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Project number:	HNS 193
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Report:	Annual report, February 2018
Previous report:	Annual report, February 2016, 2017
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Date project commenced:	01 May 2015
Date project completed	31 December 2017

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

AUTHENTICATION

We declare that this work was done under our supervision according to the procedures described herein and that the report represents a true and accurate record of the results obtained.

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CONTENTS

SCIENCE SECTION

Introduction

The final year of the trial allowed us to identify trends in the nutrient status of the shrubs tested. The trials were on a reduced crop range for the final year which enabled a more intensive equipment reading and analysis programme to be carried out. Trials were set up in Greenmount College in Northern Ireland (NI), PCS at Destelbergen near Ghent in Belgium and Coles of Leicester (Syston site). In addition Osberton Grange nursery successfully used the atLEAF+ device on an Azalea crop.

At all sites a fully replicated, randomised trial involving three plant species and four levels of fertiliser was set up. The hand-held devices were used at the trial sites to gather data, which were then compared to traditional laboratory samples of leaf tissue and substrates. Additionally, the setup of the trials enabled the collection and analysis of leachate from a number of pots in each treatment to assess nutrient loss. Irrigation and rainfall inputs were recorded at all sites.

Materials and methods

Plants

Plant species used were Tradescanthia, Viburnum and Buddleja with Azalea at Osberton nurseries only.

Plants were grown in 1.5 litre pots and in identical physical substrates to the previous two years.

Six plants were placed in a Pour-thru device (or lysimeter) that collected any excess rainfall and irrigation. The water volume was measured at monthly intervals or more if the volume collected was excessive. In periods of heavy rainfall this was more frequent than periods of irrigation. In periods of heavy rainfall 500 ml samples were retained in a darkened refrigerator and added to a 500 ml sample taken at the end of the month for nutrient content analysis. A further 30 plants were placed around the Pour-thru device for the purposes of destructive and non-destructive tissue and substrate sampling. Substrate and tissue samples were not taken from the Pour-thru device plants and only one substrate sample was taken from a pot to avoid substrate volume reduction affecting results.

Equipment

Full details of the equipment used in year 3 of the project are described in the year 1 report.

atLEAF$^+$

The atLEAF$^+$ has proved a cost effective chlorophyll measuring device. After a period of production inactivity, the device is now available for purchase again.

There are two LED emitters in the upper part of the aperture at two wavelengths, red at 660 nm (nano meters) and near infra-red (NIR) at 940 nm. Light filtered through the leaf is captured by a sensor below which measures the absorbance of the leaf. The difference in transmission of the filtered wavelengths gives a measure of chlorophyll content in atLEAF$^+$ units (1-100).

The atLEAF$^+$ measured the chlorophyll levels in the plants well but is measured in open daylight. When measuring it was important that the main vein of the leaf was not in the measuring point. We did not find any influence of daylight on the readings.

Apogee MC100

The Apogee MC 100 was introduced into the project to replace the unavailable atLEAF$^+$ device. It measures chlorophyll with the near identical wavelength led light sources as the atLEAF$^+$ but it is within a closed chamber so avoids any daylight variations. It also uses a smaller sample size of leaf making readings easier.

The Apogee model MC 100 chlorophyll concentration meter measures relative chlorophyll content and outputs. The chlorophyll concentration is measured by two light emitting diodes, one emitting 931 nm and one emitting 653 nm with paired detectors giving chlorophyll concentration in units of µmol per m^2 of leaf surface. The ratio of radiation transmittance is measured at two different wavelengths and outputs chlorophyll concentration automatically. The Apogee MC 100 used at Greenmount and the CCM 200 used at PCS are the same piece of equipment differently badged.

Green Index

Green Index is an iPhone App that uses the phone camera to establish the colour of the leaf to determine chlorophyll colour. The device uses a colour board to create a reference for the camera and gives a figure that is called a Dark Green Colour Index (DGCI) between 0 and 1. This App was developed to capture differences in 'greenness' between maize leaves. The green and yellow discs present on this reference board are known colours (standards) used by the software to calibrate differences in light conditions; the pink background increases contrast and reduces noise, and the grey colour calibrates the white balance.

Procheck

The Procheck with GS3 sensor measures the electrical conductivity (EC) of the growing media and is easy to use and closely follows the tissue nitrogen levels. The device is inserted into the substrate and gives an immediate reading showing the conductivity in Micro-Siemens (µS). The sensor, GS3, measures soil moisture, temperature, and EC of the substrate. The probe has three steel needles that improve sensor contact in porous substrates such as peat or perlite. By measuring EC in the substrate solution, the sensor measures the total amount of salts dissolved in pore water. It does not give information on the amount of a specific nutrient. However, since the majority of salts in the substrate are macronutrients, EC can be used as an indicator of the presence of macronutrients in the growing medium.

Substrate mixes

<u>Product Specification</u>

Main ingredients

3 parts Dark peat 18 MM
6 parts Light peat 18 MM } 80% of the mix

1 part Sod peat 18 MM

FOREST GOLD® ------------- 20% of the mix

Nutrients added per m³:
1 KG BASE 15-10-20+TE

0.1 KG UREA

3 KG LIME/DOLODUST

0.4 LITRE WETTING AGENT

CRF additions:

2, 4, 6 and 8 kg/m^3 of 8-9 month Osmocote Pro 8-9 month 16:11:10 (standard release product). This is the second generation material and would be considered comparable to Nutricote, Basacote, Plantacote products of similar longevities.

Laboratory analysis

All plant tissue, substrate and water analysis was carried out at the same UK laboratory to avoid differences in the results and analysis methods. Results were given in standard UK units of milligrams/litre (mg/l), with pH measured using the water extract. EC was measured in micro Siemens (µS). Tissue samples were taken from new stems and the foliage selected was taken from the first fully matured new young leaves. Leaf tissue was taken from all three replicates and placed into one sample, according to the substrate CRF rates. Similarly substrate samples were taken from the guard pots from all three replicates and bulked into one. Substrate samples were taken from the mid pot area, avoiding the top and bottom.

'Pour-thru' collection and analysis:

Every four weeks the volume of drained water in the lower container of the pour-thru was measured, sampled for water analysis and emptied. At the same time substrate samples were also taken and sent to the laboratory. Samples from each site were sent to the same laboratory to avoid any differences in procedure. Temperature and rainfall were measured by weather stations present at each site. The amount of water received by the plants (rainfall plus irrigation) was measured by rainfall gauges. In this way run-off and nutrient leaching could be monitored to give researchers an accurate idea of the level of nutrients being used by the plants and lost through leaching.

Weather records

Half hourly weather records were taken at each site covering air temperature, humidity, rainfall and light levels. Rainfall was measured in mm and light levels were measured in watts/m^2. See appendix 2, Figures 36 and 37

Statistical Analysis Protocol

The data comprised the treatment means for four treatments (2, 4, 6, 8kg) observed on each of 3 sites for a number of variables. Not all variables were observed and results were concentrated on the relationship of the equipment data to plant tissue analysis results. This data was available for three species (Viburnum, Tradescanthia, Buddleja). The analysis comprised an ANOVA test of the variables and regarding the sites as randomised blocks and ignoring the dependencies between measurements taken on a plot a different times. These were regarded as being independent measurements for the purposes of the analysis.

Results

Laboratory results:

All the plants had been potted at or around week 19 and whilst the initial leaf tissue nitrogen percentage (N%) levels were quite good at or near potting, the N% figures dropped for plants after the beginning of June and in many cases resulted in N% tissue levels being below standard values (stated in Mills and Benton Jones, 1996) expected for such plants. Figures 5-7 show the N% in tissue from June until October and clearly show the decreasing level of nitrogen (N) in the tissue. This may well reflect the initial release of fertiliser from the CRF granules ahead of the plants really establishing, and also supports the initial high levels of nutrients in the leachate, Figures 8-10. The result of leaching in the early stages after potting at Greenmount meant that there was insufficient N levels in the CRF for optimum growth later in the season.

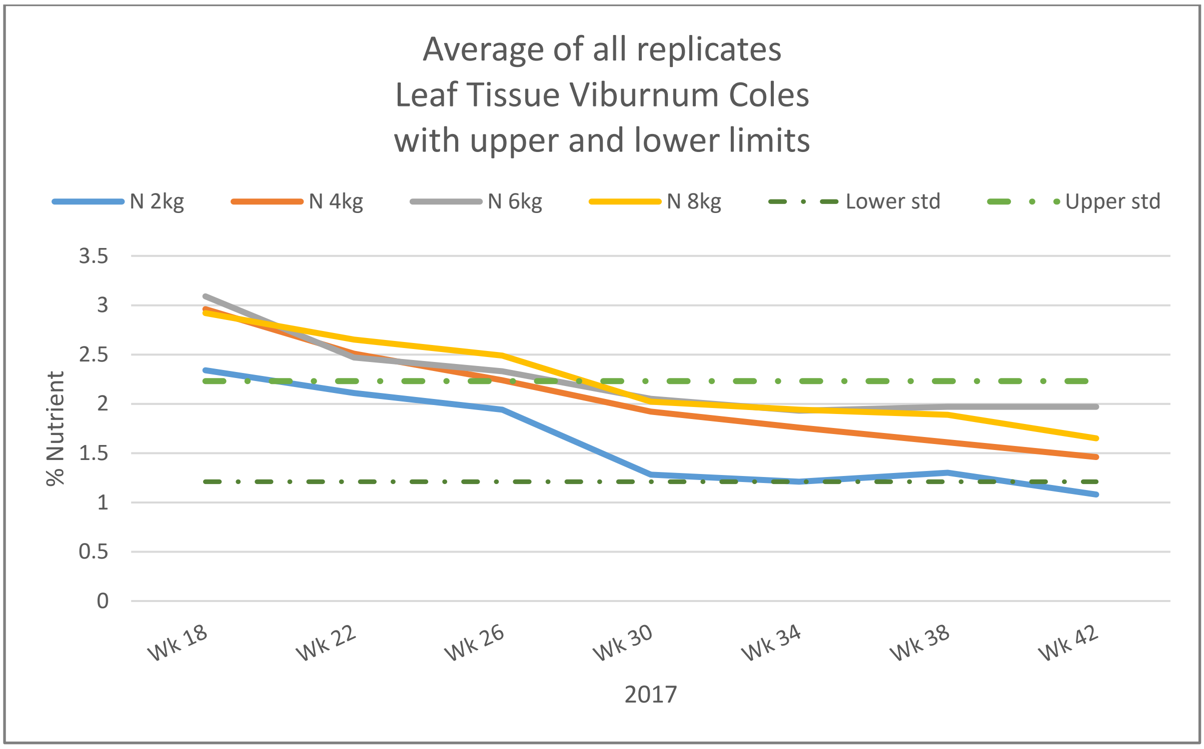


Figure 5 shows the N% in the leaf tissue levels for Coles including standard values (Mill and Benton Jones, 1996).

