Effects of Harvest Maturity on Extra Virgin Olive Oil Year 3 – SFF Project #07/1

Requejo-Jackman C, Farrell M, Olsson S, Ogwaro J, Beresford M, Harker R, McGhie T, Blanche A, Fara E, Wohlers M, Farr J, Wibisono R, Wang Y, Wong M, Fan C, and Woolf A

March 2011

A report prepared for:

Olives New Zealand

Requejo-Jackman C, Farrell M, Olsson S, Beresford M, Harker R, Wohlers M, Farr J, Wibisono R, and Woolf A.

Plant & Food Research, Auckland

McGhie T.

Plant & Food Research, Palmerston North

Ogwaro J, Wang Y, Fan C and Wong M.

Institute for Food, Nutrition and Human Health, Massey University

SPTS No. 5244

Contents

Executive summary

The year 2010 constitutes the third and final year of this research examining olives grown in New Zealand. The primary aim was to develop new locally appropriate measures of assessing the maturity of New Zealand fruit. A secondary aim was to collect a robust range of data on phytochemicals in the oil of cultivars from most growing regions in New Zealand. With this purpose, in this last year of research the phenolic composition was monitored to examine how it changed as fruit matured in different growing environments throughout New Zealand.

Fruit samples were received from orchards in Northland, Auckland, Waiheke, Hawke's Bay, Wairarapa, Nelson, Marlborough and Otago. Fruit were collected by the growers at several stages of maturity (usually two to three weeks apart) from the small very immature green stage through to the black stage of fruit coloration (from February/March to July 2010). The aim was to determine the impact of progression and timing of fruit maturation harvested from different regions on fruit characteristics at harvest, and on the chemical profiles of the oil.

Physical measures such as dry matter, fruit weight and colour maturity index were performed on the fruit immediately after harvest. Oils were obtained by the accelerated solvent extraction (ASE) method and analysed for changes in total phenolics over the harvest period. Selected oils from harvests close to commercial harvest dates for each grower were also analysed for fatty acid, tocopherol and sterol compositions.

In summary,

- As fruit matured, there was a progressive accumulation of oil with a levelling off later in the season in most cultivars and growing locations. Oil accumulated at different rates between regions, with the earliest maturing orchard in the Auckland region reaching maximum oil content six weeks earlier than the latest maturing orchard in the Otago region.

- There was significant variation in the oil content of olives between cultivars, growing regions, and growers within a region. In general, orchards in the northern regions (Northland and Auckland) developed fruit with higher oil content than those in the southern regions. The maximum oil content (on a dry basis) for 'Frantoio' and 'Leccino' ranged from approximately 28% to 48%, and 'Koroneiki' ranged from approximately 32 to 52%. There was much less variability in oil content for the orchards in the South Island.

- The 2010 data confirmed that total solids per fruit (expressed in grams) is a better measure than % dry matter content and shows strong correlations with % oil content and fruit maturity. This simple tool can be used by growers to track oil accumulation over the season and make decisions on harvest timing.

- There were poor correlations between oil content and the colour maturity index for the North and South Island orchards, especially for 'Frantoio' for the data combined for the whole country.

- Total phenolics contents measured in the oil decreased with maturity and it was observed that the incidence of rain at the time of harvest could also influence this rate of decline.

- The dry matter measured as a percentage did not relate well with (%) oil content, probably because of differences in water content of the fruit grown in the different climates and the influence of rainfall. It was observed that % dry matter values had different correlations with oil content was region-dependent.

- Variability in climates and microclimates throughout New Zealand has influence on the olive fruit development and maturation rates. Thus, fruit maturity is achieved at different times during the season depending on the orchard location, but normally earliest in the northern regions and latest in the South Island regions.

- In general, fruit weights tended to be largest in the northern regions and decreased as the regions progressed south.

- Cultivar differences were found in the different cultivars in terms of sterols. Oil extracted from 'Barnea' was found to contain the highest concentration of plant sterols - between 1402 and 1873 mg/kg, while 'Koroneiki' contained the least 685 – 1114 mg/kg. In the ASE extracted oils, the campesterol present was found to be greater than 4% in both 'Barnea' and 'Koroneiki' oils. This is consistent with reports from other olive oil producers. The ASE-extracted oils from the other cultivars had sterol percentages within the international standards for olive oil.

- No regional differences were seen with the tocopherol (Vitamin E) composition in the oils, but differences were observed in the cultivars. 'Frantoio' and 'J5' oil had the lowest concentrations of tocopherols, 155 – 290 mg/kg, while the other cultivars analysed had concentrations ranging from 306 to 758 mg/kg.

This three-year study has provided extensive information on New Zealand olives in terms of changes in maturity and the chemical makeup of the oils, particularly from the last season. The range of techniques examined for measuring maturity led to the recommendation that two are most appropriate for use by growers and/or processors – total solids per fruit (g) and fruit firmness. The information obtained is very useful for a number of applications, including development of quality standards, access to markets, and maximising oil yield, quality, stability and flavour. Results obtained here in New Zealand have shown significant differences from those of the Mediterranean and Australian olive industries.

For further information please contact:

Cecilia Requejo-Jackman
The New Zealand Institute for Plant & Food Research Ltd
Plant & Food Research Auckland
Private Bag 92 169
Auckland Mail Centre
Auckland 1142
NEW ZEALAND
Tel: +64-9-925 7000
Fax: +64-9-925 7001
Email crequejo-jackman@plantandfood.co.nz

1   Background

This project was undertaken with the aim of finding a maturity index for producing extra virgin olive oil of consistent quality. In addition, this project also provided an opportunity to collect baseline information on the variability in oil composition, such as fatty acids, polyphenols and other health-promoting compounds (such as tocopherols and sterols), of which there was a dearth of information for New Zealand-grown olives. Over the last two years we have studied the maturity of the olive fruit from different cultivars grown in different New Zealand regions to examine the relationship between maturity and extra virgin olive oil characteristics. In the first year, 2008, the study concentrated on the three main cultivars grown in New Zealand, 'Koroneiki', 'Leccino' and 'Frantoio'. These were harvested at three stages of maturity from four orchards within the greater Auckland region. The fruit were collected at three nominated harvest maturities, early, mid and late harvest, to determine the effect of harvest maturity on fruit characteristics, as well as on the physical, chemical and sensory profiles of the oil. In the second year of studies, 2009, the research focused on the 'Frantoio' cultivar, produced by two orchards in two different regions: South Auckland (Simunovich Olive Estate) and Hawke's Bay (Matapiro Olive Estate). Similarly, fruit were collected at several stages of maturity. Physical measurements such as dry matter, fruit weight, colour maturity index and firmness were performed on the fruit immediately after harvest. Oils were obtained by the accelerated solvent extraction (ASE) method and by the cold pressed method. The oil obtained was analysed for chemical quality measures as well as fatty acid composition, quantification of tocopherols, total phenolics and antioxidants.

Among other important results, the data showed that the amount of oil and its composition depended on the maturity stage of the fruit. Determination of optimal time of harvest to produce 'high quality oil', therefore, related to the grower's goal, e.g. maximise oil yield or oil with specific and distinct flavour characteristics. However, oil content determination is not easy and it certainly cannot be done in the orchard. Subsequent investigations, therefore, have focused on finding alternative fruit measures that correlate with oil quantity and quality that can be used in the field to assist with the harvest decision making.

Two fruit variables (fruit firmness and total solids (dry matter on a per fruit basis)) were correlated to oil content in the 2008 and 2009 studies. It was proposed that these two measures could be used to predict oil content and may give an indication of phenolic composition at the time of harvest. Firmness and total solids can be relatively easily and rapidly measured and measurement of total solids in the fruit is viable and applicable by growers. However, there is currently no economically feasible instrument to measure fruit firmness in the field – thus fruit firmness was not examined during 2010 season.

2   Introduction

Olive oil from New Zealand is recognised overseas for its unique organoleptic qualities. The organoleptic qualities of olive oil are usually described through their balance in fruity, bitter and pungent flavours. Extensive research has shown that phenolics are the chemical compounds responsible for the pungency and bitterness levels in oil (Beltrán et al., 2007; Gawel & Rogers, 2009; Mateos et al., 2004; Skevin et al., 2003). In addition, phenolics are closely related to oil stability and therefore crucial for its shelf life (Ayton et al., 2007; Baccouri et al., 2008; Ben Temime et al., 2006; Cinquanta et al., 2001; Gutiérrez et al., 2001; Kiralan et al., 2008; Tsimidou et al., 1992). Thus, finding out how phenolics change with fruit maturity is important to

assist with the harvest time decision for New Zealand growers and to achieve a high quality product in terms of both flavour and longevity.

In the northern hemisphere, olives are grown in hot and dry climates where the optimum time of harvest in terms of oil content and oil flavour is estimated using the colour maturity index. In New Zealand, olives are grown in 12 regions throughout the country, from Northland to Central Otago, each with very different microclimates, ranging from dry to wet and cool to warm environments. The colour maturity index is a visual and very subjective measure. Our two previous years of research have shown that while the maturity index can be a good indicator of maturity development in warm dry climates in New Zealand, it is not reliable in cooler climates. This has been demonstrated particularly in areas where there are shorter summers and cool autumns or for cultivars that mature late in the season when temperature are cool. In this third and last year of research we examined how the oil characteristics change as fruit mature in different growing environments throughout New Zealand. We expanded the sampling area to cover most growing regions throughout New Zealand. Samples of fruit were received from orchards in Northland, Auckland, Waiheke, Hawke's Bay, Wairarapa, Nelson, Marlborough and Otago. Samples of fruit were collected by the growers at several stages of maturity (usually two to three weeks apart), starting at the small and very green stage (i.e. well before commercial harvest) through to the black stage of fruit coloration. This determined the progression and timing of fruit maturation in these different regions and its impact on at-harvest fruit characteristics and the chemical profiles of extracted oil.

3   Aim

Investigate the characteristics of olive fruit and extracted oil quality for the three most widely grown cultivars ('Frantoio', 'Leccino' and 'Koroneiki'), and a range of other cultivars (such as 'Picual', 'Barnea' and 'J5'), over a wide distribution of growing areas in New Zealand:

The patterns of changes in fruit maturity included:
- Colour maturity index (external and internal colour)
- Dry matter
- Oil content
- Fruit weight/size
- And the corresponding changes in polyphenols in the extracted oils (monitored over three harvest dates.

Olive oil characteristics included important phytochemicals (potentially healthful compounds):
- Fatty acids
- Sterols
- Tocopherols.

4 Materials and Methods

4.1 Olive fruit sample collection

Olive (*Olea europaea*) samples were obtained from 20 commercial orchards from eight olive growing regions of New Zealand (Figure 1). In this study, fruit were harvested from all regions except Kapiti and Canterbury (Table 1).

Figure 1. Map of the main olive growing areas of New Zealand. In this study, fruit were harvested from all regions except Kapiti and Canterbury (see Table 1 for orchards and regions).

At the start of the season, olive growers randomly selected and tagged three replicates of trees per cultivar (trees at least 8 years old), each replicate comprising three trees with similar tree structure and fruit loads. Fruit were harvested from these same sets of trees throughout the trial period. Six different cultivars were studied: 'Frantoio', 'Leccino', 'Koroneiki', 'Picual',' Barnea' and 'J5', although most effort was made to obtain fruit from the first three cultivars.

Between 300 and 1000 g of olives (depending on the time in the season) were sampled from each replicate every two weeks, at least eight times during the growing season (from as early as February through to late harvest, sometimes July 2010). The fruit samples, in plastic bags were couriered overnight to Plant & Food Research (PFR) laboratories located in Mt Albert, Auckland. Fruit from the Auckland region were harvested into plastic bags, by the PFR team, and directly transported back to the PFR laboratory.

A summary of the orchards' locations, orchard names and cultivars sampled is found in Table 1.

Table 1. Olive cultivars, location and orchard sampled during 2010.

SUMMARY OF SAMPLES COLLECTED DURING the 2010 SEASON

Region/Orchard/Cultivar	'Frantoio'	'Leccino'	'Koroneiki'	'Barnea'	'Picual'	'J5'
Northland						
Quail Ridge Olives		X	X			X
Toru Waiwai	X	X	X			
Auckland						
River Estate	X		X			
Simunovich Olive Estate	X				X	X
Armour	X	X	X			
Waiheke						
Matiatia Olive Estate	X	X				
Watch Dog			X			
Putiki		X			X	
Hawke's Bay						
The Village Press	X	X		X	X	
Matapiro Estate	X	X		X		
Waimarama				X		
Wairarapa						
Olea Estate	X	X			X	
Olivo				X		
Stone Valley			X			
Nelson						
Nelson EPF	X	X				
Neurdorf Olives	X					
Tasman Bay	X	X			X	
Marlborough						
Lusatori Olive Grove		X	X			
Otago						
Poverty Gully	X	X				
Cainmuir Olives	X	X				

