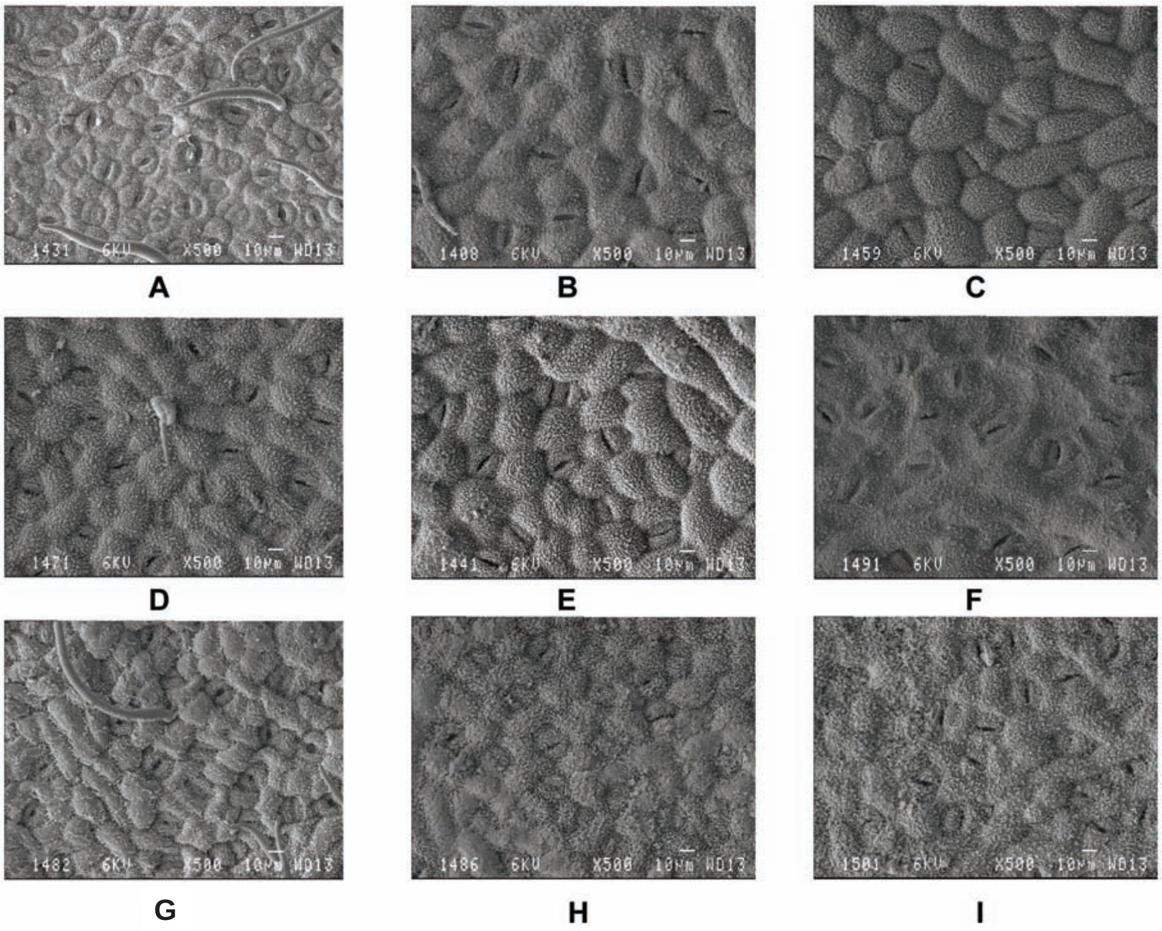

Figure 21. SEM photographs of the abaxial surface of avocado leaves sampled at different stages (A - F: cultivar Ryan, with A: soft green leaf; B: green hardened-off leaf; C: less than 50% yellow colouration; D: more than 50% but less than 100% colouration; E: 100% yellow colouration; F: 100% colouration and browning of veins. G-H: cultivar Fuerte, with G: soft green leaf; H: green hardened-off leaf; I: 100% yellow colouration).