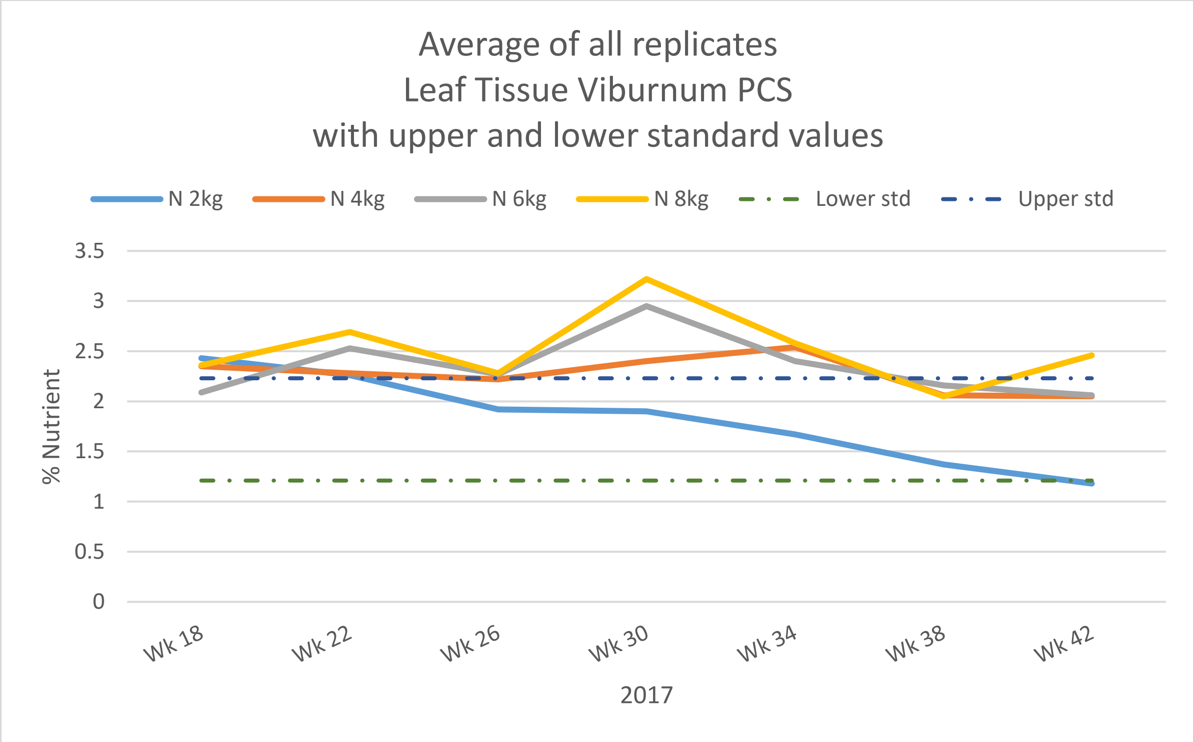


Figure 6 shows the N% in the leaf tissue levels for PCS including standard values

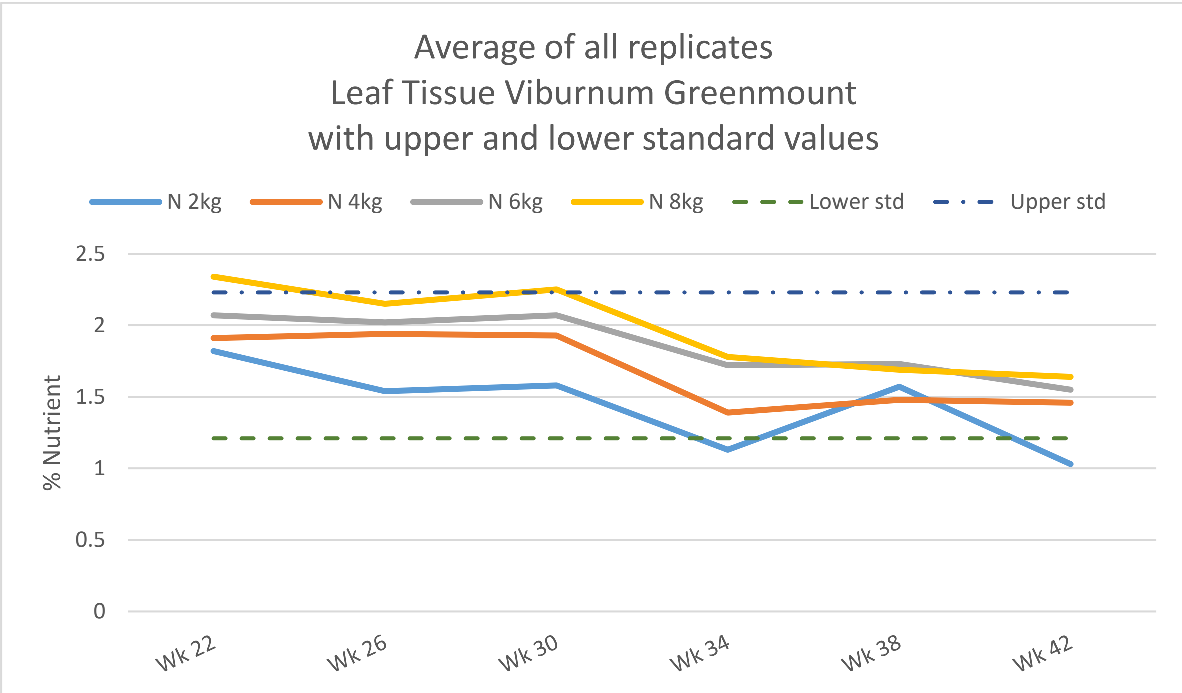


Figure 7 shows the N% in the leaf tissue levels for Greenmount including standard values

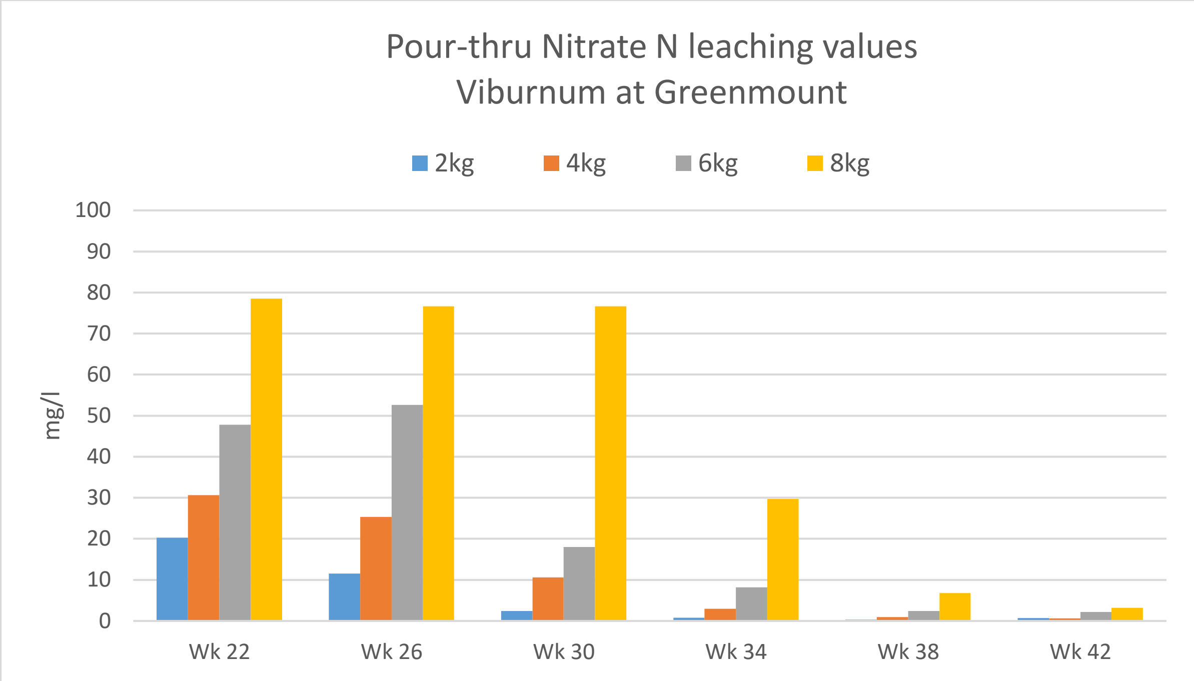


Figure 8 shows nitrate N leaching at Greenmount for Viburnum across all CRF rates