4.2 Processing of the Olives

Each of the samples of olives received was processed in the steps shown in the following diagram (Figure 2).

```
┌─────────────────────────────────────────┐
│        Reception of olive fruit          │
└─────────────────────────────────────────┘
                    │
                    ▼
┌─────────────────────────────────────────┐
│  Weigh 100 fruit per sample (3 replicates),│
│           Rate maturity index            │
└─────────────────────────────────────────┘
                    │
                    ▼
┌─────────────────────────────────────────┐
│  Mechanical grinding of 200 g whole fruit (3│
│      replicates) to give olive paste     │
└─────────────────────────────────────────┘
                    │
                    ▼
┌─────────────────────────────────────────┐      ┌──────────────────────────────────┐
│  Liquid N₂ + sealed (vacuum sealer) in vacuum│─────▶│ *Aim* : Replacing O₂ delays       │
│  bag flushed with nitrogen (oxygen free) gas │      │ enzyme activity  and  oil         │
└─────────────────────────────────────────┘      │ deterioration                     │
                    │                            └──────────────────────────────────┘
                    ▼
┌─────────────────────────────────────────┐      ┌──────────────────────────────────┐
│  Storage -80°C freezer (minimum 24 h)    │─────▶│ *Aim* : Minimize degradation      │
└─────────────────────────────────────────┘      └──────────────────────────────────┘
                    │
                    ▼
┌─────────────────────────────────────────┐      ┌──────────────────────────────────┐
│  Freeze dry = lyophilization (~3 days) = % dry│─────▶│ *Aim* : Removal of water by       │
│        matter calculation                │      │ sublimation to preserve oil       │
└─────────────────────────────────────────┘      │ qualities.                        │
                    │                            └──────────────────────────────────┘
                    ▼
┌─────────────────────────────────────────┐
│     Seal in foil bags (vacuum sealed)    │
└─────────────────────────────────────────┘
                    │
                    ▼
┌─────────────────────────────────────────┐      ┌──────────────────────────────────┐
│   Accelerated Solvent Extractor (ASE)    │─────▶│ *Aim* : Extraction of oil under repeatable│
└─────────────────────────────────────────┘      │ and controlled conditions         │
                    │                            └──────────────────────────────────┘
                    ▼
┌─────────────────────────────────────────┐      ┌──────────────────────────────────┐
│  Rapid Vap to recover the oil (using nitrogen)│─────▶│ *Aim* : Evaporation of solvents in│
└─────────────────────────────────────────┘      │ oxygen-free environment           │
                    │                            └──────────────────────────────────┘
                    ▼
┌─────────────────────────────────────────┐
│   Olive oil = Total olive oil content (%)│
└─────────────────────────────────────────┘
                    │
                    ▼
┌─────────────────────────────────────────┐
│  Total phenolics/antioxidants and        │
│  compositional analyses                  │
└─────────────────────────────────────────┘
```
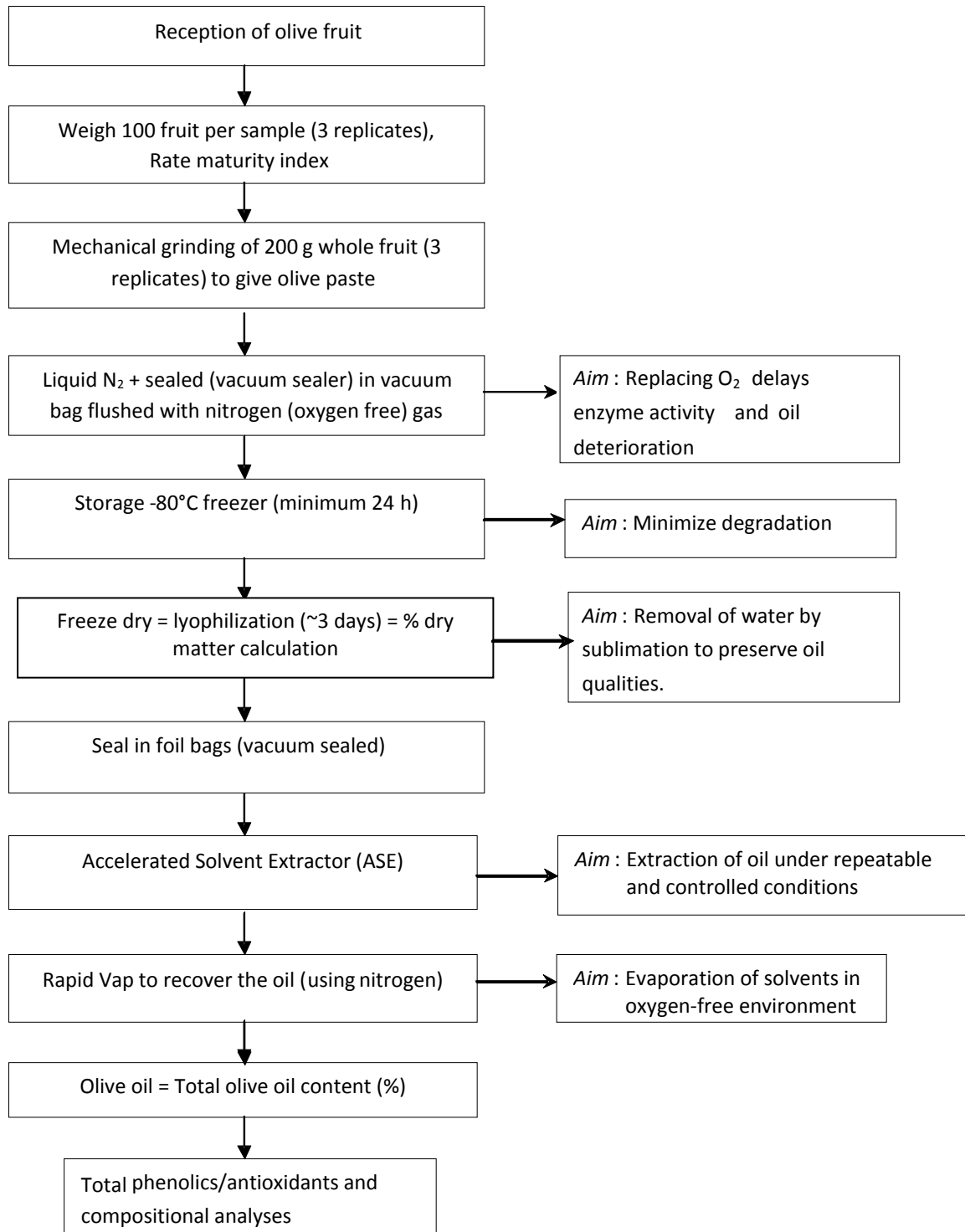
Figure 2. Steps used to process olives at Plant & Food Research.

4.2.1 Physical measurements on the fruit

The following instrumental measurements of fruit were performed on each of the three replicates on the day the fruit were received at the laboratory.

4.2.1.1 Dry matter percentage and sampling for oil extraction

Approximately 200 g of whole olive fruit was mechanically ground in a hammer mill grinder and placed in a pre-weighed dish. The weight of the ground paste was recorded, then snap frozen using liquid nitrogen, placed in an aluminium foil bag, flushed with nitrogen gas (N_2) (food grade, oxygen free; BOC, Auckland, New Zealand), heat sealed and stored at -80°C freezer for at least 24 hours. Later the dish containing the paste was removed from the foil bag, freeze dried for 72 hours at 20°C and the dried weight recorded immediately after removal from the freeze dryer. The dry weight of the tissue in each replicate dish was divided by the original fresh wet weight of the tissue and multiplied by 100 to obtain the percentage (%) dry matter of that harvest. The dried tissue was then used for oil extraction using an Accelerated Solvent Extractor machine (ASE 300®, see below for details).

4.2.1.2 Fresh fruit weight

The weight of one hundred fruit from each replicate was taken to record the average changing fruit weight between harvests.

4.2.1.3 Visual colour maturity index (MI)

Maturity index (MI), also called visual colour rating, is a numerical measure of olive maturity that growers can use to predict the optimum stage for harvesting. MI was determined according to the method described by Kiritsakis (1998) and Boskou (2006). One hundred olives were randomly drawn from the olive sample. The fruit were then graded for both external and internal (pulp) colour according to the colour scale shown in Figure 3. The internal colour was assessed for all the olives with an external MI score of four after the fruit were cut into half to continue the assessment.

Where: A, B, C, D, E, F, G and H represent the number of fruit in each Class 0, 1 2 3, 4, 5, 6, and 7 respectively. The maturity index (MI) of the sample was calculated using the following formula.

$$MI = \frac{A \times 0 + B \times 1 + C \times 2 + D \times 3 + E \times 4 + F \times 5 + G \times 6 + H \times 7}{100}$$

External Assessment:

Class 0= Intense green skin

Class 1= Green yellowish skin

Class 2= Green skin with red-purple gradient in less than half the fruit

Class 3= Red-Purple skin in more than half the fruit

Internal Assessment:

Class 4= Purple/black skin with white or green pulp

Class 5= Purple/black skin and less than half the pulp is purple

Class 6= Purple/black skin and more than half the pulp is purple

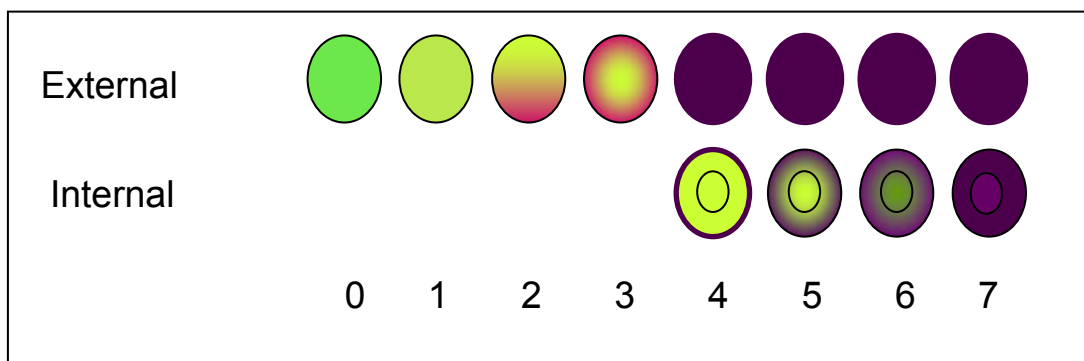Class 7= Purple/black skin and purple pulp all the way to the stone.

Figure 3. Colour maturity index for olives.

4.3   Determination of oil content using Accelerated Solvent Extractor (ASE®)

The three replicates of freeze dried tissue were mixed in equal parts and ground further in a domestic coffee grinder. Two analytical replicates of this mixed tissue sample were processed using the Accelerated Solvent Extractor (ASE® 300 Dionex Corporation, Sunnyvale California) in pure hexane (95% HPLC /GC pesticide grade) solvent. Extraction conditions were as follows: five minutes' sample heating time at 60°C followed by 100 min total extraction time at 1500 psi. The run was split into five cycles of 20 min with nitrogen gas (oxygen free, 99.99% purity, BOC gases) purge cycle of 90 seconds. The oil dissolved in the solvent was auto-collected in glass bottles. The glass bottles were nitrogen flushed during and after extraction. The oil in hexane solvent was recovered by the RapidVap unit (RapidVap N_2 Evaporation Systems, Labconco® Corp. Kansas City, MO). The percentage oil content in the sample of fruit was calculated and expressed on a percent dry weight basis i.e., excluding the moisture content of the fruit.

4.3.1 Chemical measurements in the oil-Oil compositional analyses

Because of budget constraints throughout the harvest period, harvest samples were selected for further compositional analyses as follows:

Table 2. Analyses performed on the olive fruit samples.

Chemical analyses performed	Harvest samples analysed
Total oil content (% dry weight)	On all fruit harvest samples
Total phenolics	On 3 harvest samples (including the *advised commercial harvest)
Fatty acid, tocopherol and sterol	On the commercial harvest sample (advised by grower)

*Harvest date advised by the grower

4.3.1.1 Determination of total phenolic content in the oil by Folin-Ciocalteu assay

The Folin-Ciocalteu assay is a common colorimetric method that operates by an oxidation-reduction mechanism (Vinson et al., 2001). Two mL of oil were weighed and added to 2 mL n-hexane (HPLC grade, UltimAR). The oil-hexane mixture was then transferred into a 100-mL separating funnel where phenolic compounds were extracted by adding 6 mL methanol-acidified water (80:20) and shaking vigorously for three minutes. The aqueous layer at the bottom of the funnel was then collected. This extraction cycle was repeated three times. Gas was released frequently during shaking by opening the stopper as pressure built up inside the funnel. The combined aqueous fractions were completely dried in the Rapid Vap for 90 min at 40°C at a shaking speed of 70. The dried extract was redissolved in 3 mL of absolute methanol and gently mixed thoroughly. This methanolic extract was used to determine the total phenolic content using Folin-Ciocalteu reagent (Ainsworth and Gillespie, 2007, Singleton et al., 1998). The methanolic extraction samples were centrifuged for 5 minutes at 16089 g (13000 rpm; Biofuge13Heraeus Sepatch, USA) before analysis in order to precipitate undesirable cloudiness of the extract, which may affect the absorbance reading in later stage. Because of the higher concentration of phenolics in early harvest fruit, this methanolic extract was diluted further (with absolute methanol) depending on time of harvest (higher dilutions for early harvest, and lower dilutions for late harvest).

Folin-Ciocalteu reagent (1 mL) was added into each methanolic sample (200 µL). Three to eight minutes after the addition of Folin-Ciocalteu reagent, 0.8 mL of sodium carbonate (Na_2CO_3 concentration 75 g/L) was then added. The tubes were incubated for 90 minutes in the dark at room temperature. One mL of each sample was then centrifuged at 34,010 g (Biofuge13 Heraeus Sepatch, USA) for five minutes and 200 µL of supernatant was pipetted into a microplate. The absorbance was read at 760 nm using a Spectra-Max microplate reader

(SPECTRA-Max PLUS384, MDS Analytical Devices, USA). A range of dilutions in absolute methanol were prepared fresh using a caffeic acid solution (0.5 mg/mL) (Sigma-Aldrich, New Zealand) to obtain a standard curve. Caffeic acid one of the phenolic compounds present in olive oil, and it is a recommended standard as it gives the most sensitive response, relative to other standards such as gallic acid and tannic acid (Ayton et al., 2007; Blekas et al., 2002; Boskou et al., 2006; Kiritsakis, 1998). Results are expressed as milligram caffeic acid equivalent per kilogram of oil.

4.3.1.2 Determination of fatty acid composition in the oil

The fatty acids were saponified and methylated using the method described by Hartman & Lago (1973). A Hartman reagent was prepared comprising ammonium chloride dissolved in methanolplus concentrated sulphuric acid. The fatty acids were saponified with 0.5M methanolic-potassium hydroxide and methylated with the Hartman reagent. Hexane was added to extract the fatty acid methyl esters and the supernatant layer was injected into the gas chromatograph (GC), with a DBwax capillary column heated to 250°C. The individual fatty acid composition of the oils was determined as a percentage of the total peak area.

4.3.1.3 Determination of individual tocopherols by high performance liquid chromatography (HPLC)

The individual tocopherols (α (alpha), β (beta), δ (delta), and γ (gamma)) were analysed by high performance liquid chromatography (HPLC) based on the method of Indyk (1988). The oil was saponified in an ethanol (containing pyrogallol) and potassium hydroxide solution. The unsaponifiable fraction containing the tocopherols was extracted with a hexane and di-isopropyl ether mixture. After centrifugation, the unsaponifiable fraction was evaporated to dryness under nitrogen. The residue was dissolved in ethanol then injected into the HPLC system. The Shimadzu HPLC system consisted of a 5-μm Lichrospher Si-60 (250 X 40 mm) column and a Guard column (Silica 4 X 3.0 mm AJ0-4348); detection was by UV-Vis detector at 294 nm. A mobile phase of n-Hexane : isopropanol (98.9 : 1.1) was used. External standards were used and peak areas were used to calculate individual tocopherol concentrations. Standards were: α-tocopherol (≥98% HPLC grade, Fluka), β-tocopherol (+90%, Supelco), δ-tocopherol (approx. 90%, Sigma), (γ-tocopherol ≥97%, Fluka).

4.3.1.4 Determination of phytosterols by gas chromatography-mass spectrometry (GCMS)

The oil samples were saponified using the method described by Indyk (1988) and as for tocopherols. The trimethylsilyl (TMS) ether derivatives of sterols were prepared by adding 0.20 mL derivatization agent to the concentrated extract (Thompson & Merola, 1993). The derivatization agent-mix was equal parts of (1) bis(trimethylsilyl)-trifluoroacetamide (BSTFA) containing 1% TMCS and (2) pyridine. The trimethylsilyl (TMS) ether derivatives were analyzed by Gas Chromatography-Mass Spectrometry (GCMS). The GC column used was a RTX-5 silica capillary column (60 m x 0.25 mm ID, crossed bonded 5% diphenyl -95% dimethyl polysiloxane (0.25um film) (Restek Corp., USA). Peaks were identified by comparing retention times with those of the authentic standards and by comparing mass spectra of unknown peaks with the spectra of known standards and referring to literature sources (Dutta et al. 1998, Li et al., 2007). The internal standard used was 5a-chloestan-3b-ol (approx 95%).

5   Results and Discussion

5.1   Differences in climate throughout New Zealand

The northern and Hawke's Bay regions of the North Island are generally known for their earlier and warmer springs, warm to hot summers and late cooler autumns. In contrast, the regions throughout the South Island are known for their later and more changeable springs, which can lead to periods of very cold, or warm to hot weather, which can result in generally shorter summers, with earlier cool to cold autumns. Frosts during springtime and autumn are often associated with these areas.

One way of summarising differences in climate data is the use of Growing Degree Days (GGD; Orlandi, Ruga et al., 2005; Orlandi et al,. 2009). GDD is a measure of ambient heat accumulation (number of days above a base-line temperature, usually 10°C). It can thus be used as a rough "rule of thumb" as to how weather conditions are affecting a crop physiologically (although it can of course be complicated by extreme conditions such as drought or other extremes in temperature e.g. frost). This information can be used in a variety of ways, including regional or seasonal comparisons, or to predict when a crop will reach maturity.

Historically, the Northern and Hawke's Bay regions have a much larger number of growing degree days in spring and autumn than the southern regions (Table 3). For example, the Auckland region had a GGD of 858 for spring and autumn (September-November + March-May), compared with 445 for the same period in the Nelson region. All regions generally have a high number of sunshine hours, ranging from around 1900 to 2400 hours per year (Table 4).

In contrast, rainfall differs among regions, with the Auckland and Northland regions having the highest rainfall (approximately 1300 mm per year), Hawke's Bay, Wairarapa and Nelson having a lower rainfall (c. 950-1050 mm), and the Central Otago area having the least rainfall (approximately 360-900 mm).

These differences in climate mean that olive fruit maturity is achieved at different times during the season, depending on the orchard location, and normally this will be earlier in the North Island regions and later in the South Island regions. For instance, commercial harvest time (as advised by the growers) in the North Island orchards starts in early April and ends in early June, as opposed to the South Island, where commercial harvest time is only starting in June and can be extended until the beginning of July.