Anatomical work

No structural difference was observed between the cuticles of the 'Fuerte' (Control) and 'Ryan' (**Figure 21**). The 'Fuerte' and 'Ryan' cuticles of the young immature leaves were thin (**Figure 21A** and **G**) but thicker at the other stages (**Figure 21B** to **F** and **H** to **I**). In none of the stages were any of the stomata blocked to such an extent that gas exchange was impossible, indicating that cuticles probably did not affect the functionality of the leaves which could therefore possibly still support the developing tissue with photosynthates. No defined abscission layer could be found for abscised 'Ryan' leaves (**Figure 22**). This phenomenon will be further investigated.

Chemical applications to prevent excessive leaf abscission

Leaf abscission was measured as the percentage of leaves that dropped from the time of application (approximately six weeks before flowering) until fruit set. For the "high leaf abscission" 'Ryan' block no significant differences were observed between the control and any of the treatments (**Figure 23**). For the "low leaf abscission" 'Ryan' block, leaf drop was significantly higher in the 30 mg/ℓ NAA, 20 mg/ℓ sodium nitroprusside and 3% kaolin treatments when compared with the control (**Figure 23**). Fruit set was recorded as the number of fruit set per inflorescence two weeks after full flowering. Fruit set was negatively affected in all the treatments in the "low leaf abscission" 'Ryan'

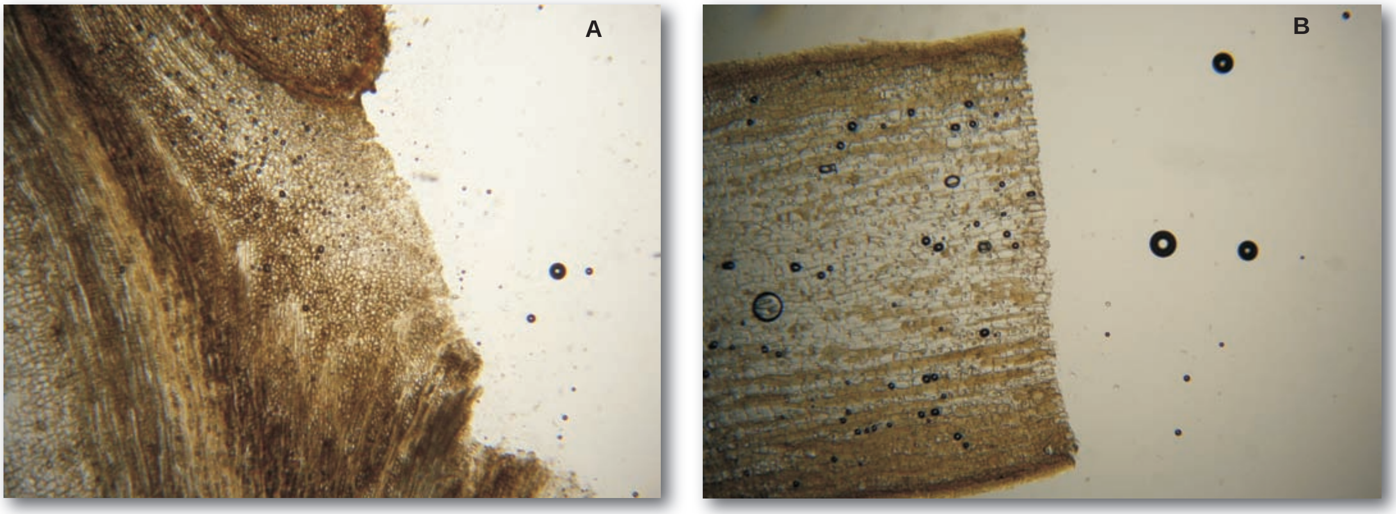

Figure 22. Light microscopy photographs showing leaf abscission for yellow coloured 'Ryan' leaves (A: attachment site on the main stem; B: attachment site of the petiole).