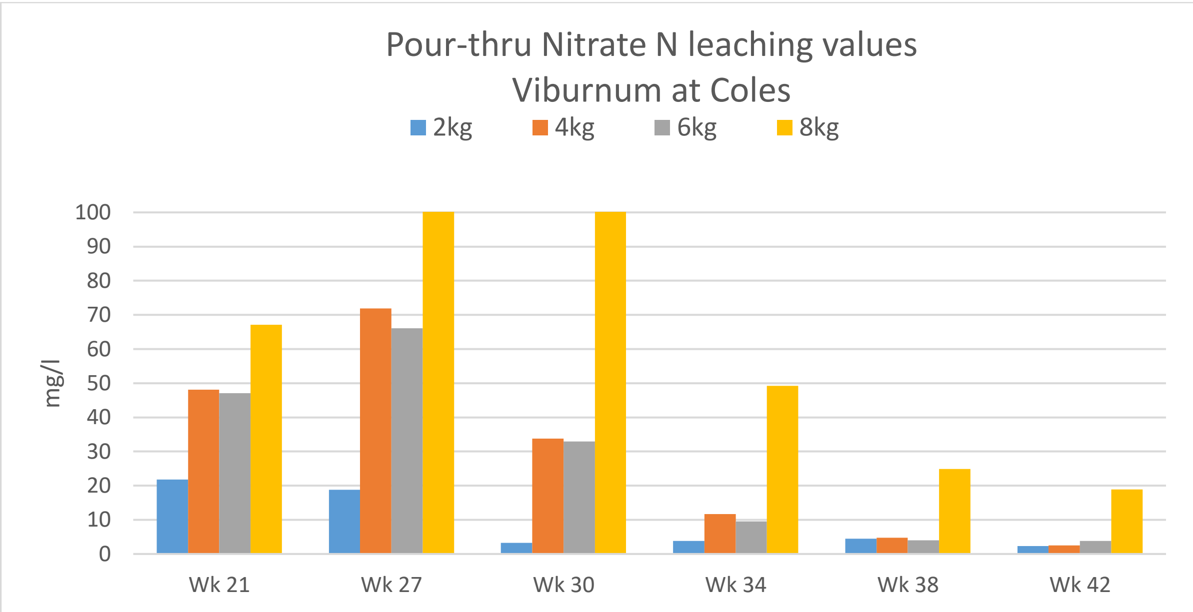


Figure 9 shows nitrate N leaching at Coles for Viburnum across all CRF rates

The leaching of N at all sites was very similar in profile. High leaching rates after potting decreased rapidly (especially with the lower rates of CRF) once the root system was established. The leaching levels of N at the high rate of 8kg/m³ show that this rate is far above that which Viburnum can use.

The increased N leaching that results from trimming back the Buddleja was seen again in the leachate analysis, in this last year of trials.

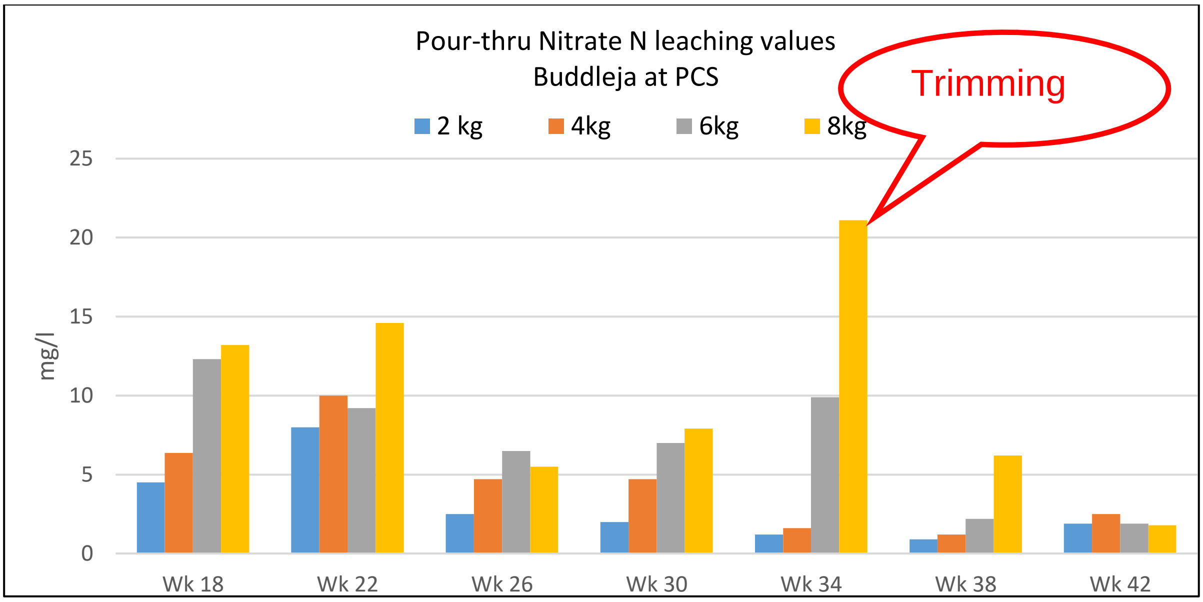


Figure 10 shows nitrate N leaching effects at PCS when trimming Buddleja.

The early high release rates of the CRF resulted in a lack of N availability at the end of the season. The tissue N levels on all four CRF rates is at or below the minimum standard for optimum growth.

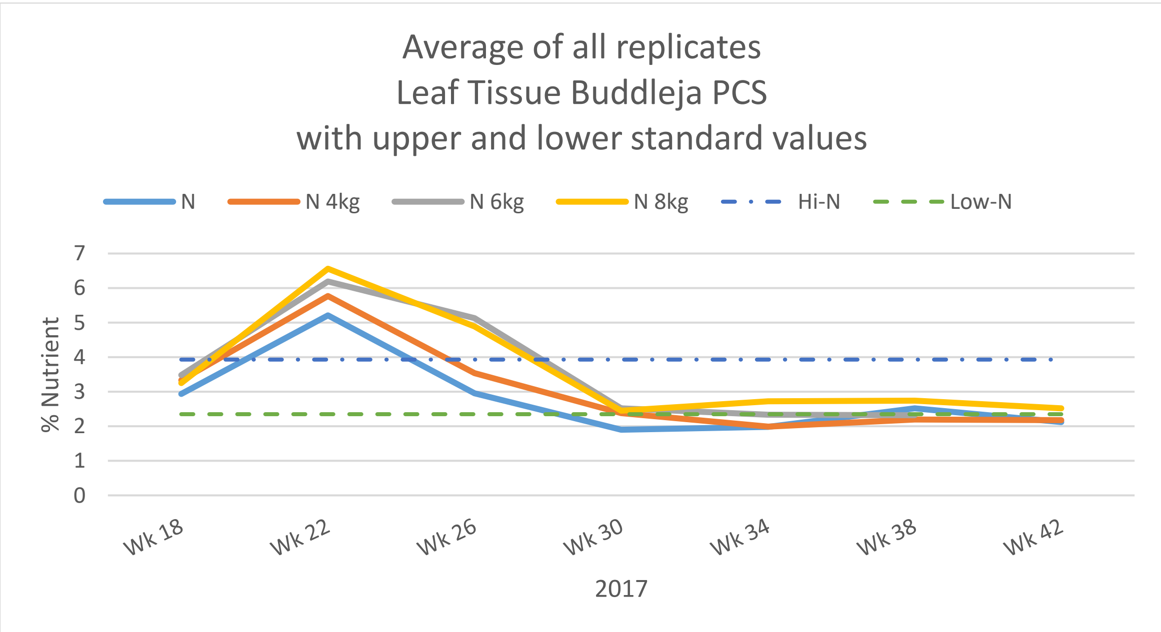


Figure 11 shows tissue levels of N for Buddleja at or below minimum standard levels through much of the growing season.

Equipment ability to register N levels in a crop

atLEAF+ and Apogee MC 100/CCM Chlorophyll measurement

The data collected and shown below indicates the relationship between the device output and the total N levels from the plant tissue analysis which includes all treatments. The trend line shows the resultant relationship between the tissue analysis results and the equipment results. The slope algorithm is shown on the graph. The atLEAF+ device was used at Coles and on the Azalea crop at Osberton. The Apogee MC100/CCM device was used at PCS and Greenmount. The Green Index app and Procheck were used Coles, PCS and Greenmount.

Buddleja and Viburnum have strong, significant relationships between N% tissue and the two device measurements, indicating that is good at monitoring the N% levels. However the Green Index measurements, although significant, vary a lot more between sites. The purple trend line and slope formula shown in each graph represents the average of all the sites.

atLEAF+

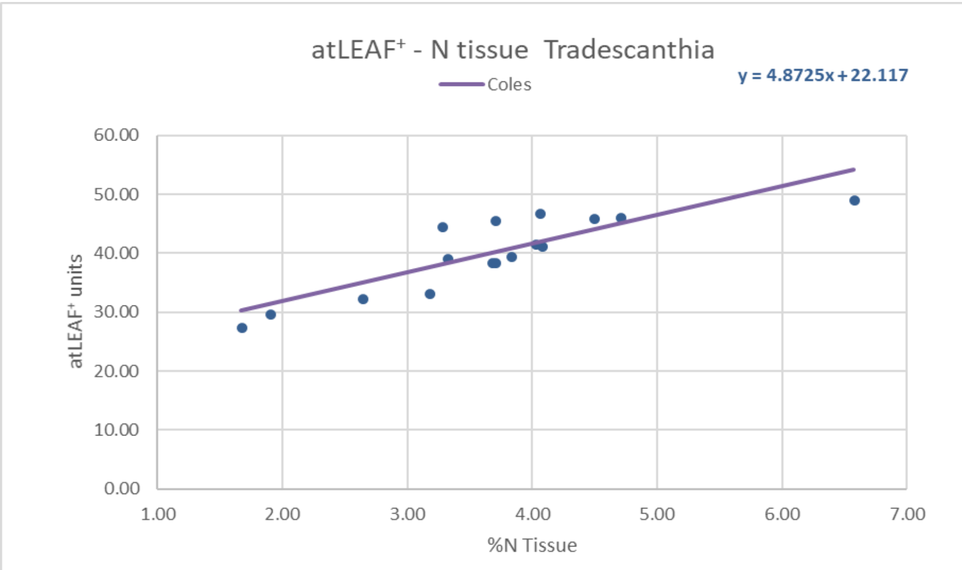


Figure 12 shows the relationship between tissue analysis and atLEAF+ for Tradescanthia p= <0.01