Table 3. Monthly average growing degree days (GDD) for regions in New Zealand, baseline 10°C (1971 – 2000).

Number of Growing Degree Days (GDD)

Region	Total	Sept.	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.	May	Jun.	Jul.	Aug.
Northland	**2326**	133	127	210	330	317	311	296	215	153	85	54	96
Auckland	**1738**	14	116	151	221	275	259	241	166	110	54	32	38
Hawke's Bay	**1415**	47	104	146	217	251	227	197	119	55	20	14	19
Wairarapa (Masterton)	**1253**	49	45	138	250	214	220	168	96	43	10	0	20
Nelson	**1180**	29	77	124	194	236	213	177	84	31	6	3	6
Otago	**960**	20	57	104	175	228	188	131	43	9	1	0	3

Source: The National Climatic Data Base-NIWA (http://cliflo.niwa.co.nz/).

Table 4.: Annual average rainfall (mm) and sunshine hours for regions in New Zealand (1971 – 2000).

Region	Rainfall (mm)	Sunshine (hours)
Northland	1334	2070
Auckland	1240	2060
Hawke's Bay	1051	2180
Wairarapa	979	1915
Nelson	970	2405
Otago	360	2025

Source: The National Climatic Data Base-NIWA (http://cliflo.niwa.co.nz/).

5.2 Regional differences in fruit maturation and oil content for the cultivars 'Frantoio', 'Leccino' and 'Koroneiki'

Because 'Frantoio', 'Leccino' and 'Koroneiki' are the most commonly planted cultivars in New Zealand, the main focus of this report is on these three cultivars.

5.2.1 Oil content

In the 2010 season, the total oil content (on a dry basis) for 'Frantoio' and 'Leccino' grown in the different regions throughout New Zealand ranged from approximately 28 to 48%, and for 'Koroneiki' ranged from approximately 32 to 52% (Figures 4, 5, Figure 6). In general, orchards in the northern regions (Northland and Auckland) developed fruit with higher oil content, with all but one orchard reaching a plateau at over 45% oil for all three cultivars. The further south the orchards were, the more the potential maximum oil content decreased. In the Matapiro orchard in Hawke's Bay, where a larger number of samples were taken, maximum oil content for the 'Frantoio' and 'Leccino' cultivars reached 40-42%, while in the Olea orchard (in the Wairarapa region), the oil content for these varieties was lower again, at c. 23-27%. Oil content in 'Koroneiki' from the North Island orchards reached between 40 and 44% before it showed a tendency to reach a plateau. There was much less variability in oil content for the regions in the South Island, with fruit from all regions reaching similar potential maximum oil content of c. 30-35% for 'Frantoio' and 'Leccino', and 36% for 'Koroneiki', reflecting their regions' geographical and climatic similarities (Figure 4 to 9).

The timing of maximum oil accumulation also differed across the regions. In general, fruit from the orchards in the northern regions achieved maximum oil content earliest, followed by those in Hawke's Bay and lastly those from orchards in the South Island (which all had very similar oil accumulation rates and timings). The differences in accumulation rates between regional extremes were large, with fruit from the earliest maturing orchard in the Auckland region reached maximum oil content around 6 weeks earlier than those from the latest maturing orchard in the Otago region.
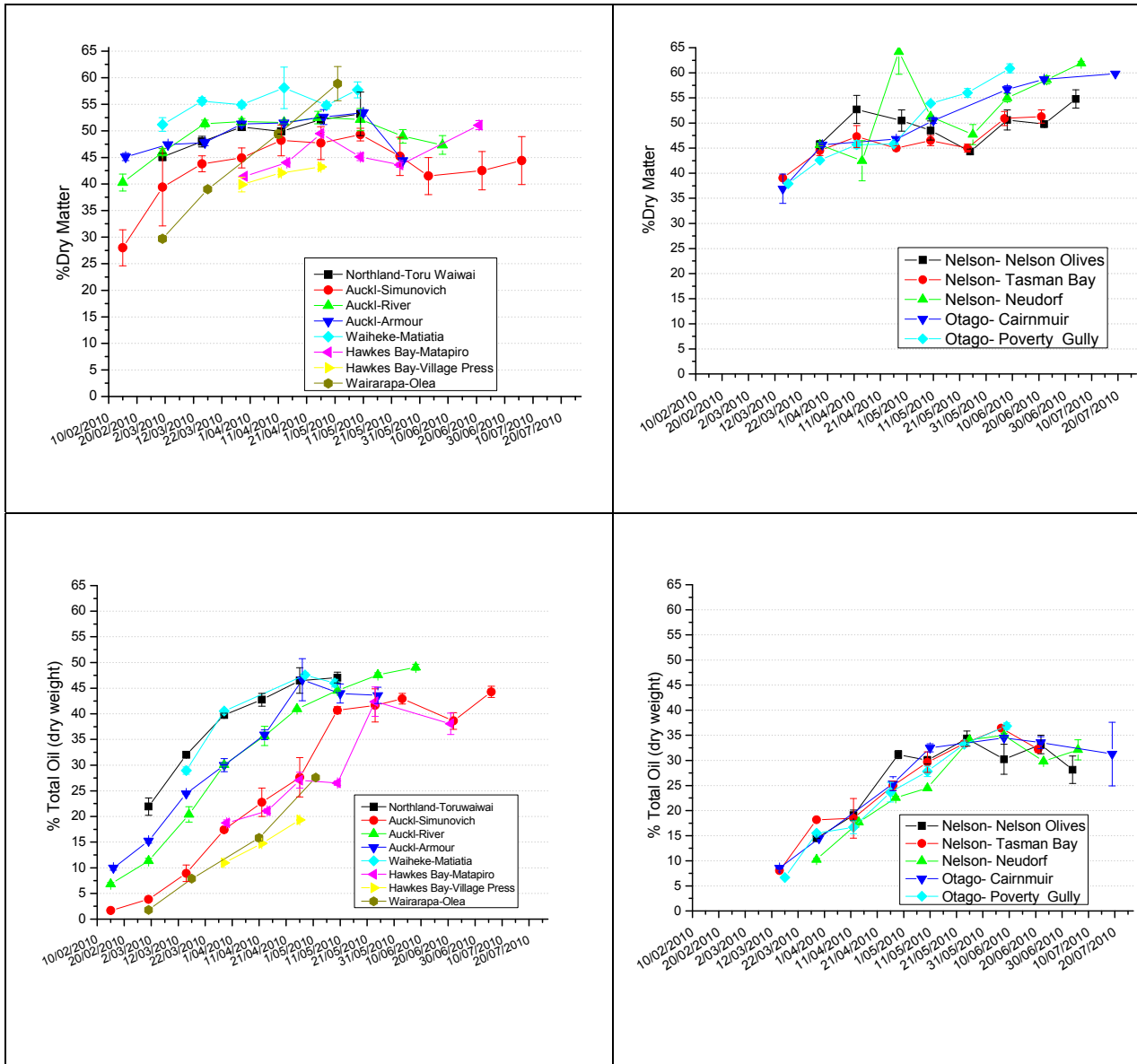
Figure 4. Percentage dry matter and total oil (obtained by solvent) of 'Frantoio' fruit harvested during the 2010 season.
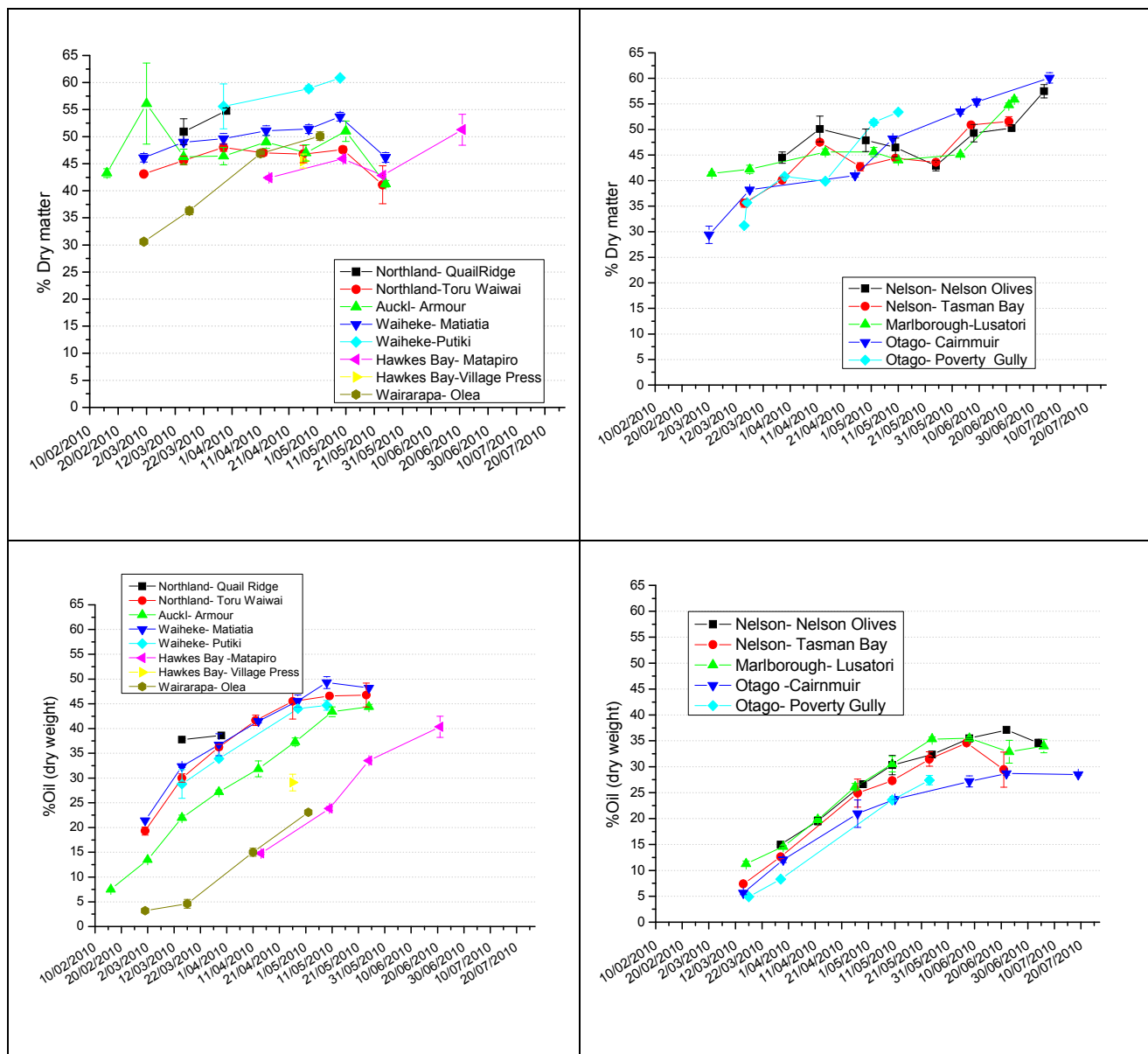
Figure 5. Percentage dry matter and total oil (obtained by solvent) of 'Leccino' fruit harvested during the 2010 season.
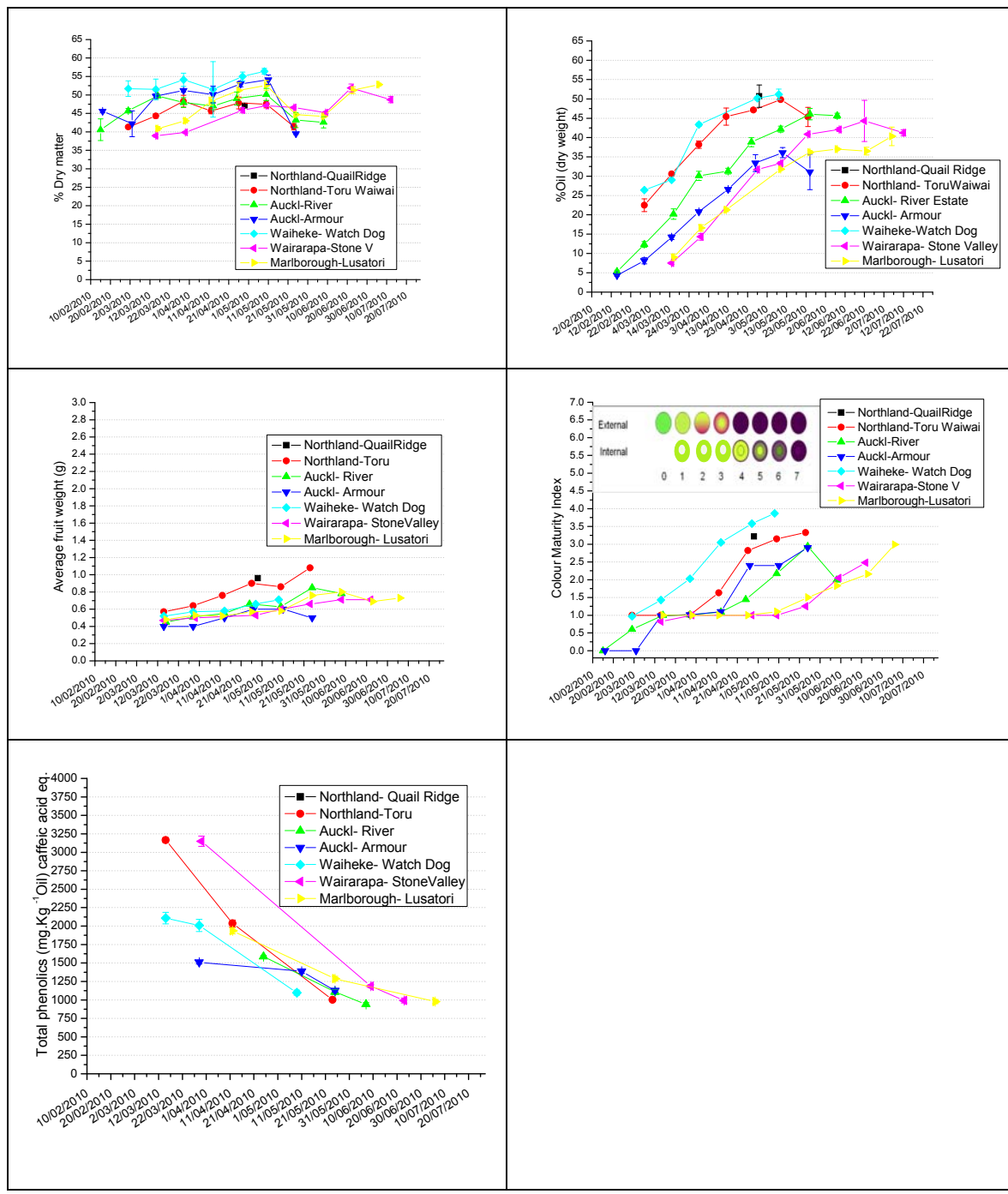
Figure 6. Percentage dry matter, total oil (obtained by solvent), fruit weight, colour maturity index and total phenolic content in oil, of 'Koroneiki' fruit harvested during the 2010 season.
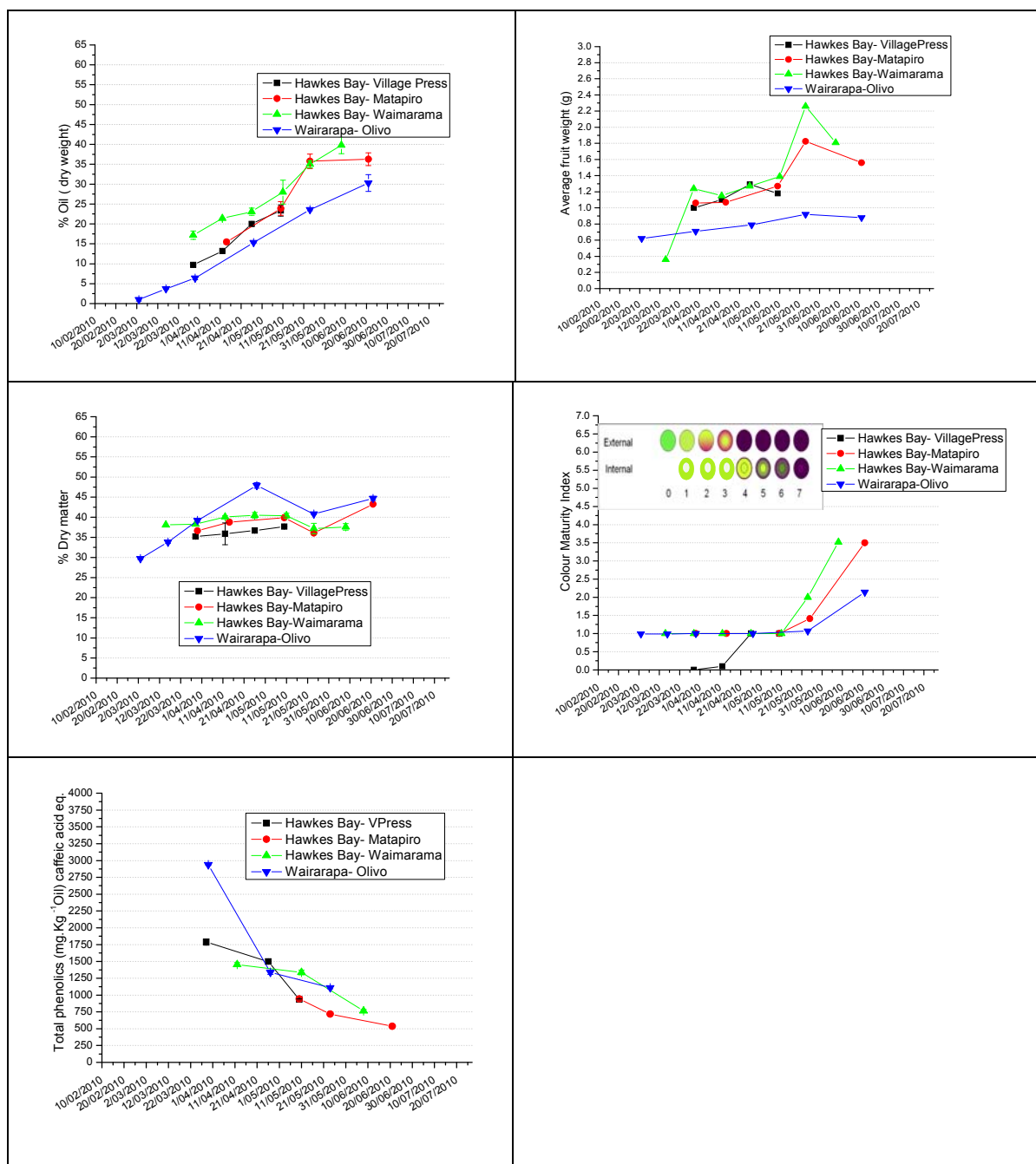
Figure 7. Percentage dry matter, total oil (obtained by solvent), fruit weight, colour maturity index and total phenolic content in oil of 'Barnea' fruit harvested during the 2010 season.
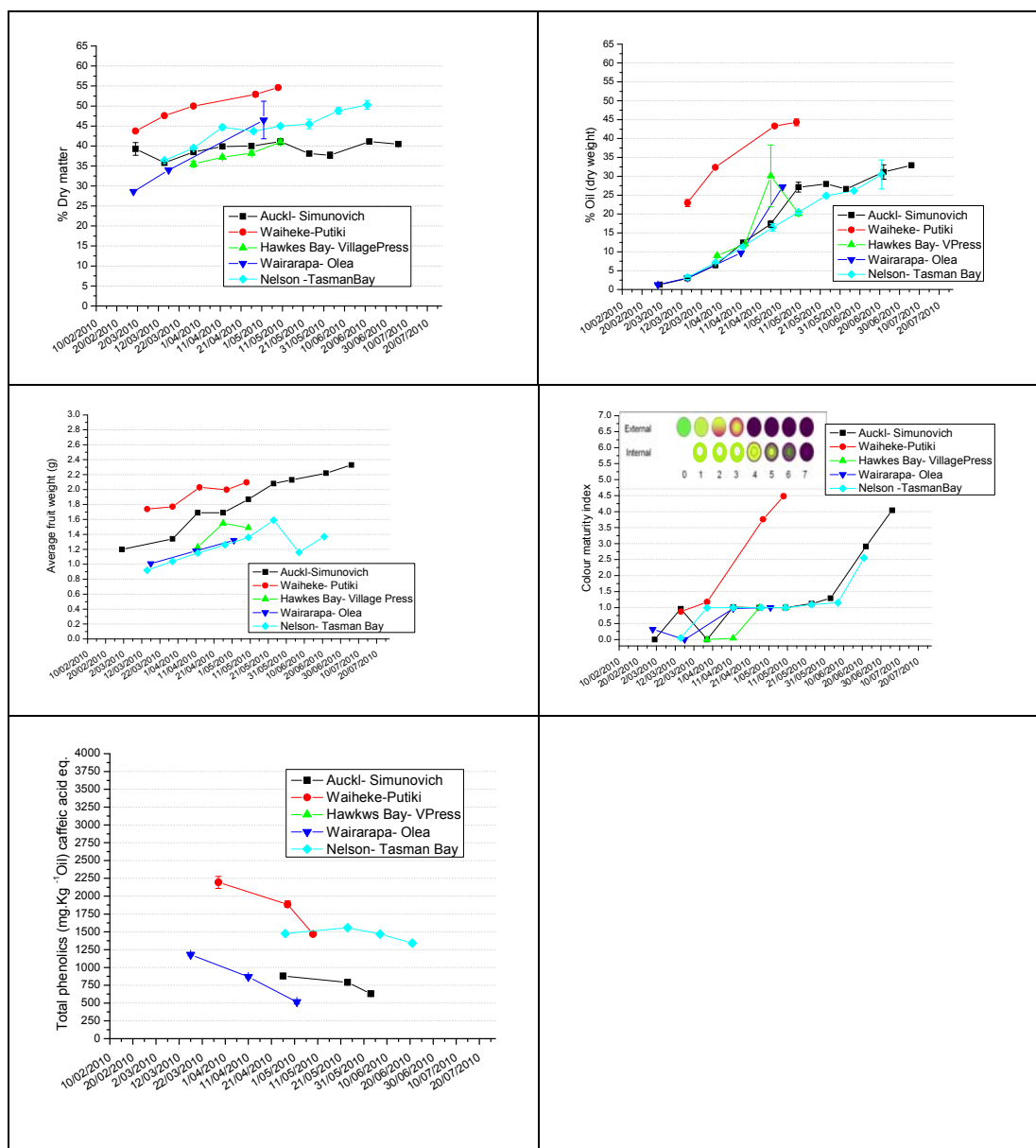
Figure 8. Percentage dry matter, total oil content (obtained by solvent), fruit weight, colour maturity index and total phenolic content in oil of 'Picual' fruit harvested during the 2010 season. (N.B. No phenolic analyses were carried out for oils from The Village Press.)
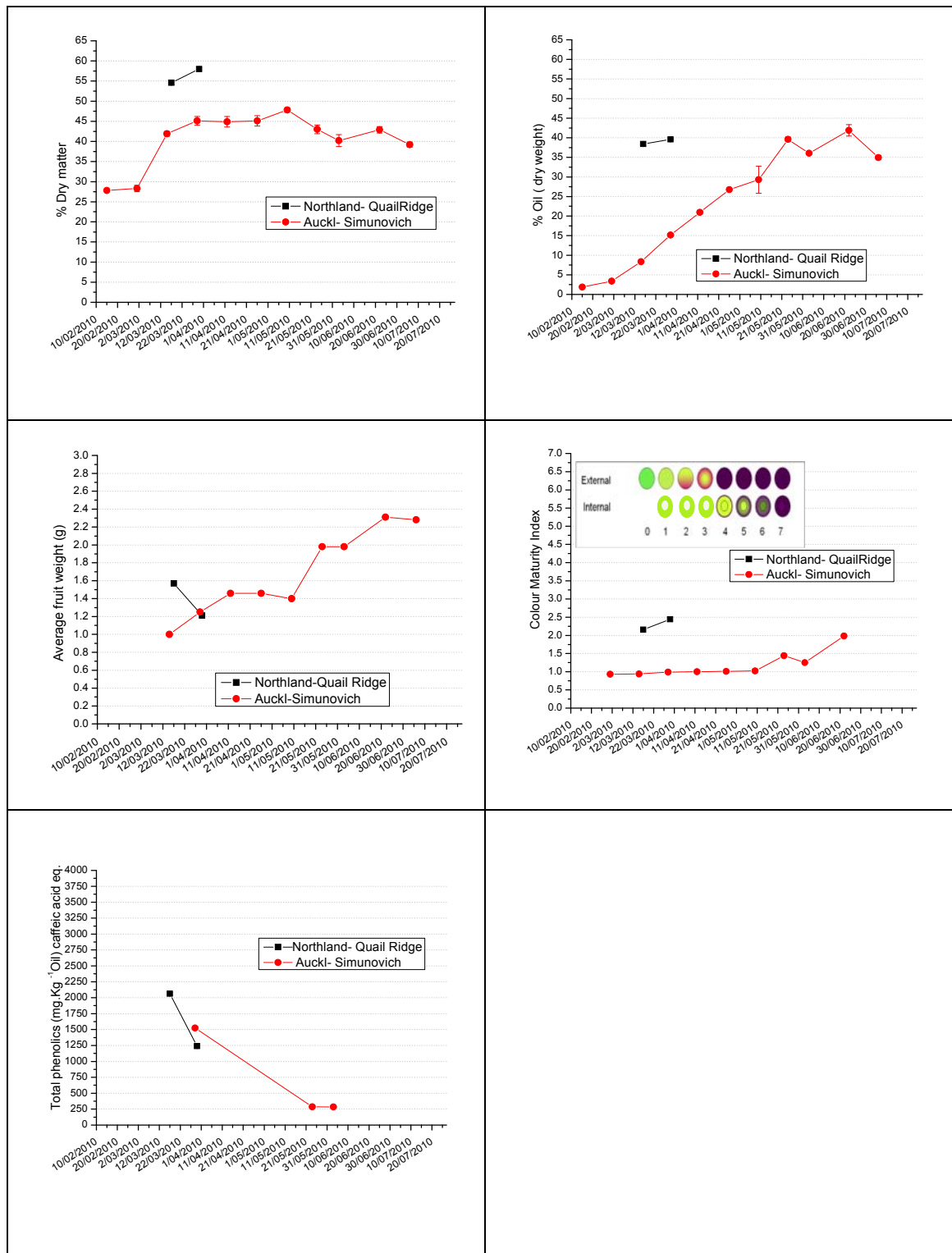
Figure 9. Percentage dry matter, total oil (obtained by solvent), fruit weight, colour maturity index and total phenolic content in oil of 'J5' fruit harvested during the 2010 season.