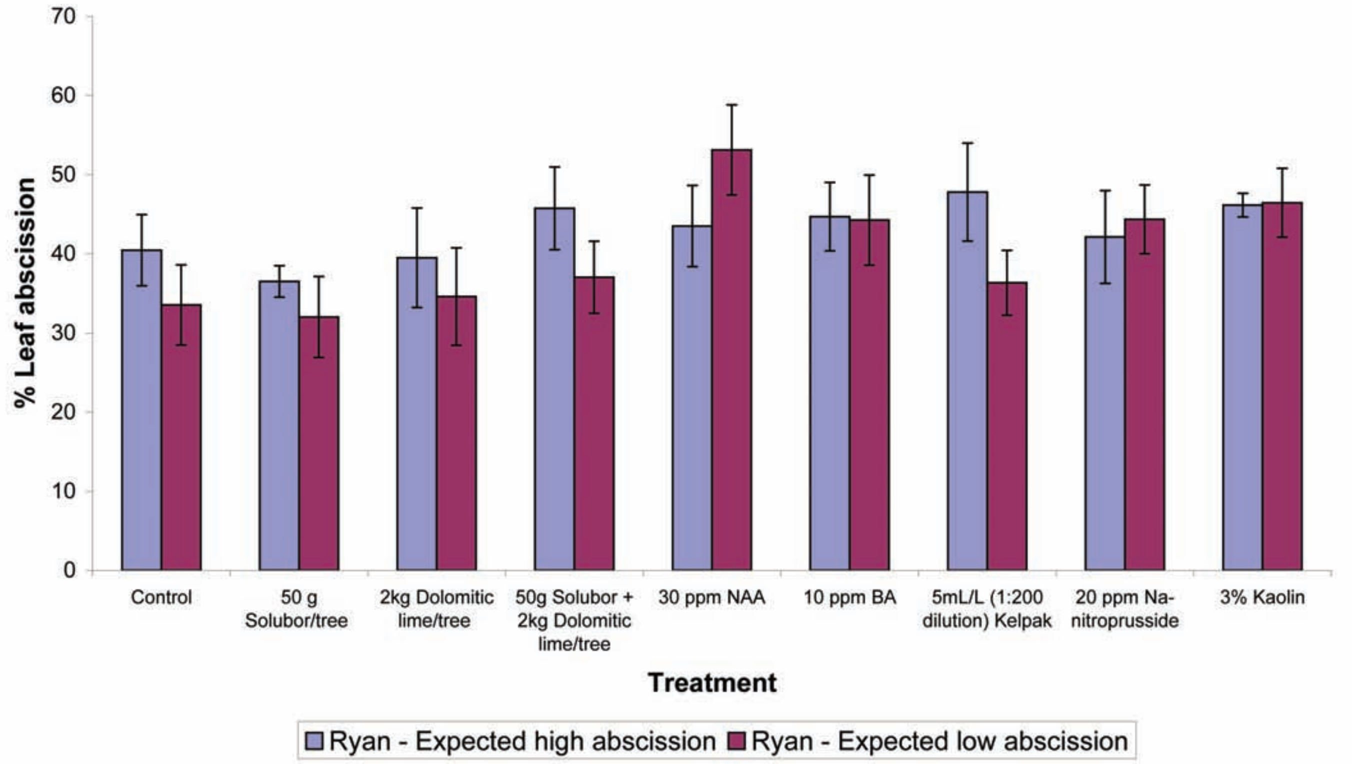

Figure 23. Effect of chemical applications on leaf abscission of 'Ryan'.

block, but no such effect was found for the “high leaf abscission” ‘Ryan’ block (**Figure 24**). Most probably there is an optimum period during the season when the application of chemicals should have its maximum effect on the prevention of leaf drop but without affecting fruit set negatively. Leaf abscission was less severe during the 2005 season, which could explain why the chemical applications did not have the desired effect. Abscission can also be promoted in cases where chemical applications are made at the wrong physiological stage (Abeles & Rubinstein, 1964). It is therefore important to determine the optimum time for the application of chemicals to prevent or retard leaf abscission.

CONCLUSION

Leaf abscission for the cultivar Ryan was not significant during the 2005 season. Although there was a decrease in nitrogen and phosphorus content of leaves prior to flowering, it appears to be a normal phenomenon. The trend observed for leaf iron levels could indicate that the ‘Ryan’ blocks in which the experiments were performed could have experienced drought stress. However, there was no drastic difference in the leaf RWC between ‘Ryan’, ‘Hass’ and ‘Fuerte’. Possibly, ‘Ryan’ trees could be more sensitive to drought stress conditions. No structural changes could be found for ‘Ryan’ to indicate that the leaves became so non-functional with aging that leaf abscission was initiated. The effect of chemical applications on leaf retention and fruit set differed between the two ‘Ryan’ blocks, showing either no effect or a negative effect. It is therefore important to determine the optimum application time for the application of chemicals.