R^2= 44.9

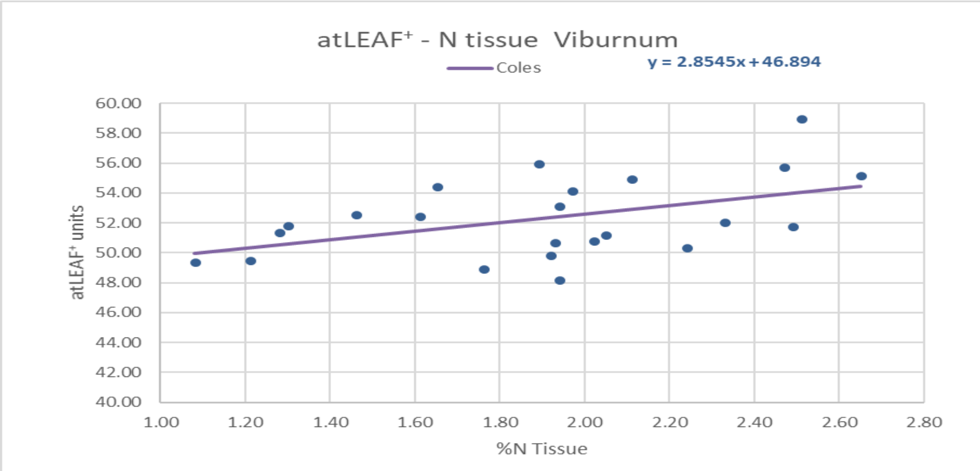


Figure 13 shows the relationship between tissue analysis and atLEAF+ for Viburnum p= <0.05, $R^2 = 47.8$

Apogee/CCM

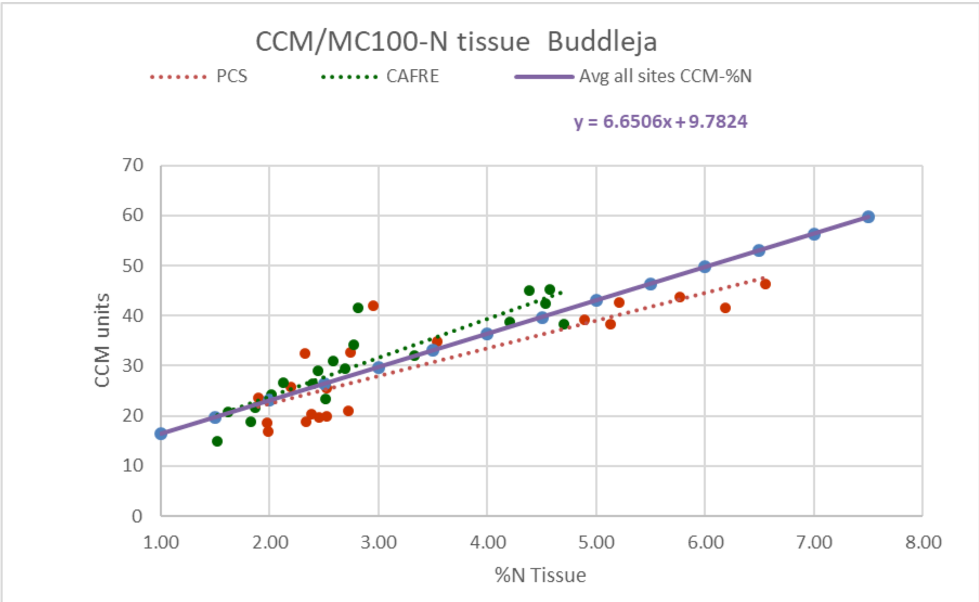


Figure 14 shows the relationship between tissue analysis and Apogee/CCM for Buddleja

p= <0.001, R2 = 82.8

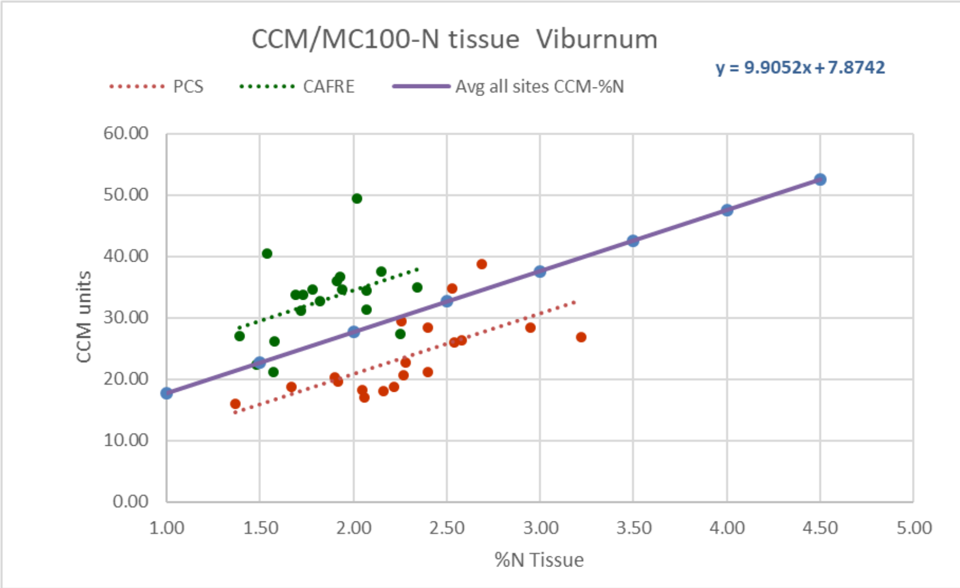


Figure 15 shows the relationship between tissue analysis and Apogee/CCM for Viburnum

p= 0.001, R^2 = 1535.1

Green Index

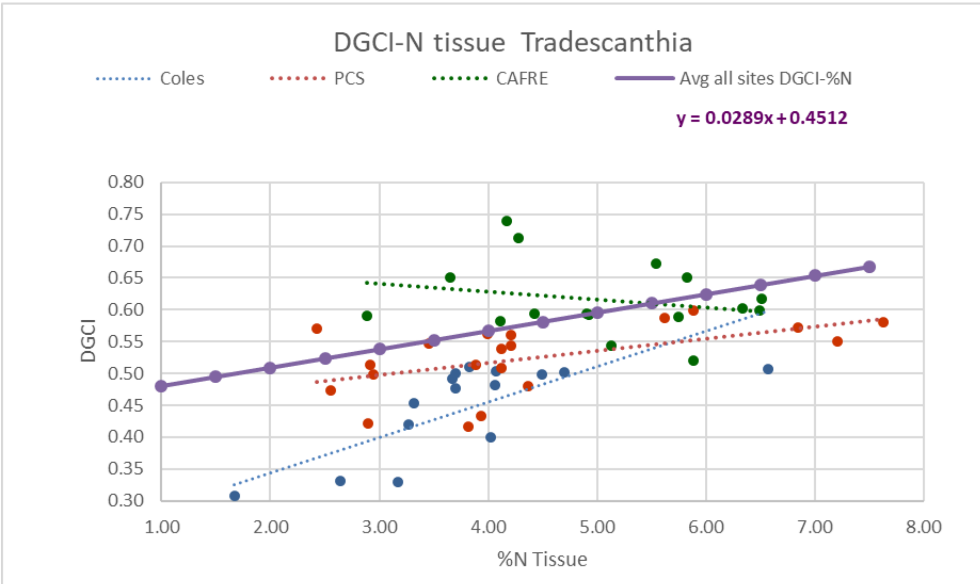


Figure 16 shows the relationship between tissue analysis and Green Index for Tradescanthia

p= <0.001, R^2 = 44.9

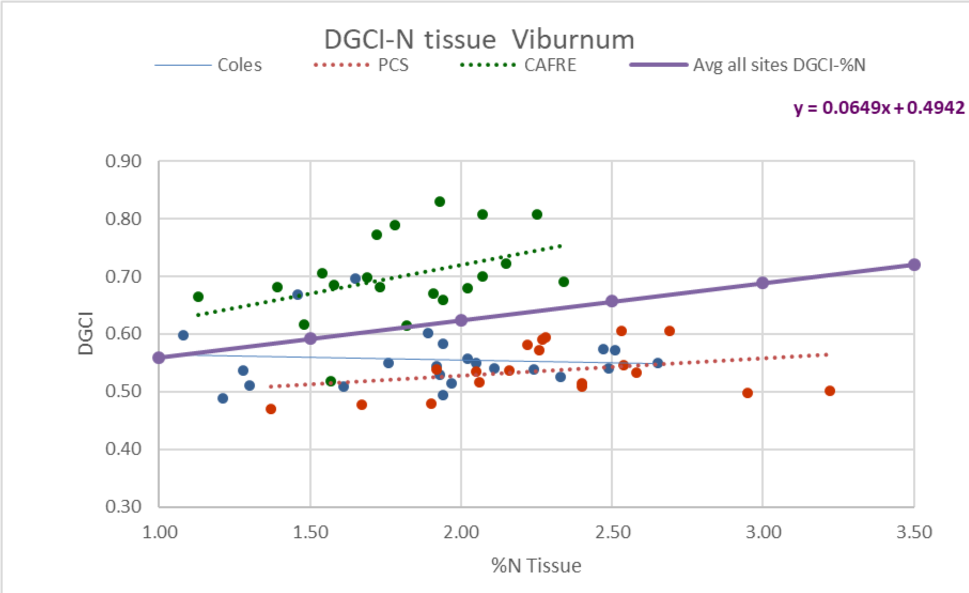


Figure 17 shows the relationship between tissue analysis and Green Index for Viburnum

p= >0.05 (0.84), R^2 = 58.1

Procheck + GS3 sensor

The Procheck device with a GS3 sensor measure the electrical conductivity of the substrate. It is therefore subject to variations in output due to rainfall or irrigation, substrate temperature and plant husbandry operations. This accounts for the greater variation between sites. The significance of the outputs does however show the ability of the equipment to give a good guide to leaf tissue nitrogen levels.