5.2.2   Dry Matter Percentage

Dry matter content (%) of fruit is considered an indicator of the oil content. The current results were similar to previous seasons where, although there were often similar trends in accumulation rates, actual dry matter content related to different values of oil content depending on the region. For example, one orchard in the Wairarapa had a maximum dry matter concentration of 58% that related to 27% oil content, compared with a Waiheke orchard that had 57% dry matter but 48% oil content (Figures 4 to 9).

This discrepancy is probably reflected in the different amounts of moisture (water) in the fruit at harvest, which affects the dry matter percentage. Water content in the fruit can be affected by rain, drought, or irrigation practices (Kiritsakis, 1998). For this reason, the total solids (expressed in g) approach was developed in the 2009 research, which eliminates the water effect on the dry matter results. As explained in the results from 2009, using this technique the correlation with oil content is much higher (Requejo-Jackman et al., 2010).

5.2.3   Fruit Weight

Fruit weights for 'Frantoio' and 'Leccino' were variable but generally higher for orchards in the North Island than in the South Island. For instance, Table 5 shows samples of 'Leccino' fruit from Northland and Otago, where the differences in fruit development rates for the same cultivar at very similar harvest times are illustrated. Maximum fruit weight for North Island orchards ranged from 1.0 to 2.2 g for 'Frantoio' and 1.2 to 4.5 g for 'Leccino'. For South Island orchards, fruit weight in general was lower but less variable, ranging from 0.8 to 1.2 g for 'Frantoio' and 1.0 to 1.8 g for 'Leccino'. The 'Koroneiki' cultivar fruit weight, with typically a small sized fruit, was less influenced by region being generally low and ranging from 0.5 to 1.1 g. In general, fruit weight tended to be largest in the northern regions and to decrease further south (Figures 4 to 9).

This is probably a reflection of the longer growing seasons for the northern regions, although there are other factors not measured here that can affect fruit weight (such as crop load, tree age and water relations (e.g. lack of irrigation)).

Table 5. Characteristics of samples of 'Leccino' fruit from A) North Island (Northland) and B) South Island (Otago).

A

B

- Cultivar: 'Leccino'
- Region: Northland
- Orchard: Toru Waiwai
- Harvest: 24-05-2010
- Total oil (dry basis): 46.8%
- Average fruit weight: 3.1 g
- Colour maturity index: 4.4

Commercial harvest date*: 26-04-2010 at which:

- Total oil (dry basis)[+]: 45.5%
- Average fruit weight: 2.2 g
- Colour maturity index: 4.1

- Cultivar: 'Leccino'
- Region: Otago
- Orchard: Cairnmuir
- Harvest: 11-05-2010
- Total oil (dry basis): 23.7 %
- Average fruit weight: 1.3 g
- Colour maturity index: 1.2

Commercial harvest date*: 22-06-2010 at which:

- Total oil (dry basis)[+]: 28.8%
- Average fruit weight: 1.3 g
- Colour maturity index: 2.8

*commercial harvest date as advised by grower
[+] oil extracted by solvent

5.2.4 Colour Maturity Index (MI)

The colour maturity index (MI) was variable for both North and South Island orchards (Figures 6 to 9, 11 and 12) and patterns of colour development (using for calculating the index) did not usually relate well to the changes in oil content accumulation, particularly for the cultivar 'Frantoio'. Figure 10 shows poor correlations (r^2 less than 0.5) between oil content and the colour maturity index for this cultivar for the North and South Island orchards, and for the data combined for the whole country for this particular cultivar, respectively.

The MI is designed specifically to assist harvest timing and is commonly used in hot dry climate countries such as Spain and Australia. In a very different growing climate such as New Zealand, the colour maturity index has shown to be not appropriate as an indicator for oil content and harvest maturity for the cultivars studied.

For example, for 'Frantoio', fruit from some orchards in the hotter northern regions reached a maturity index of 3.5 (green background with >50% purple blush) and colour development tracked the increase in oil content quite well (Figure 11). However, fruit from another Auckland orchard achieved only a maturity index of 2.5 (green - yellow skin fruit with <50% purple blush) at harvest despite having similar oil content to those from the other Northern orchards. In addition, in the cooler regions, particularly in the South Island, the maturity index for orchards was much lower, ranging from 1.0 (green skin) to 2.5 (green background with < 50% purple blush) at harvest time. In these South Island orchards, where fruit maturation is occurring during cooler weather, there seems to be a dislocation of fruit colour development and fruit development i.e. the fruit's colour development system is slower than in the North Island and therefore less in synchrony with the oil accumulation fruit system.

For 'Leccino' in the North Island, MI was generally higher than for 'Frantoio' (ranging from MI 3.4 to 5.2 at commercial harvest; Figure 12). Although the maturity index related better to oil accumulation for fruit from the North Island orchards (r^2 = 0.83; Figure 13A), the correlation in the South Island was weaker (r^2 = 0.57).