Acknowledgements

The authors would like to thank SAAGA for the financial support, and Mr Chris van der Merwe for his excellent help with the use of the electron microscope.

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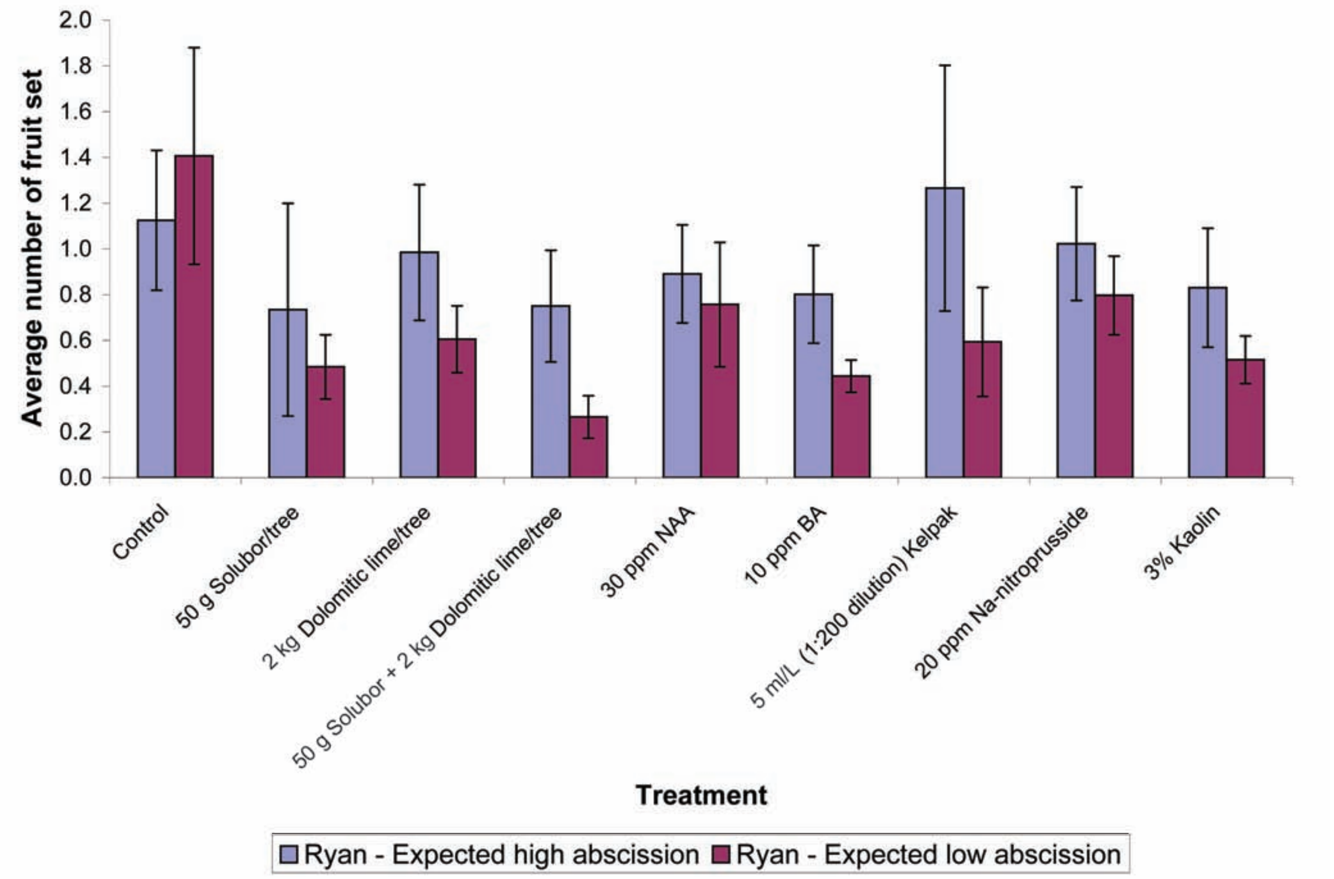

Figure 24. Effect of chemical applications on fruit set of ‘Ryan’.

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