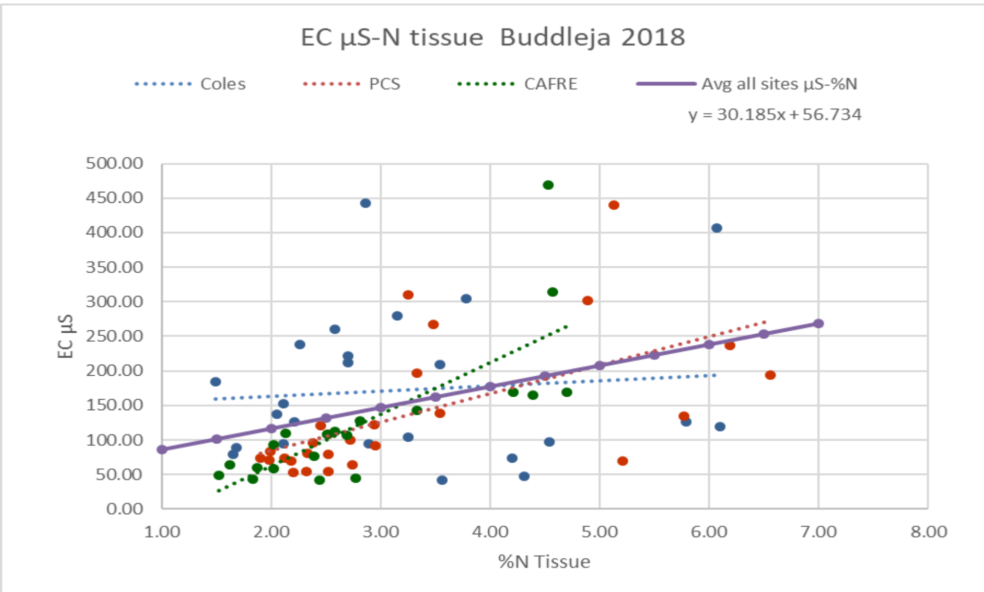


Figure 18 shows the relationship between tissue analysis and Procheck for Buddleja

p= <0.001, R^2 = 34.8

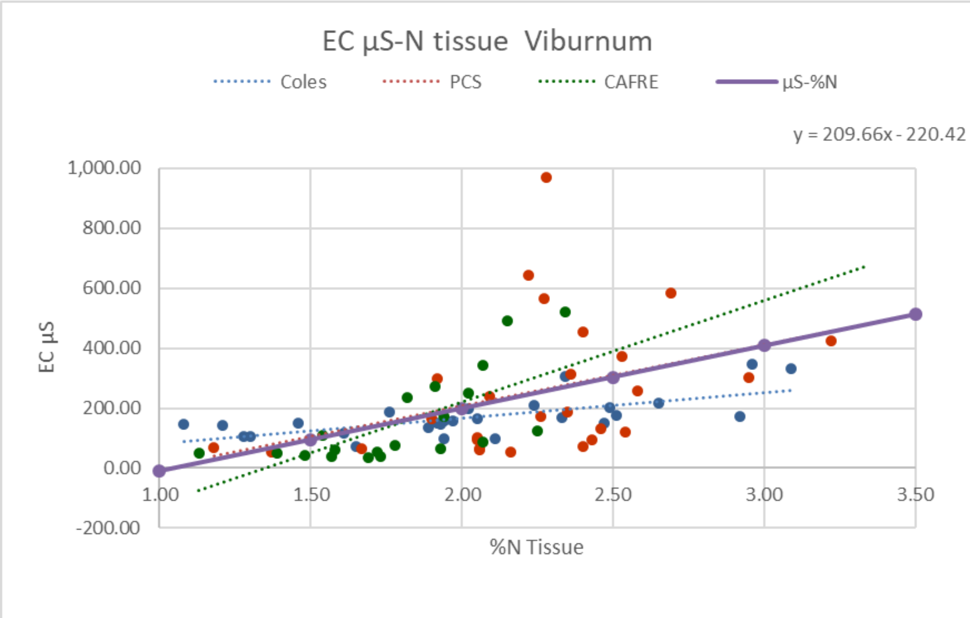


Figure 19 shows the relationship between tissue analysis and Procheck for Viburnum
p= <0.001, R2 = 61.2

Leaching

The leaching of nutrients through the growing season is one important aspect of this project as it shows a plant's ability to assimilate the available nutrients. By demonstrating leaching levels it is possible to ensure greater compliance to ground water regulations, coupled with reduced fertiliser costs. There is a high initial N release into the drainage water at potting which decreases over the season. The crop characteristics show that there is the potential to categorise crops into high, medium and low feeding groups to target nutrient levels and reduce leaching.

In the case of Buddleja, the crop assimilated the N from all four CRF rates and when leaching and tissue levels are compared they show the crop, once the root system is established, would be in the 'high feeding' category. Note the 8kg/m³ spike when trimming took place (Figure 20).

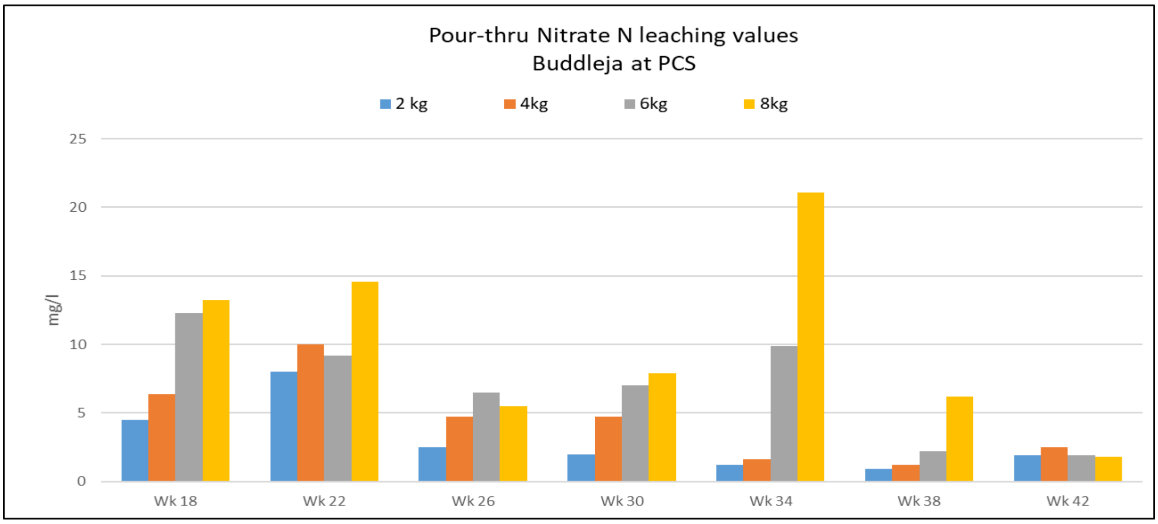
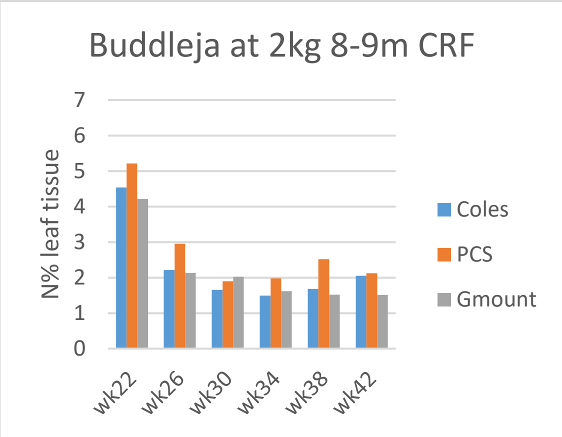
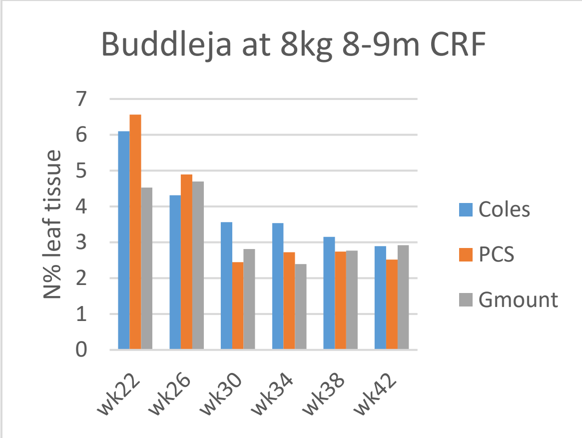


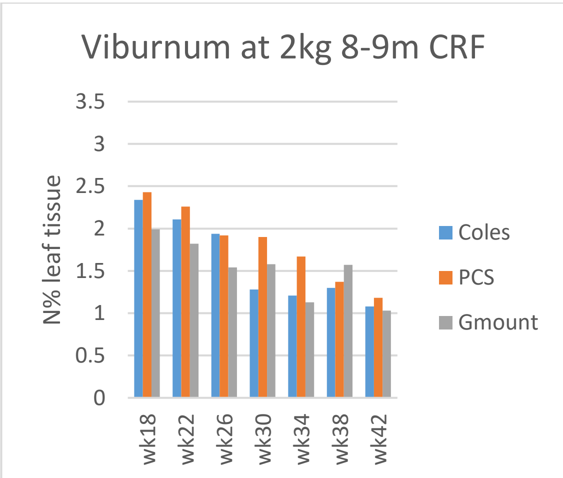
Figure 20 shows the Nitrate N leaching levels for Buddleja

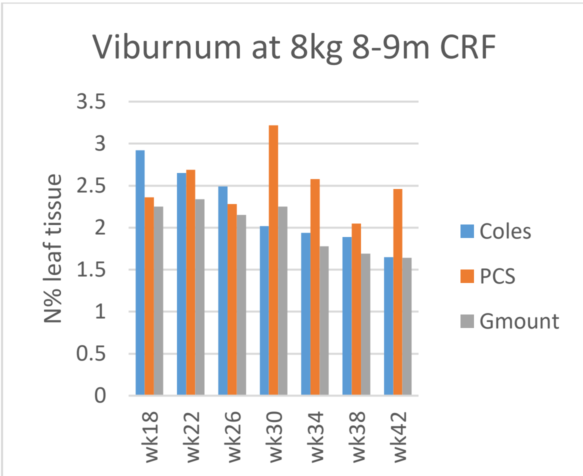




Figures 21 and 22 show N tissue levels for Buddleja with the 8-9 month (m) CRF product

The viburnum crop would be placed in the 'medium feeding' group. The data shows that 2-4kg/m³ is the optimum rate of CRF incorporation for the plant with 8kg/m³ showing no real increase in N uptake or assimilation by the crop (Figures 23 and 24), and a high degree of leaching (Figure 25).





Figures 23 & 24 show N tissue levels – Viburnum with the 8-9 month (m) CRF product

The leaching data shows the high rate of N discharge from the 8kg/m³ CRF rate and the low rate of 2kg/m³. At 2kg/m there is very little leaching of N.