For 'Koroneiki' fruit, the increase in MI during fruit maturation followed similar trends to oil content accumulation for individual orchards, but again did not relate well across orchards and regions ($r2$ = 0.67; Figure 14). For example, all but one orchard reached a maturity index of 3 or more. If the decision were to harvest at maturity index 3, then one orchard would never reach clearance to harvest, despite having accumulated 45% oil content, three of the orchards would be harvested at a maximum oil content and two orchards would be harvested before maximum oil content were achieved. As a further example, a maturity index of 3 for an Auckland orchard (Armour) correlated to c. 30% oil, whereas a maturity index of 3 for a Northland orchard (Toru Waiwai) correlated to nearly 50% oil content.
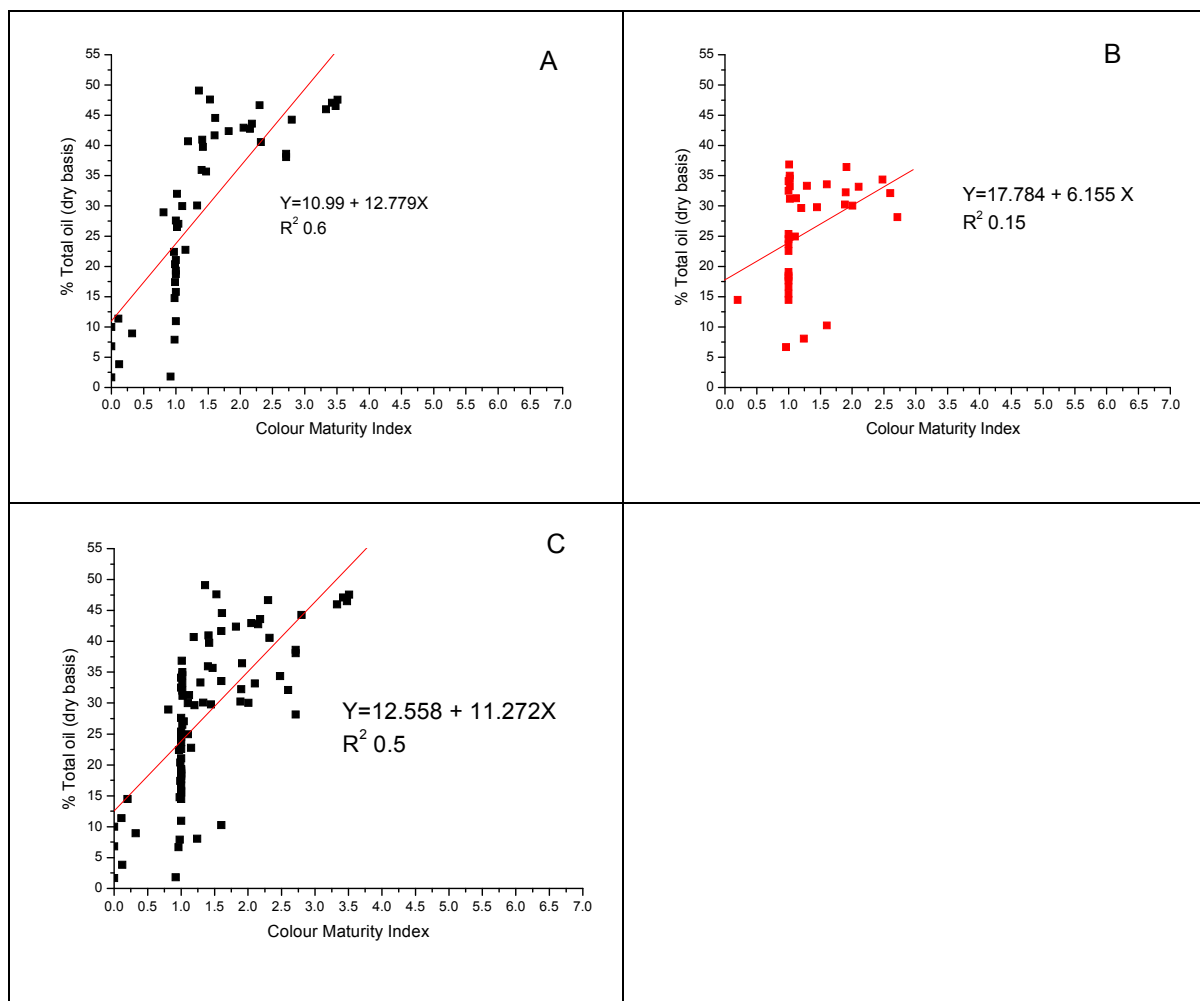
Figure 10. Correlation between colour maturity index and percentage total oil (dry basis) for 'Frantoio' in the A) North Island B) South Island C) North and South Islands harvested during the 2010 season.

Figure 11. Fruit weight, colour maturity index and total phenolic content in oil of 'Frantoio' harvested in the 2010 season.

Figure 12. Fruit weight, colour maturity index and total phenolic content in oil of 'Leccino' harvested during the 2010 season.

Figure 13. Correlation between colour maturity index and percentage total oil (dry basis) for 'Leccino' in the A) North Island, B) South Island C) North and South Islands harvested during the 2010 season.
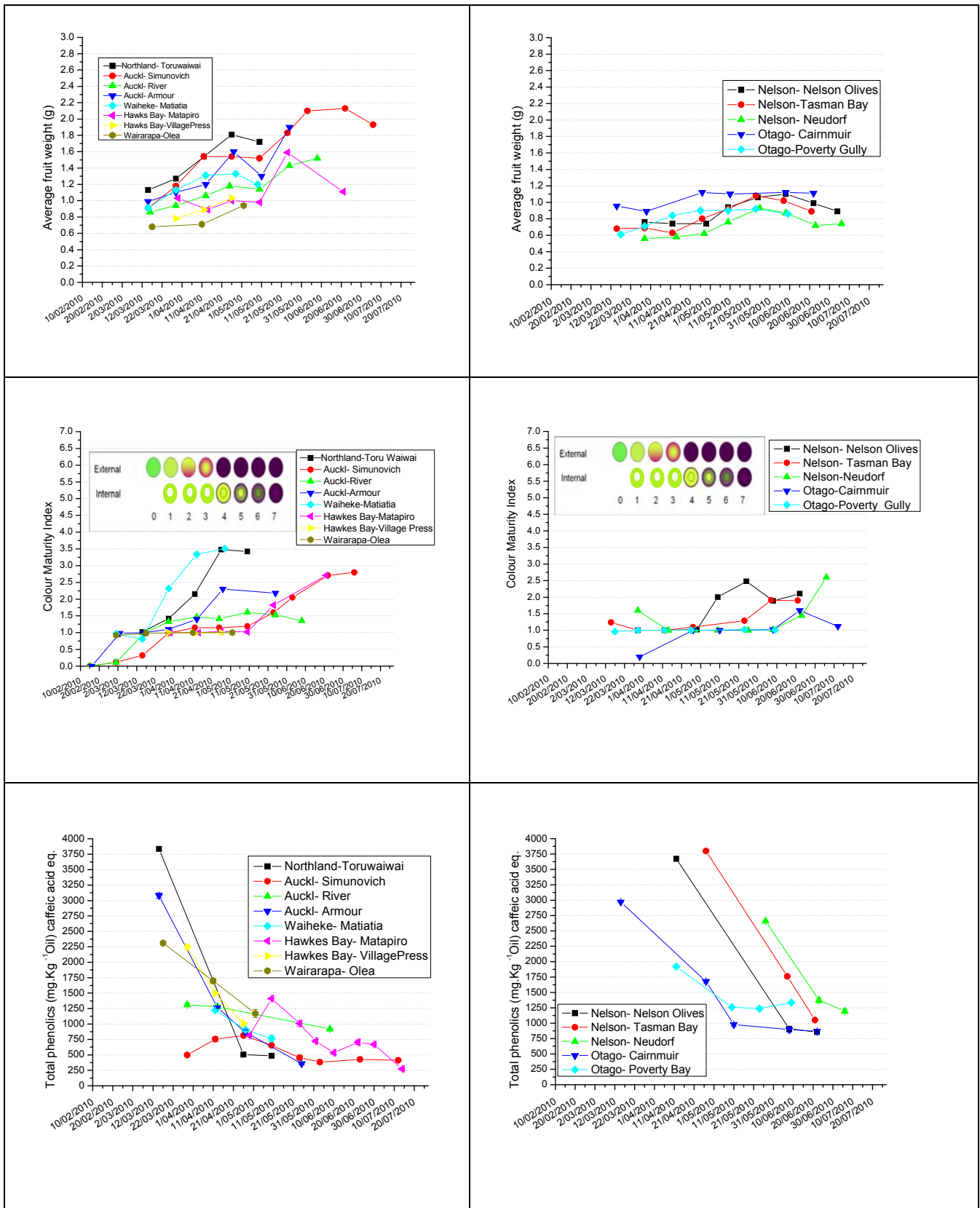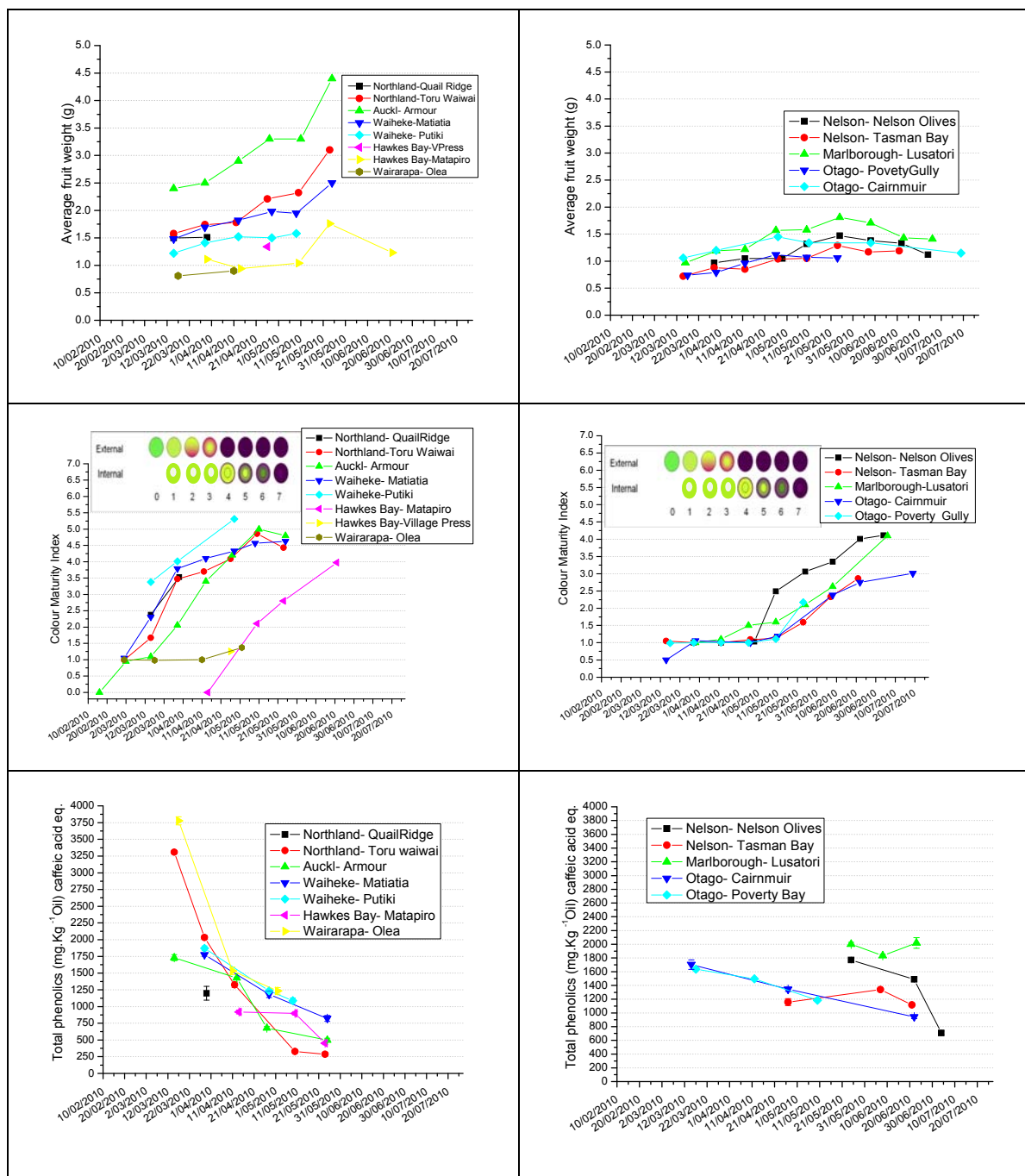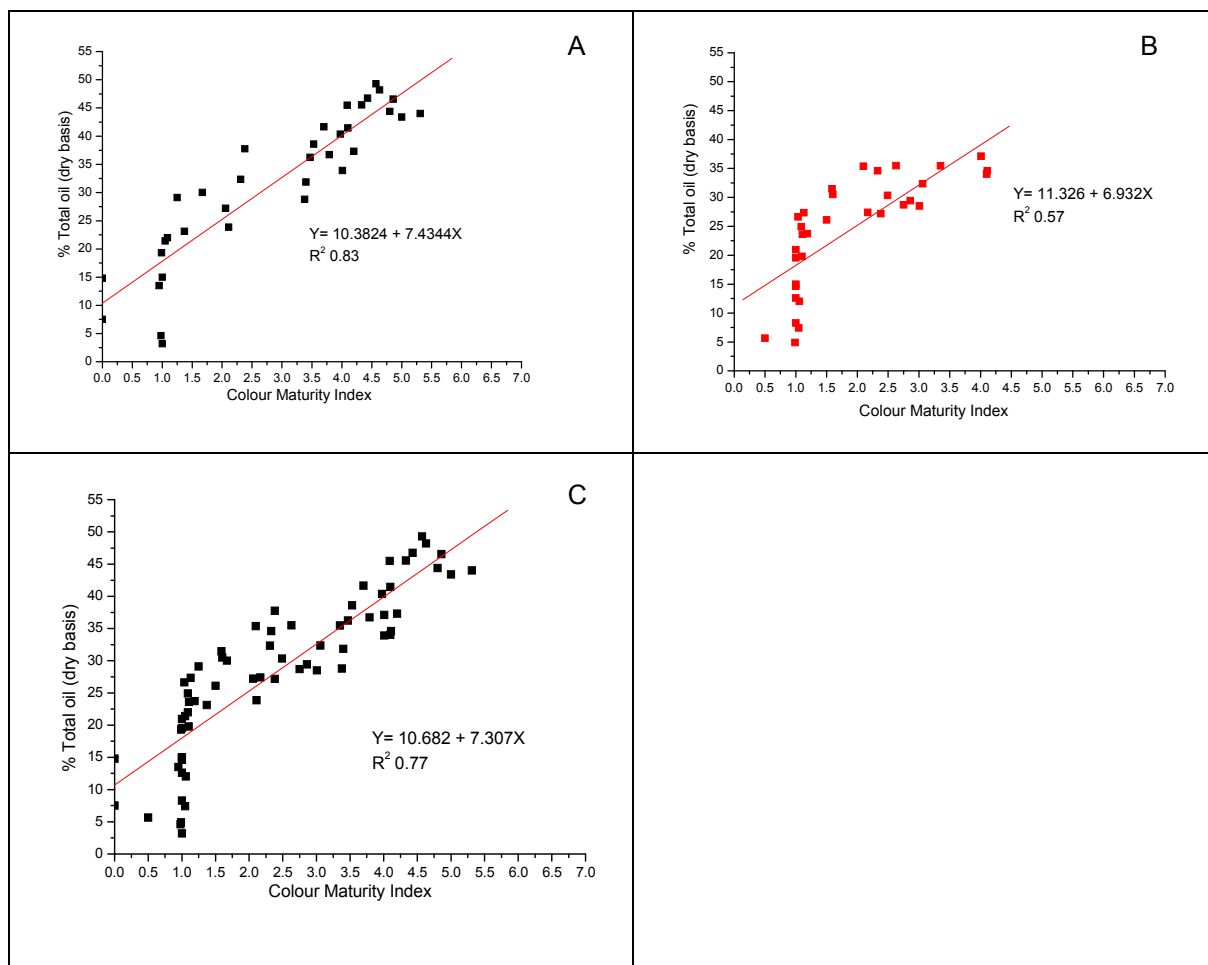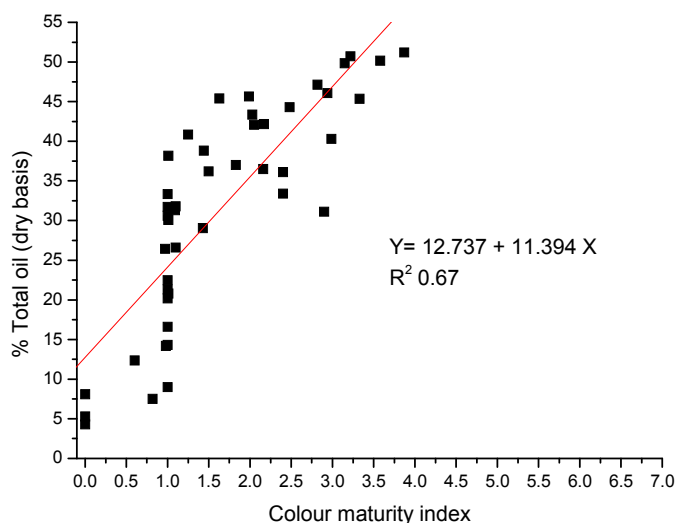
Figure 14. Correlation between colour maturity index and percentage total oil (dry basis) for 'Koroneiki' for North and South Islands orchards (although there was only one orchard in the South Island), during the 2010 season.

5.2.5   Total phenolics content

Total phenolics content decreased with maturity in all regions and cultivars studied (Figure 6 to 12). The rate of decline in the 'Frantoio' and 'Leccino' cultivars was slower and the amount of phenolic compounds measured at the end of the harvest season was higher in the South Island than in the North Island orchards. These higher concentrations of phenolics at the end of the season could be explained by the cooler climates that cause fruit from South Island orchards to mature later than those in the North Island. Cooler climates mean a shorter and slower growth cycle, which in turn means that by harvest time the phenolics in the South Island fruit have not decreased to the concentrations they did in the North Island.

To obtain an estimate of the general trends in oil percentage and total phenolics over time for each cultivar in the North and South Islands, prediction curves were fitted (Figure 15). To calculate the overall prediction curves, individual curves with a common shape were fitted to each orchard and then averaged. This had the advantage of producing an equation that was not biased towards predicting only the orchards with the greater number of measurements over the trial period. The resulting equations (Tables 6 and 7) can be used to predict an average oil percentage or total phenolic concentration at a certain time point, where time is given as days after 15 February (which was the earliest observed date in the trial). Predictions should be made only within the observed time period, as extrapolating outside this can lead to very unreliable results (for example negative values), since it assumes that the trends observed at the end points of the trial continue.
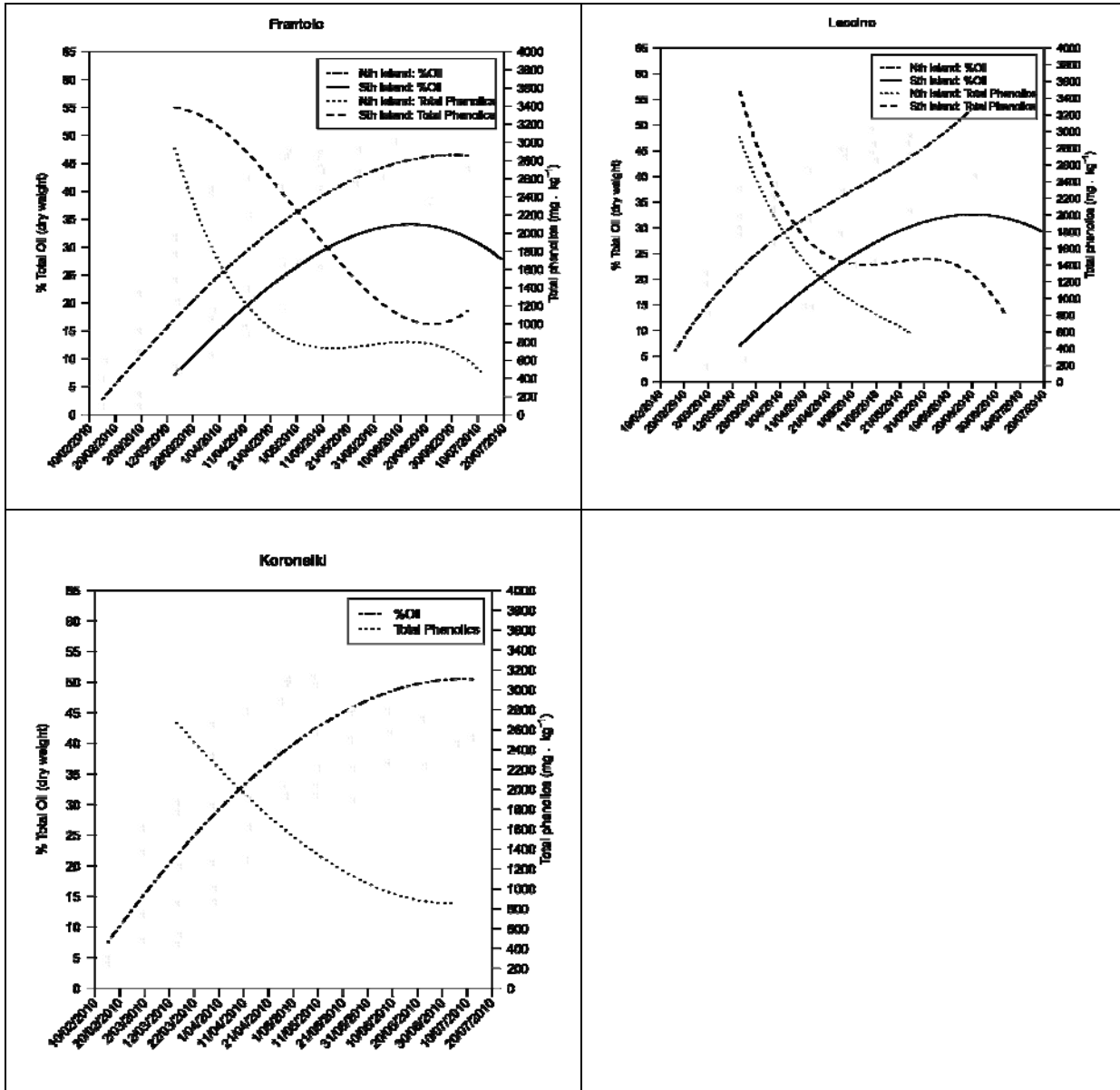
Figure 15. Fitted curves for pooled orchards for North and South Islands, showing patterns of oil accumulation and total phenolic content in oil of 'Frantoio', Leccino and 'Koroneiki' harvested in the 2010 season.

Table 6. Prediction equations for the proportion (%) of olive oil over time in the season by cultivar for the North and South Islands.

Cultivar	Region	Prediction Equation for % Oil (predictions only valid within time period)	Time Period
'Frantoio'	North Island	$2.83 + 5.40t \times 10^{-1} - 8.37t^2 \times 10^{-4} - 5.68t^3 \times 10^{-6}$	15th Feb to 6th Jul (t=0 to 141)
'Frantoio'	South Island	$-6.213 + 4.76t \times 10^{-1} + 5.33t^2 \times 10^{-4} - 1.42t^3 \times 10^{-5}$	15th Mar to 19th Jul (t=28 to 154)
'Leccino'	North Island	$5.30 + 7.48t \times 10^{-1} - 6.37t^2 \times 10^{-3} + 2.76t^3 \times 10^{-5}$	16th Feb to 21st Jun (t=1 to 126)
'Leccino'	South Island	$-5.31 + 4.47t \times 10^{-1} + 9.39t^2 \times 10^{-5} - 9.98t^3 \times 10^{-6}$	15th Mar to 23th Jul (t=28 to 158)
'Koroneiki'	Both	$7.51 + 5.51t \times 10^{-1} - 1.40t^2 \times 10^{-3} - 2.44t^3 \times 10^{-6}$	15th Feb to 19th Jul (t=0 to 147)

Table 7. Prediction equations for the concentration of total phenolics in oil over time in the season by olive cultivar for the North and South Islands.

Cultivar	Region	Prediction Equation for Total Phenolics (predictions only valid within time period)	Time Period
'Frantoio'	North Island	$6677.10 - 178.75t + 1.76t^2 - 5.69t^3 \times 10^{-3}$	15th Mar to 12th Jul (t=28 to 147)
'Frantoio'	South Island	$2826.06 + 46.00t - 1.06t^2 + 4.62t^3 \times 10^{-3}$	15th Mar to 6th Jul (t=28 to 141)
'Leccino'	North Island	$6237.96 - 1.60t \times 10^{-2} + 1.69t^2 - 6.56t^3 \times 10^{-3}$	15th Mar to 25th May (t=28 to 99)
'Leccino'	South Island	$7823.21 - 214.47t + 2.35t^2 - 8.45t^3 \times 10^{-3}$	15th Mar to 4th Jul (t=28 to 139)
'Koroneiki'	Both	$3498.58 - 30.83t + 3.09t^2 \times 10^{-2} + 3.90t^3 \times 10^{-4}$	15th Mar to 6th Jul (t=28 to 141)

5.3 Specific regional effects on the three key cultivars: 'Frantoio', 'Leccino' and 'Koroneiki'

5.3.1 Specific regional effects on 'Frantoio'

- Maximum oil content ranged from 28 to 48%, with cooler or later maturing regions producing fruit with lower oil content. Oil content in the North Island ranged from 28 to 48% and in the South Island ranged from 28 to 37%.

- The dry matter percentage did not relate well with oil content, probably because of differences in water content of the fruit grown in the different climate regions.

- Fruit weight ranged from 1.0 to 2.2 g in the North Island and 0.8 to 2.2 g in the South Island

- Colour development for was generally low, with the maturity index ranging from 1.0 to 3.5 in the North Island and 1.0 to 2.5 in the South Island, and the colour maturity index did not relate well to oil content.

5.3.2 Specific regional effects on 'Leccino'

- Maximum oil content ranged from 24 to 48%, with cooler or later maturing regions producing fruit with lower oil content. Oil content in the North Island ranged from 24 to 48% and in the South Island ranged from 27 to 37%.

- The dry matter percentage did not relate well with oil content, probably because of differences in water content of the fruit grown in the different climate regions.

- Fruit weight ranged from 0.9 to 4.4 g in the North Island and 1.0 to 1.9 g in the South Island.

- Colour development was generally moderate to high, with the maturity index ranging from 4 to 5.4 in the North Island and 3 to 4 in the South Island, and the colour maturity index was moderately well related to oil content.

5.3.3 Specific regional effects on 'Koroneiki'

- Mostly grown in the North Island.

- Maximum oil content ranged from 32 to 52%, with cooler or later maturing regions having less effect on total oil content. It is important to note, however, that most sampling was carried out in the Northland and Auckland regions and only one orchard was sampled from each of the more southern Wairarapa and Marlborough regions. Oil content in the North Island ranged from 32 to 54% and in the single orchard in the South Island had fruit with an oil content of 40% at commercial harvest time.

- The dry matter concentration did not relate well with oil content, probably because of differences in water content of the fruit grown in the different climate regions.

- Fruit of this cultivar are generally smaller, and fruit weight ranged from 0.5 to 1.1 g throughout all regions.

- Colour development was generally moderate, with the maturity index ranging from 2.5 to 4 throughout the regions, and the maturity index was moderately well related to oil content for individual orchards, but not when compared across different orchard in different regions.

5.3.4 Specific regional effects on 'Picual'

- Maximum oil content ranged from 27 to 45%, with cooler or later maturing regions producing fruit with lower oil content. Oil content in the North Island ranged from 27 to 45% and fruit from the single orchard in the South Island had an oil content of 30% at commercial harvest time.

- The dry matter concentration did not relate well with oil content, probably because of differences in water content of the fruit grown in the different climate regions.

- Maximum fruit weight ranged from 1.4 to 2.4 g throughout all regions.

- Colour development for 'Picual' was generally variable, with the maturity index ranging from 1 to 4.5, and the colour maturity index was not well related to oil content.

5.3.5   Specific North Island regional effects on 'Barnea'

- Maximum oil content ranged from 30 to 40%.

- The dry matter concentration did not relate well with oil content, probably because of differences in water content of the fruit grown in the different climate regions.

- Maximum fruit weight ranged from 0.9 to 2.3 g throughout the two regions.

- Colour development was generally moderate to low, with the maturity index ranging from 2 to 3.5, and the colour maturity index was not well related to oil content.

5.3.6   Specific North Island regional effects on 'J5'

- Maximum oil content ranged from 47 to 58%, with fruit from the single Northland orchard having higher oil content than those from the single Auckland orchard at commercial harvest time.

- The dry matter concentration did not relate well with oil content, probably because of differences in water content of the fruit grown in the different climate regions.

- Maximum fruit weight ranged from 1.6 to 2.3 g across the two regions.

- Colour development for was generally moderately low, with the maturity index ranging from 2 to 2.5, and the colour maturity index was not well related to oil content.

5.4   The impact of regional rain on percentage dry matter and total phenolics on the 'Frantoio' cultivar

'Frantoio' is the most widely planted olive in New Zealand and we focused more resources and analyses on this cultivar. As has been discussed above, in New Zealand, 'Frantoio' percentage dry matter is not a good indicator of oil content, as it does not reflect advancement in maturity. In addition, percentage dry matter can be influenced by weather conditions such as rainfall. Figure 16 shows the variability in responses on percentage dry matter due to regional rainfall on the different orchards studied. For instance, in the Auckland region the percentage dry matter response to an increase in rainfall in May-June is noticeable for the Simunovich orchard where percentage dry matter dropped from almost 50 to 40%, while for the orchards in the Nelson region, rainfall did not affect dry matter percentage. Once more, these inconsistent changes in percentage dry matter due to environmental moisture make this variable an unreliable tool for prediction of maturity.
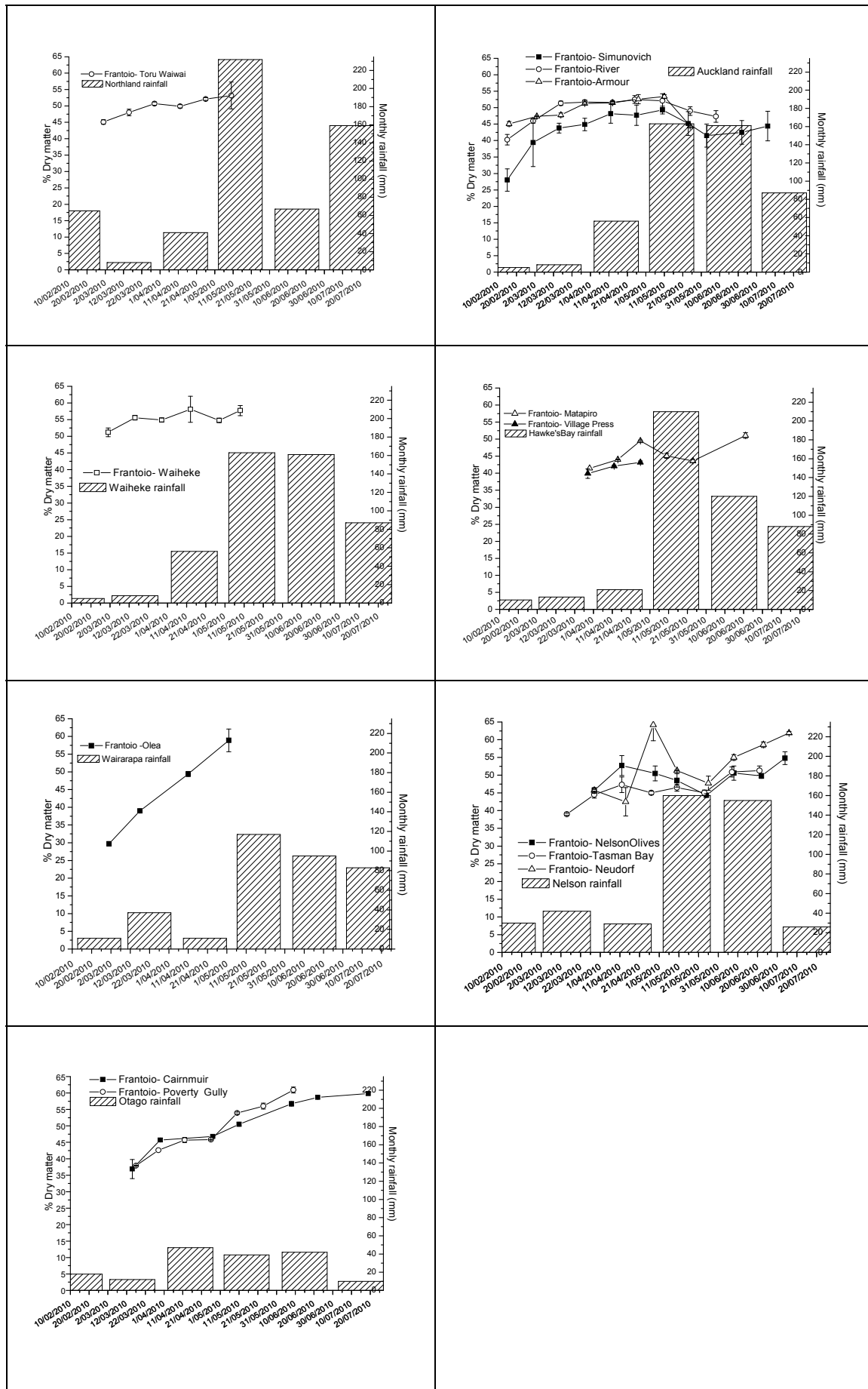
Figure 16. Dry matter of 'Frantoio' fruit and regional monthly rainfall during the 2010 season.