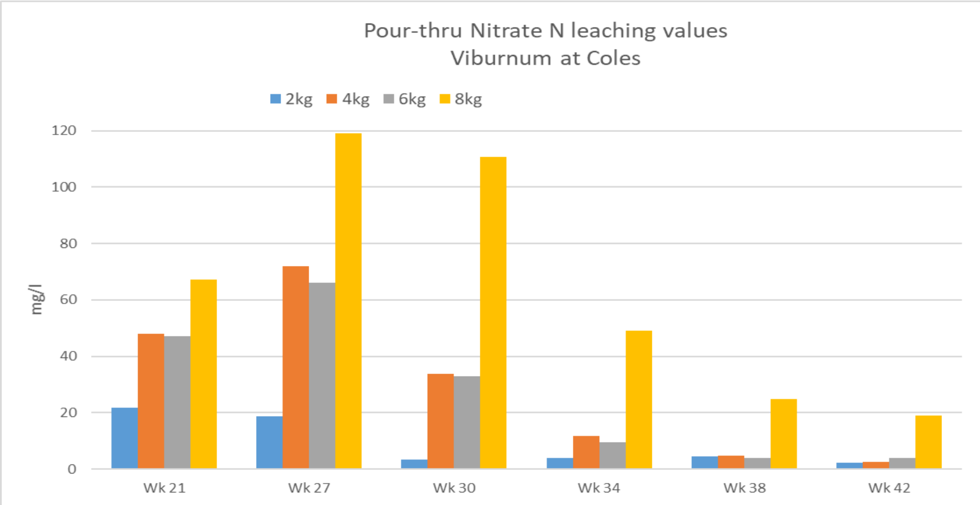


Figure 25 shows N leaching levels for Viburnum

By way of contrast, Tradescanthia assimilated all the nutrients that were available once root establishment had taken place. Even at 8kg/m³ CRF rate the leaching of N was very low. Therefore this crop would be in the 'high feeding' group.

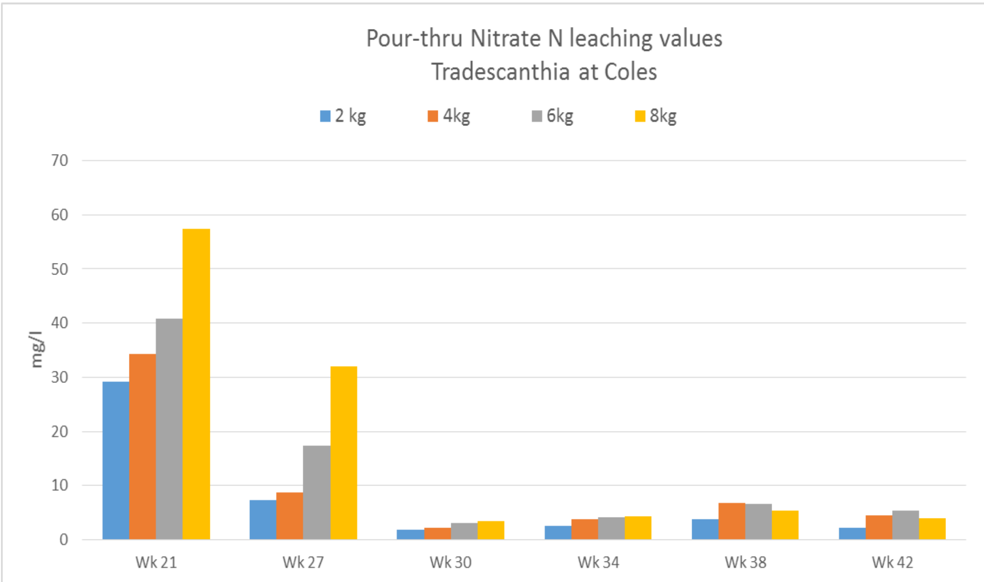
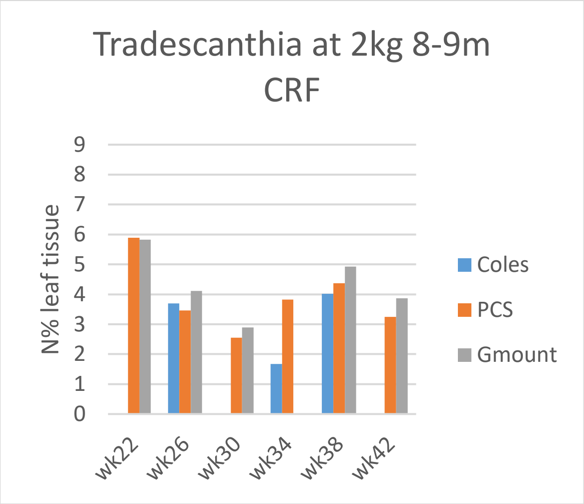
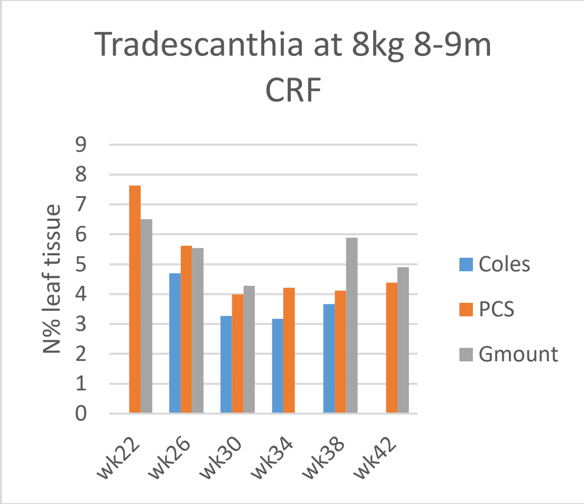


Figure 26 shows N leaching levels for Tradescanthia





Figures 27 and 28 show the difference in tissue N levels between 2kg/m³ and 8kg/m³ is small, showing the ability of the crop to assimilate and retain large quantities of N.

Leachate Nitrogen levels were not closely related to rainfall, air temperature or irrigation.

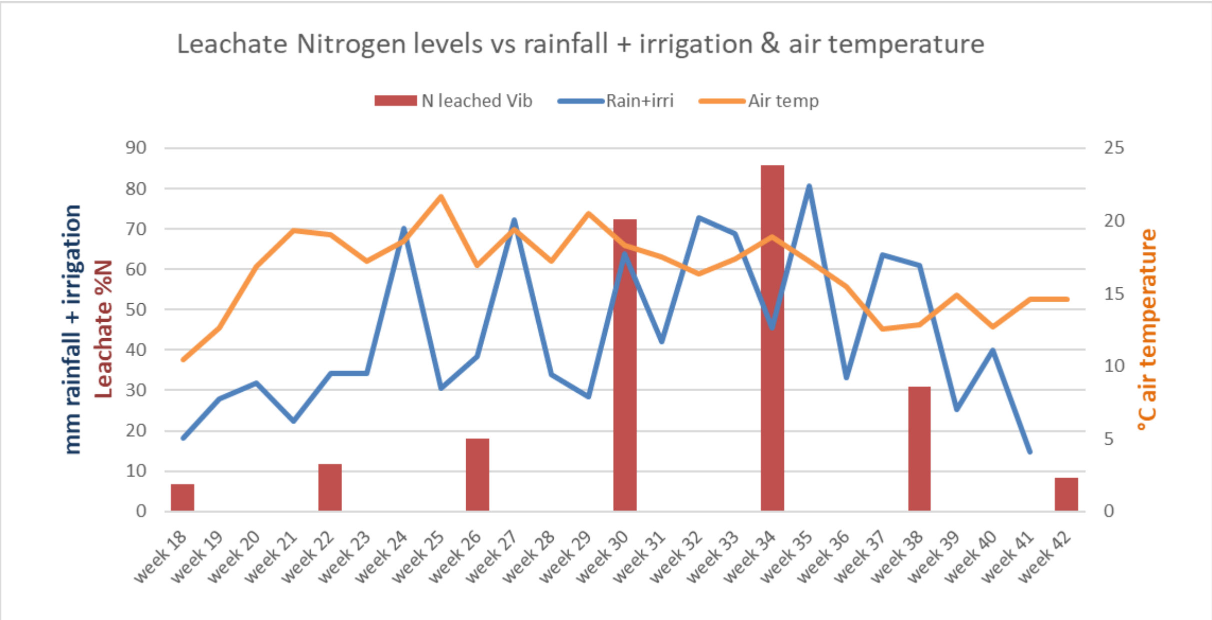


Figure 29 shows very little relationship between rainfall, irrigation levels and air temperature to the average leachate Nitrogen levels of all CRF rates on Viburnum at PCS.

Discussion

The aim of this project was to examine whether it was possible for growers to gauge the nutritional status of plants, using hand-held equipment on-nursery, rather than destructive sampling sent off site to laboratories for analysis. The latter is seen as relatively expensive and is generally only done if there appears to be a problem with plant growth, by which time it may be too late to recover plant quality.

During the first and second years of the project several pieces of sap testing equipment were removed from the trial. Obtaining sap for the tests was difficult on nursery stock subjects. The leaf tissue did not yield sufficient sap volume and was too discoloured. This made the colour checks of the Merck Nitrate test strips difficult to achieve. The Horiba Laquatwin Nitrate kit gave suitable output figures for nitrate content when using a filter process but the procedure necessary to obtain a reading was not suitable for nursery use and was more suited to laboratory conditions.

Several pieces of equipment tested in the second year of the project showed that doing real-time sampling of plant material does have the potential to give growers a quantitative record of nutrient status before the eye can see differences. In year three the most suitable pieces of equipment identified by the previous years' work showed their ability to indicate tissue N level trends. The best tools to use on the nursery were deemed to be the spectral reflectance equipment (atLEAF+ and CCM100) and the Pro-check equipment. The use of spectral reflectance to measure crop nutritional status is being rapidly developed worldwide.

What was evident from our data was that measurement taken are unique to a site, and that it is not possible to cross reference N levels between sites. When taking a series of readings on a site and studying the trends it easily identifies N levels rising, falling or remaining static. From that a grower can regulate nutrient levels accordingly.

Taking regular readings is necessary to identify the trends early as the equipment can show N levels before any visual symptoms are seen in the crop, and nutrient levels can be corrected before the crop suffers. However, the caution here is that to get consistent data to see these trends, a nursery should dedicate one person to such data collection - this may be the same person who looks after pest and disease monitoring for the nursery.

The basis of 'one size fits all' regarding CRF incorporation rates was shown in this project to be unsatisfactory, as shown by the different assimilation characteristics of the three crops monitored. The Buddleja crop could be categorised as a gross feeder and performed well even at 8kg/m^3. The Viburnum, however, is very much the opposite and would perform with 2-4 kg/m^3. This shows that crops do need to be categorised into different feeding groups; low, medium and high feeding requirements.

The high levels of N leached at the time of potting which did not decrease until the root system establishes in the pot, may be reduced by a high phosphate liquid feed 2-3 weeks prior to potting to stimulate root activity in advance (Hiron, 1985; Ericasson and Ingestad, 1988).

The equipment we have used is constantly being updated. The new atLEAF+ device has an updated number of readings that can be stored, with new software and includes analytical tools to display plant parameters as graphs. The Procheck device is currently undergoing the development of a mobile phone link for easier data transfer and storage.

Conclusions

- Chlorophyll measuring devices can provide an indication of nitrogen levels in the tissue; this ability declines as senescence increases through the season.
- Substrate levels of controlled release fertiliser (CRF) need to be regulated to reduce levels of leached nutrients. ‘One size’ does not fit all.
- Choosing a responsive crop to act as the monitor plant enables easy leaf measurement.
- Variegated and hairy foliage crops can be difficult to monitor.
- Plants need to be categorised into feeding level groups of “High”, “Medium” and “Low” according to their rate of nutrient assimilation and leaching.
- For early spring growth, chlorophyll measuring systems can detect reducing nitrogen levels in plant leaf tissue before it is detectable by eye when using LED technology.
- The low level of nutrients at the end of the season may be due in part to the initial high release rate.
- Frequent measurements need to be taken with the chlorophyll and conductivity systems to achieve regular monitoring of crop nutrient status.
- Air temperature has a greater effect in leachate Nitrogen levels than rainfall and irrigation.

Knowledge and Technology Transfer

A stand was taken at the Four Oaks Show and the equipment was demonstrated to growers who visited the stand. A Master Class on Nutrition at Chichester as part of the two day GroSouth event gave delegates “Hands on” time with the equipment where they could try the various systems available. At a Growing Media Workshop at F P Matthews, the equipment was demonstrated across a range of different growing media.

A two day workshop and nursery tour is planned for November 2018 in Belgium which will include a presentation of similar work carried out in Germany, leaching work being carried out on a German research station as well as reports on CP 095: Sustainable resource use in horticulture and the FERTINNOWA Project.

References

Ericsson, T and Ingestad, T 1988 Nutrition and growth of birch seedlings at varied relative phosphorus addition rates, Physiologia plantarum 72: 227-235

Hiron, B at Kirton ADAS Leaflet page 909 (1985)

Mills, H.A. And Benton Jones, J. 1996. Plant analysis Handbook II: A Practical Sampling, Preparation, Analysis and Interpretation Guide. Micromacro Publishing Inc., Athens.

Appendices

Appendix 1 - Equipment data

atLEAF+

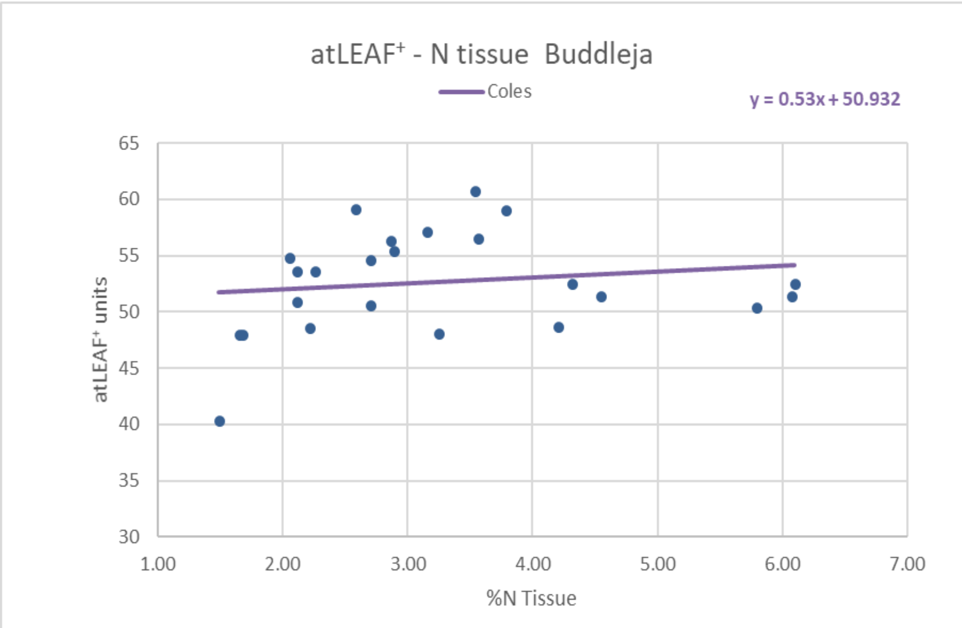


Figure 30 shows the relationship between tissue analysis and atLEAF+ for Buddleja p= >0.05 (0.41), R^2 = 27.9

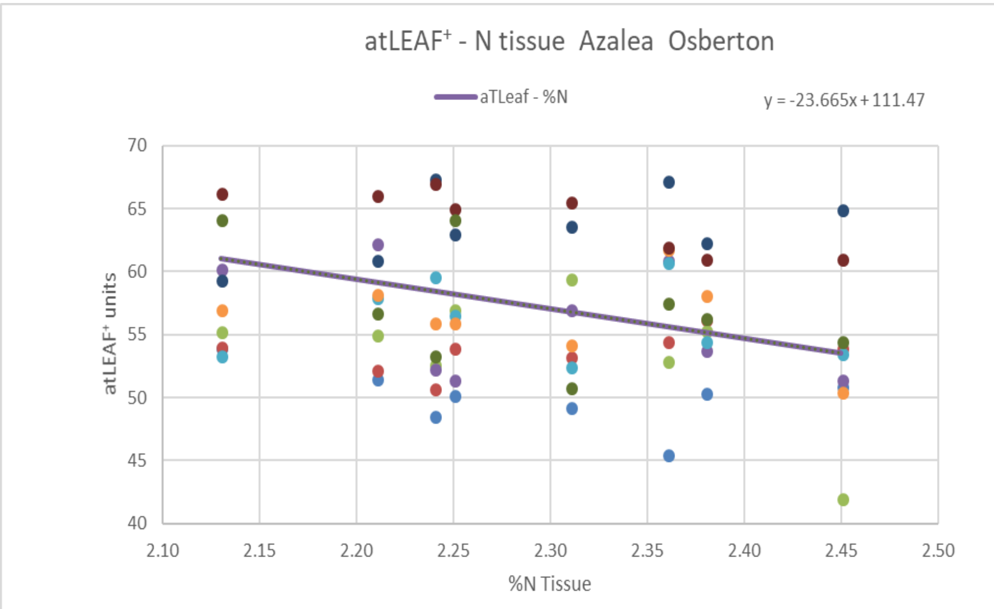


Figure 31 shows the relationship between tissue analysis and atLEAF+ for Azalea

The results of the Azalea were not as expected with the atLEAF+ device and the device readings did not follow the tissue analysis as the season progressed. The data was very erratic which would seem to be the result of readings taken from a very hairy leaf, which may have affected the closing of the device's aperture, causing daylight to interfere with the reading. When tissue was related to readings there was a negative result, though this was not significant, $p>0.05$ (0.193)

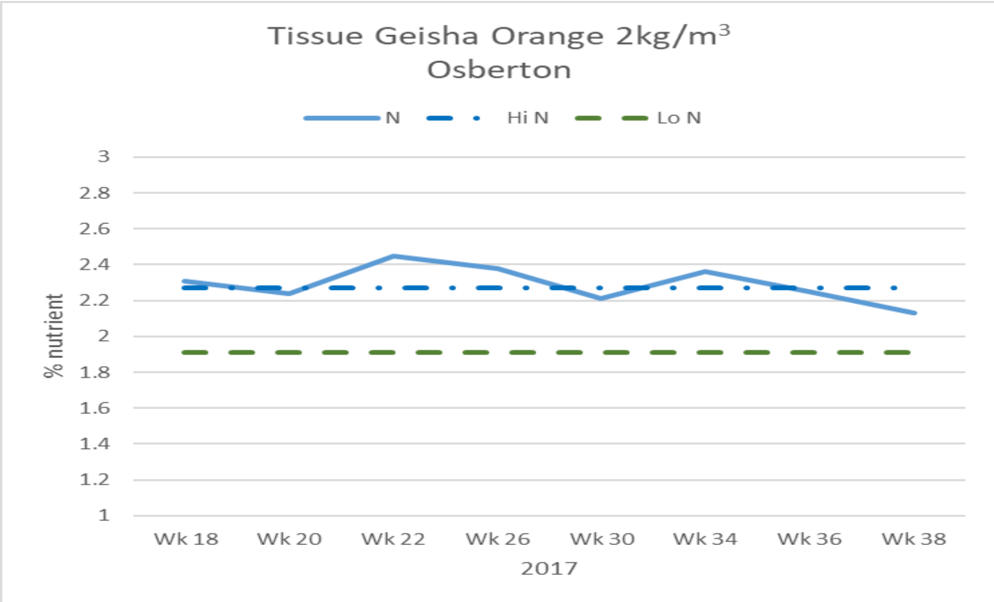


Figure 32 shows the Tissue N% over the trial period and standard levels for Azalea