The general trend in all regions of New Zealand is for phenolic content to decrease as the fruit matures. Although individual orchards' rainfall and irrigation data were not collected, Figure 17 also shows that rainfall could also be further contributing to the decrease in total phenolic content in the fruit and therefore the oil. Results found here are in accordance with other research (Ben Temime et al., 2006; Romero et al., 2003).

In some orchards such as in the Waiheke region, the full impact of rainfall on the total phenolic content is not as evident, as the fruit were harvested before the rainy months arrived.

Figure 17. Total phenolic content of 'Frantoio' fruit and monthly regional rainfall during the 2010 season.

5.5 The relationship between % oil content and % dry matter, and % oil content and total solids per fruit (g) for the 'Frantoio' cultivar around New Zealand

As in 2009, the relationship between percentage dry matter and percentage oil content was investigated for 'Frantoio' in 13 New Zealand orchards during the 2010 season. In this season, the combined data collected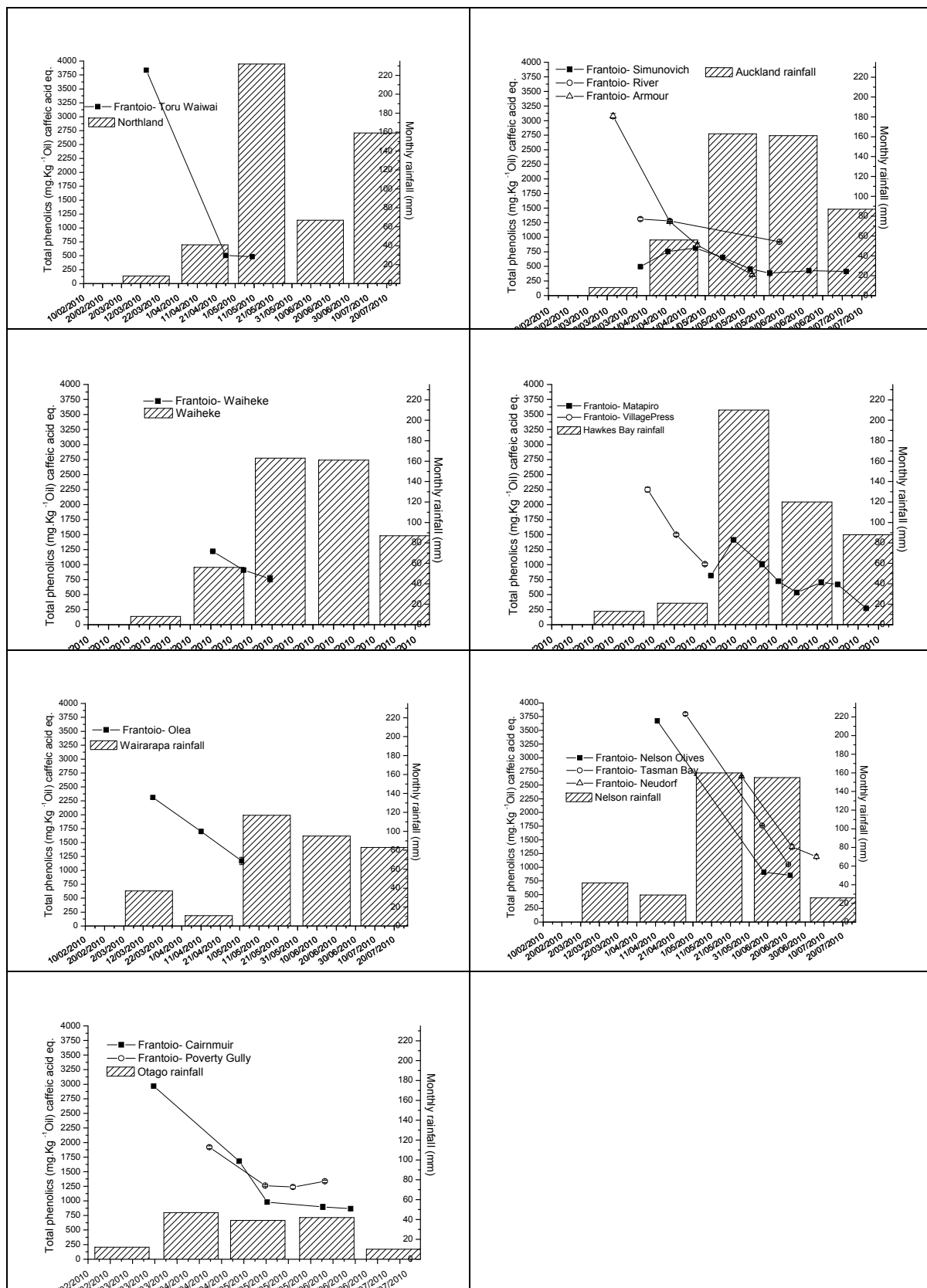 from orchards in the North Island and the South Island were analysed. As reported in 2009 (Requejo-Jackman et al., 2010), in 2010 dry matter measured as a percentage had a very poor correlation to oil content, confirming that this fruit characteristic is not a good predictor of fruit maturity (Figure 18).
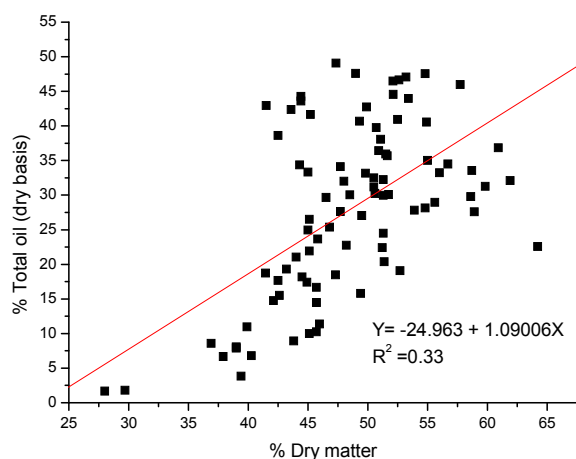
Figure 18. Correlation between percentage total oil (dry basis obtained by solvent extraction) and percentage dry matter for 'Frantoio' in the North Island and South Island during the 2010 season.

We also examined the usefulness of dry matter expressed <u>on a grams per fruit basis</u>, which we will refer to from here on as "total solids" (to avoid confusion with "dry matter" (dry matter on a % basis). Total solids for 'Frantoio' in the North Island and in the South Island during 2010 are shown in Figure 19. The total solids increased everywhere as the season progressed. For the North Island orchards, there was a trend of increasing separation between orchards as the fruit gained total solids at the time of commercial harvest (ranging from 0.5 to 0.95 g). For the South Island orchards, the total solids accumulated in fruit was more consistent across the orchards, ranging from 0.4 to 0.5 g at commercial harvest time. The wider range and variability in total solids gained per fruit in the North Island shows the significant influence of the climate and microclimates in these regions can have on the olive fruit development and maturation rates.
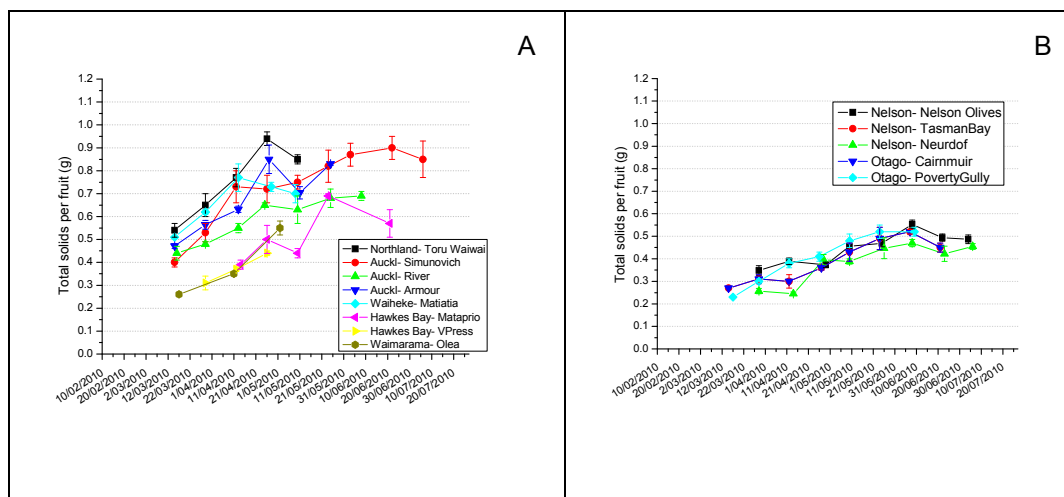
Figure 19. Total solids per fruit (g) for 'Frantoio' in A) North Island and B) South Island during the 2010 season.

- Further analyses found in 2010 again showed a strong correlation between oil content and total solids per fruit for the 'Frantoio' cultivar in both islands of New Zealand (Figure 20a, b).

- The relationship between oil content and the total solids per fruit was strong even when the data were combined for the whole country (Figure 20c).

- The data collected this season confirm that the relationship between oil content and dry matter, measured as a percentage, is not robust, and that total solids per fruit should be used instead to predict fruit oil content.

- The following section (Section 5.6) gives a practical guide to measuring total solids per fruit, which could be used by growers throughout New Zealand.
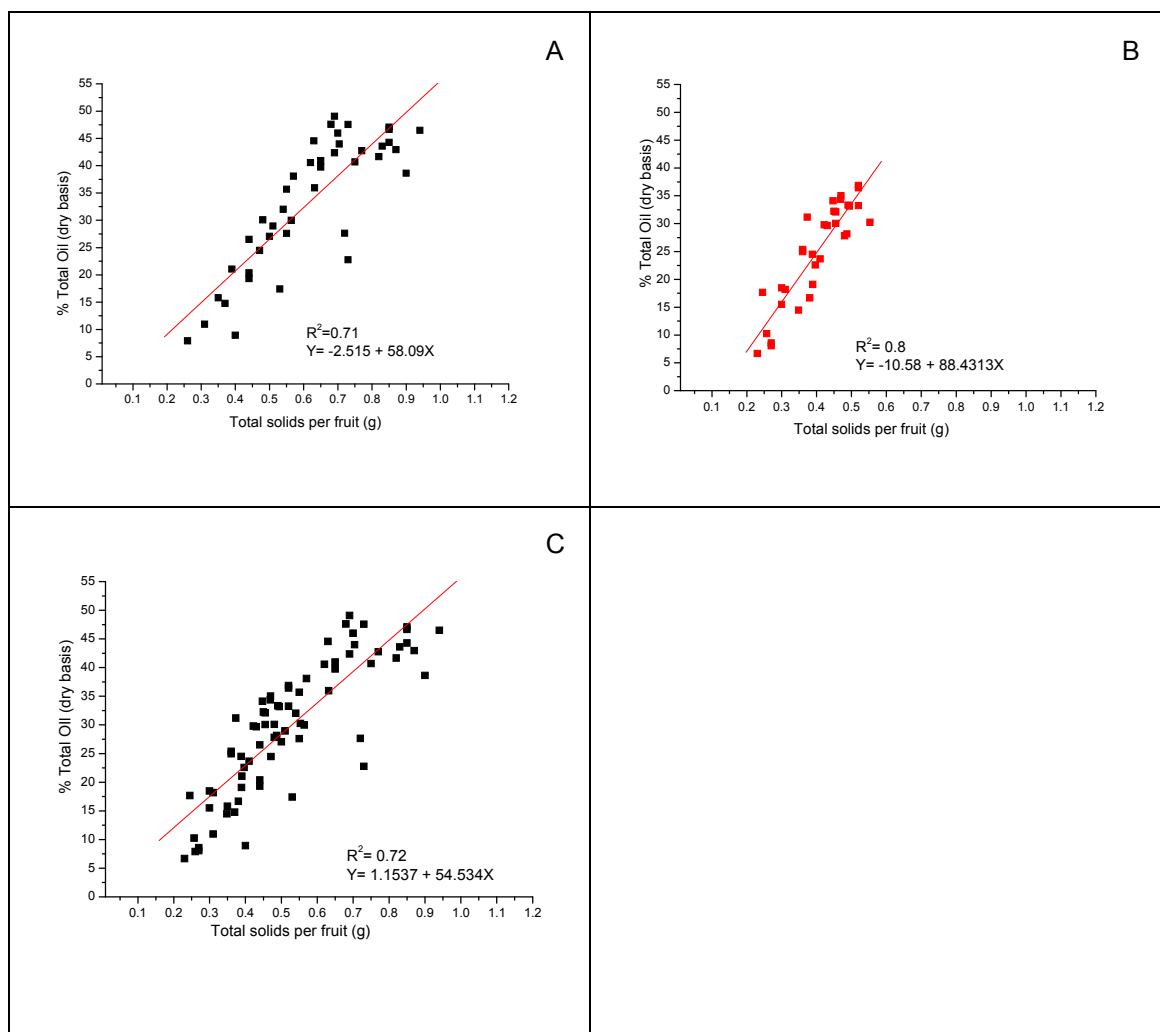
Figure 20. Correlation between total solids per fruit (g) and percentage total oil (dry basis) for 'Frantoio' in A) North Island, B) South Island and C) North Island and South Island combined during the 2010 season.

5.6   Measuring dry matter (in grams of total solids per fruit) at the orchard

From the above work, it seems that one of the useful techniques for following maturity (which correlates with oil content) is that of measuring the total solids (dry matter on a per fruit basis). While this is a relatively simple in a laboratory equipped with an appropriate hammer mill, this is not easy for individual growers, since grinding a whole olive fruit (seed or pit and flesh) is not easy. Thus, we carried out a small trial to determine the effect of tissue preparation and particle size when preparing the olive fruit for dry matter assessment. The aim was to find a practical way to measure dry matter measurements in the field.

A laboratory hammer mill grinder was used to grind the fruit (seed included), while for the cut fruit, a kitchen knife was used to make 3 – 5 slits around the fruit (to aid moisture evaporation). These two ways of preparing the fruit were compared with simply putting whole fruit in the dehydrator. The prepared tissue was placed in a dish in a domestic dehydrator (Hydraflow, Ezidri™, Ultra 1000, Hydraflow Industries Ltd, Upper Hutt, New Zealand) at 60°C.