CCM/MC100

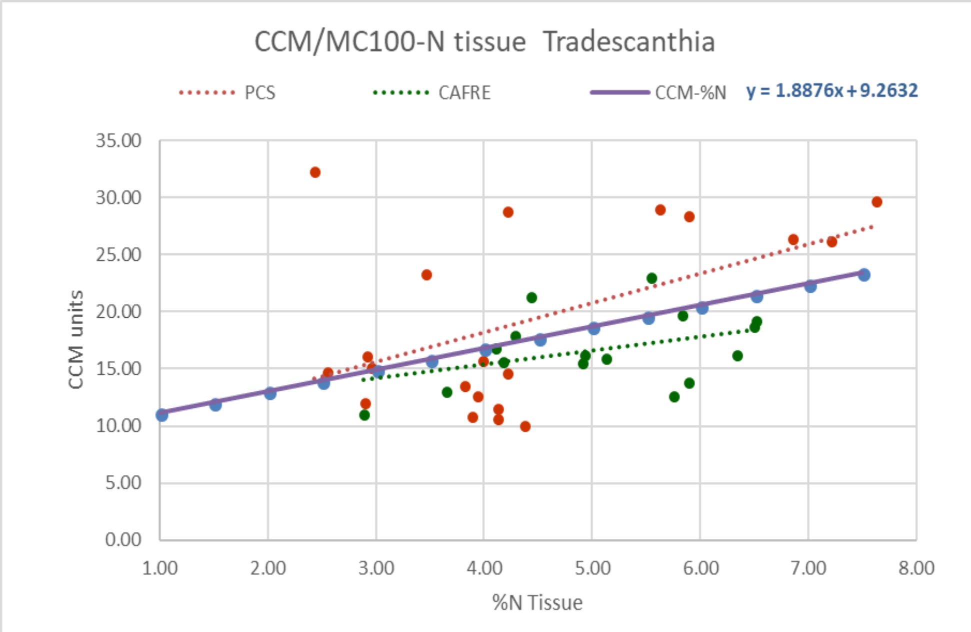


Figure 33 shows the relationship between tissue analysis and Apogee/CCM for Tradescanthia
p= <0.05, R^2 = 43.2

Green Index

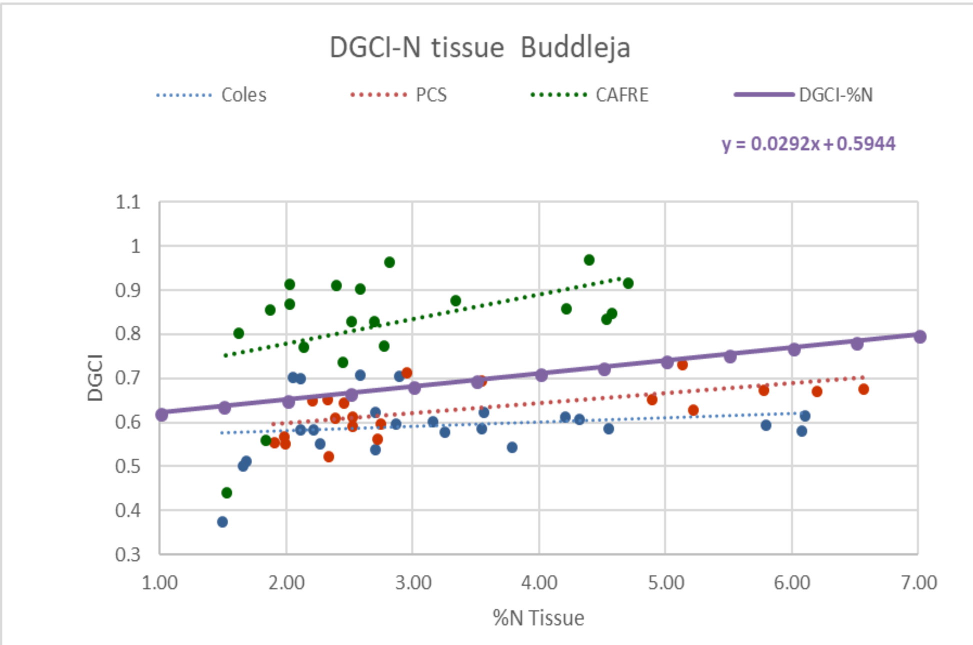


Figure 34 shows the relationship between tissue analysis and Green Index for Buddleja
p= >0.05 (0.307), R^2 = 33.5

Procheck

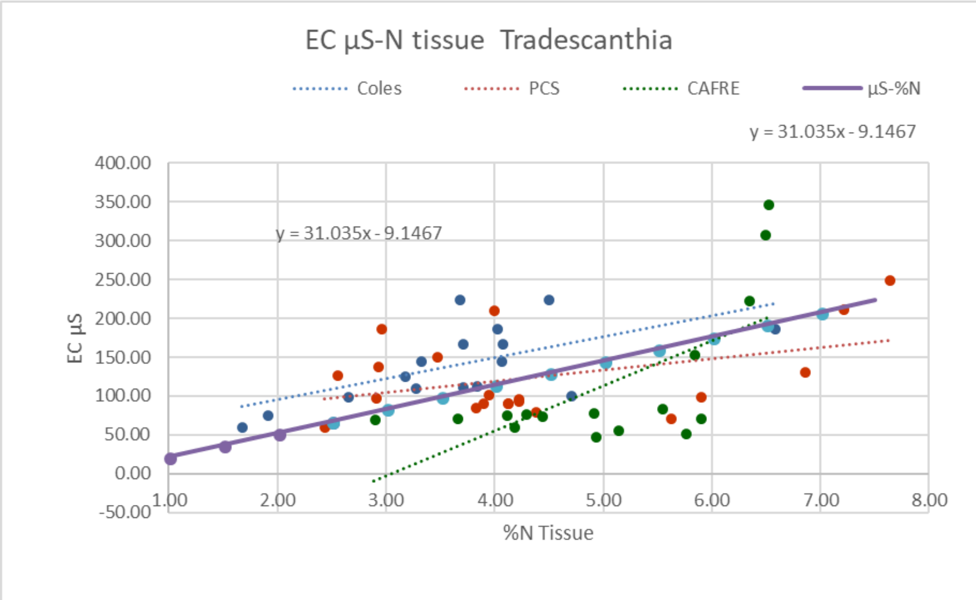


Figure 35 shows the relationship between tissue analysis and Procheck for Tradescanthia *p*= >0.05 (0.066), R^2 = 44.4

Appendix 2 – Weather data

All the sites each collect weather data on a half hourly basis. The data recorded for the trial purposes are average daily air temperature and rainfall. The comparisons between all of the sites is shown in figures 36 and 37 below.

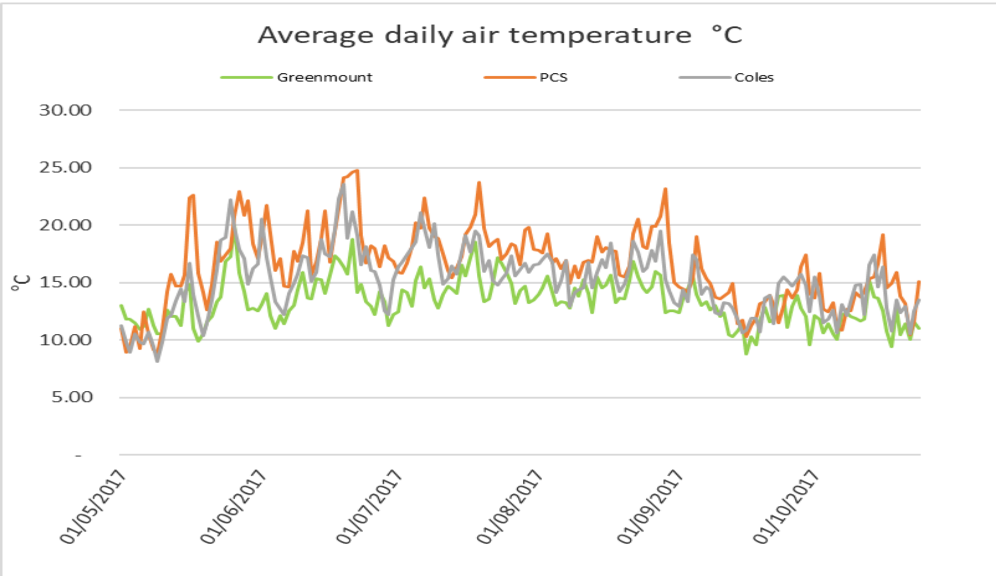


Figure 36 shows the average daily air temperatures across all 3 sites

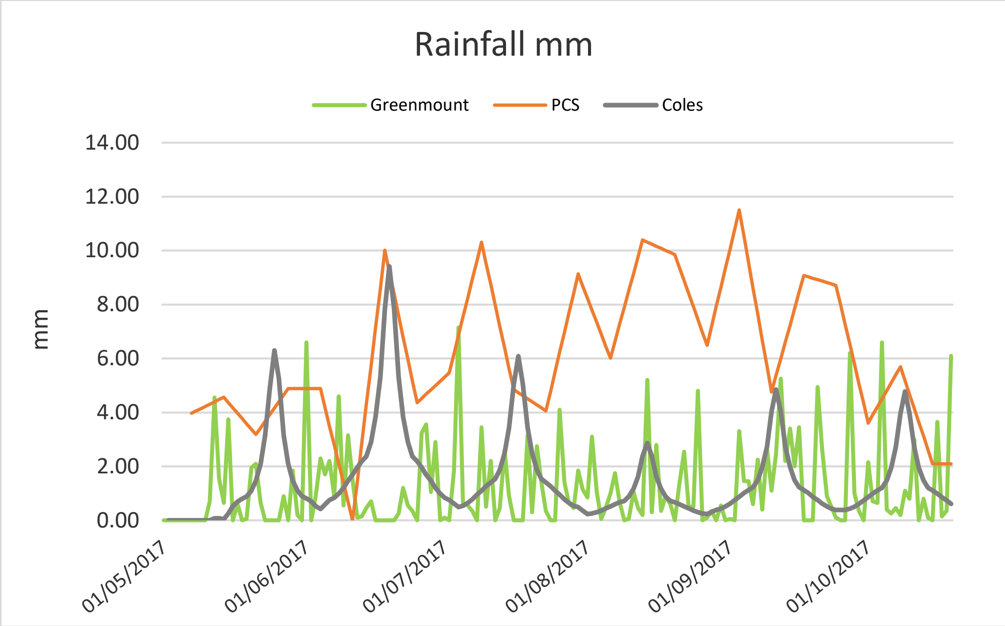


Figure 37 shows the monthly rainfall across all 3 sites