The time (in hours) to dry fruit was significantly affected by the way in which the fruit was prepared. We took the time to dry the tissue to a point that is within 0.5% dry matter of the minimum value achieved with the ground tissue (44.25%). Ground tissue reached this point after 24 hours, fruit with the skin cut took 48 hours, and whole fruit (no preparation), took 72 hours (Figure 21).
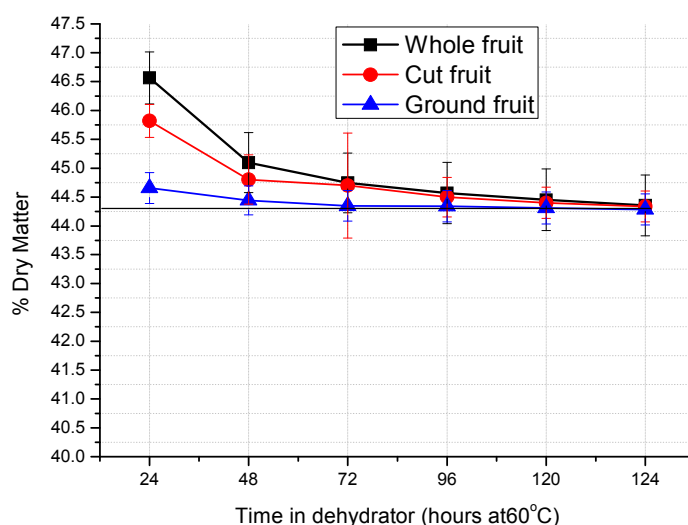
Figure 21. Dry matter content of olive fruit over time in a domestic dehydrator (Hydraflow, Ezidri™) prepared in three different ways. Whole fruit were simply fruit placed in the dehydrator with no preparation at all, cut fruit had four slits cut down the fruit (i.e. cutting the skin), and the ground tissue was the whole fruit (including seed), ground to a paste using a hammer mill.

In the previous year of research (Requejo-Jackman et al., 2010), it was shown that dry matter measured as a percentage does not reflect maturity well and that this relationship can be disturbed by water status of the olive trees (e.g. because of rainfall or drought). This year's results show the same trend, especially for the orchards in the North Island, where changes in dry matter (as a percentage) were not evident after mid March, the stage of fruit maturity where the oil accumulation was at its fastest rate. In last year's report (Requejo-Jackman et al. 2010) we proposed another way of calculating dry matter (as "total solids", total solids per fruit). This measure was more closely related to oil accumulation and therefore a commercially useful measure of fruit maturity.

The above small trial examining various means of measuring dry matter is useful for the grower, since it shows that fruit dry matter can be determined in $\cong 2$ days. Assessment of the fruit dry matter expressed as total solids (dry matter per fruit, in grams), will provide a harvest decision support tool because of its close relationship with oil content.

Based on the data collected in 2009 for the North Island, the following steps will explain how to calculate the dry matter of the fruit (as total solids per fruit in grams):

1. Obtain a random sample of fruit, c. 500 g, from a given cultivar across the orchard/block. The fruit should represent the fruit likely to be harvested. So for hand-harvested fruit, this should reflect all fruit sizes. But if fruit are to be mechanically harvested, then the fruit sample are likely to be the larger fruit and/or fruit which are most likely to be removed by the harvester.

2. Weigh and record 5 empty dishes. Place 20 fruit in each pre-weighed (heat-resistant) dish. A balance with at least one decimal point is required to achieve accurate results. If the balance is less accurate, use a large sample of tissue.

3. Record the weight of each dish containing 20 fruit. Carefully make 3-5 cuts around each fruit using a fine knife.

4. Put in a dehydrator at 60°C for at least 48 hours. A domestic oven or microwave can be used but requires careful attention to not burning the sample. The primary aim is to dry the tissue for as long as it requires to achieve a constant weight. Thus, regular reweighing of the sample is required to establish the time taken to achieve complete drying.

5. Record the weight of each dish containing the 20 dry fruit.

6. The total solids per fruit in grams can be calculated using the formula:

$$= \frac{[(\quad \text{\AA} \quad \text{\AA} \quad \text{\AA} \quad) - (\quad \text{\AA} \quad \text{\AA} \quad)]}{20}$$

Based on the second year of studies, the percentage oil content can be predicted using the relationship between total solids per fruit and oil content (in a dry basis), which is described by the following equation: *Y= -7.51 + 52.3 X* , where Y is the % oil content and X is the total solids per fruit (grams). This equation has an $r^2 = 0.82$, meaning that a one unit change in *X* can explain 0.82 change in *Y.*

This relationship is yet to be confirmed for the South Island orchards.

5.7 Chemical analyses of important phytochemicals in oils

The fatty acid, tocopherol (Vitamin E) and phytosterol composition of ASE-extracted oils was determined for the harvest closest to the commercial harvest of each orchard. For each cultivar, a range of orchards throughout New Zealand was selected. As the sample of fruit supplied was not enough to carry out a cold pressed extraction, solvent-extracted oils were analysed. Since the ASE extraction uses highly non-polar solvents, a higher concentration of the fatty acids, tocopherols and phytosterols are extracted from the olive tissue compared with cold pressed extraction. Thus, ASE-extracted oil from only one harvest, closest to the commercial harvest date, was analysed for tocopherol, sterol and fatty acid composition.

5.7.1 Tocopherols

The tocopherol compositions of selected ASE-extracted oils from various growers and cultivars are shown in Table 6. The predominant tocopherol present in olive oil was alpha-tocopherol, and the remaining tocopherols decreased in the following order: gamma tocopherol > beta-tocopherol > delta tocopherol. In many samples, delta-tocopherol was not detected in the oil. For oils extracted from 'Frantoio', the total tocopherol concentration ranged from 168 to 324 mg/kg. The tocopherol content of 'Frantoio' oils is consistent with previous data for this cultivar from North Island orchards (Requejo-Jackman et al. 2010). The tocopherol concentration was $\cong$ 35% higher in samples from the South Island, except for the sample from Tasman Bay, which is similar to northern North Island concentrations. The sample from Olea in the Wairarapa had 324 mg/kg tocopherol, which was higher than amounts from all the other regions, including the South Island.

As found in the first season of this SFF project (Requejo-Jackman et al. 2009), oil from the 'Leccino' and 'Koroneiki cultivars contained higher amounts of tocopherols than found in 'Frantoio'. Oil from 'Leccino' contained 368-515 mg/kg total tocopherols

Table 8). Again, the highest amounts of tocopherols were observed from the Olea orchard in the Wairarapa for the 'Leccino' sample. There was no clear distinction between North Island and South Island regions in terms of total tocopherols for 'Leccino' oil. Delta-tocopherol was detected in four 'Leccino' samples, which were all from the North Island regions. For 'Koroneiki', the total tocopherol content ranged from 274 to 485 mg/kg. The highest recorded value, from Armour orchard in Auckland, agreed with results reported in Year 1 of this project (Requejo-Jackman et al., 2009) for the same orchard.

Oil samples extracted from 'Barnea' had much higher concentrations of tocopherols than other cultivars, ranging from 419 to 758 mg/kg. All samples were from North Island orchards in Hawke's Bay and Wairarapa. The total tocopherol content in oil extracted from 'Picual' ranged from 306 to 487 mg/kg. Oil from 'J5' had lower tocopherol concentrations, 155-272 mg/kg, at similar rates to 'Frantoio' oil.

The Village Press samples were found to be inconsistent with results from other orchards. The 'Frantoio' sample content was more consistent with the tocopherols found in 'Barnea' fruit. The 'Picual' sample from The Village Press also showed similar tocopherols concentrations to a 'Barnea' cultivar. This was also observed in the sterol results (Table 9).

Table 8. Concentration of tocopherols in Accelerated Solvent Extraction (ASE)-extracted olive oils. 2010. Mean ± SEM (mg/kg) (ND = not detected).

Cultivar	Grower	Region	Harvest	Alpha-tocopherol	Beta-tocopherol	Gamma-tocopherol	Delta-tocopherol	Total Tocopherols
'Frantoio'	Toru Waiwai	Northland	26/04	192.55 ± 0.51	2.76 ± 0.06	4.39 ± 0.49	ND	199.69 ± 1.06
	River	Auckland	08/06	163.31 ± 1.89	1.79 ± 0.05	3.63 ± 0.11	ND	168.73 ± 1.83
	Armour	Auckland	25/05	188.37 ± 0.27	1.00 ± 0.42	4.10 ± 0.24	ND	193.47 ± 0.44
	Simunovich	Auckland	22/06	175.59 ± 0.44	1.93 ± 0.04	3.02 ± 0.09	ND	180.55 ± 0.5
	Matiatia	Waiheke	10/05	166.48 ± 1.86	0.89 ± 0.31	4.81 ± 0.45	ND	172.18 ± 2.6
	Matapiro	Hawke's Bay	30/06	168.84 ± 0.30	2.01 ± 0.08	5.55 ± 0.19	ND	176.07 ± 0.41
	The Village Press	Hawke's Bay	26/04	412.55 ± 0.45	4.34 ± 0.10	12.01 ± 0.10	ND	428.90 ± 0.45
	Olea	Wairarapa	02/05	315.73 ± 0.98	3.18 ± 0.05	5.51 ± 0.01	ND	324.42 ± 1.02
	Nelson Olives	Nelson	22/06	266.37 ± 1.44	2.97 ± 0.05	8.56 ± 0.00	ND	277.90 ± 1.39
	Tasman Bay	Nelson	21/06	182.61 ± 0.31	2.13 ± 0.02	3.91 ± 0.18	ND	188.65 ± 0.51
	Neudorf	Nelson	06/07	277.42 ± 3.16	2.00 ± 0.25	11.16 ± 0.60	ND	290.58 ± 2.98
	Cairnmuir	Central Otago	22/06	254.96 ± 0.00	2.94 ± 0.19	7.93 ± 0.11	ND	265.83 ± 0.30
	Poverty Gully	Central Otago	09/06	261.82 ± 0.21	2.42 ± 0.02	8.07 ± 0.22	ND	272.32 ± 0.46
'Leccino'	Quail Ridge	Northland	30/03	339.19 ± 4.95	4.49 ± 0.72	22.36 ± 0.53	0.04 ± 0.02	366.08 ± 5.93
	Toru Waiwai	Northland	26/04	367.85 ± 10.27	4.39 ± 0.12	25.10 ± 1.28	ND	397.35 ± 11.67
	Matiatia	Waiheke	25/05	342.53 ± 0.45	3.63 ± 0.24	24.22 ± 0.98	0.10 ± 0.10	370.47 ± 1.78
	Putiki Grove	Waiheke	09/05	348.36 ± 0.12	4.97 ± 0.88	39.85 ± 8.61	0.28 ± 0.21	396.08 ± 8.17
	Armour	Auckland	25/05	398.10 ± 0.08	4.51 ± 0.08	20.60 ± 0.28	ND	423.21 ± 0.43
	Matapiro	Hawke's Bay	21/06	381.59 ± 1.59	2.95 ± 0.05	26.88 ± 0.79	0.09 ± 0.09	411.51 ± 0.85
	Olea	Wairarapa	02/05	493.99 ± 0.13	4.48 ± 0.07	17.28 ± 0.26	ND	515.75 ± 0.20
	Lusatori	Marlborough	08/06	458.55 ± 0.06	3.73 ± 0.15	22.29 ± 1.16	ND	484.57 ±1.24
	Nelson	Nelson	22/06	365.29 ± 1.58	3.32 ± 0.03	18.46 ± 0.02	ND	387.07 ± 1.58
	Tasman Bay	Nelson	21/06	383.45 ± 17.04	1.74 ± 1.22	35.07 ± 1.45	ND	420.25 ± 19.35
	Cairnmuir	Central Otago	22/06	359.86 ± 0.00	3.75 ± 0. 36	20.55 ± 1.77	ND	384.16 ± 2.13
'Koroneiki'	Quail Ridge	Northland	30/4	381.77 ± 0.02	4.59 ± 0.01	11.10 ± 0.15	ND	397.46 ± 0.16
	Toru Waiwai	Northland	10/05	294.55 ± 0.86	3.54 ± 0.39	7.68 ± 0.07	ND	305.77 ±1.19
	River	Auckland	25/05	321.58 ± 1.85	5.68 ± 0.05	7.09 ± 0.21	ND	334.35 ±1.70
	Armour	Auckland	25/05	463.75 ± 1.15	6.24 ± 0.34	15.91 ± 5.28	ND	485.90 ± 4.46
	Stone Valley	Wairarapa	22/06	265.45 ± 0.67	2.41 ± 0.20	7.04 ± 0.30	ND	274.90 ± 0.57
	Lusatori	Marlborough	23/06	369.72 ± 0.72	4.18 ± 0.18	12.91 ± 0.14	ND	386.82 ± 0.76
'Barnea'	Matapiro	Hawke's Bay	24/05	398.49 ± 8.88	4.23 ± 0.45	18.32 ± 0.25	ND	421.04 ± 9.17
	The Village Press	Hawke's Bay	26/04	719.10 ± 0.84	9.31 ± 0.72	29.80 ± 2.51	ND	758.29 ± 2.55
	Waimarama	Hawke's Bay	08/06	381.31 ± 10.39	9.27 ± 1.84	28.34 ± 2.88	0.26 ± 0.07	419.17 ± 11.15
	Olivo	Wairarapa	24/05	587.18 ± 0.57	6.86 ± 0.09	27.19 ± 0.09	ND	621.23 ± 0.57
'Picual'	Putiki	Waiheke	0905	253.23 ±0.37	1.94 ± 0.11	51.52 ± 0.46	0.08 ± 0.08	306.78 ±0.13
	Simunovich	Auckland	06/07	291.27 ± 0.54	2.34 ± 0.40	74.20 ± 0.86	0.27 ± 0.01	368.08 ± 1.78
	The Village Press	Hawke's Bay	26/04	449.60 ± 13.01	6.36 ± 0.88	31.22 ± 2.79	ND	487.18 ±11.10
	Tasman Bay	Nelson	26/06	401.29 ± 1.39	3.88 ± 0.33	32.23 ± 0.15	ND	437.40 ± 0.92
'J5'	Quail Ridge	North Auckland	29/03	259.93 ± 31.53	2.59 ± 0.10	9.85 ± 4.68	ND	272.38 ± 1.01
	Simunovich	Auckland	06/07	150.55 ± 0.22	1.32 ± 0.06	3.85 ± 0.40	ND	155.72 ± 0.56

5.7.2 Sterol content

The sterol composition of selected ASE-extracted oils are presented in two ways, in terms of % individual sterols (%w/w; Table 9) and as absolute concentrations (mg/kg oil; Table 10). IOC standards for virgin olive oils recommend limits for the percentages of individual sterols in the oils (IOC, 2010). Absolute concentrations (mg/kg) will provide information on the actual amount of sterols present in the oil (not the % of each sterol present). As mentioned earlier, more of the phytochemicals present are extracted with solvent extraction than would be extracted using cold pressed extraction. The increase in extraction is on average 20% greater in solvent-extracted oils than in cold pressed oils. Hence, the % campesterol detected in all the cultivars except 'Barnea' would have been well within the IOC standard of $\leq 4\%$, and the standard of $\leq 4.8\%$ as recently suggested in the proposed Australian/New Zealand standards. 'Barnea' ASE-extracted oil was found to contain 5.03 – 6.32% campesterol. 'Koroneiki' ASE-extracted oils contained the second highest concentrations of campesterol, ranging from 3.94 to 5.53%. All oils except a 'Leccino' oil sample from Olea, Wairarapa, were found to have apparent beta-sitosterol concentrations > 92.5%.

'Frantoio' and 'Leccino' oils from the North Island had lower concentrations of campesterol and conversely higher concentrations of beta-sitosterol than the same cultivar from South Island orchards. This trend was more obvious when the absolute concentrations (mg/kg oil) of sterols in the oils were compared (Table 10). These trends have only been found in one season, and another season of data collection would be preferable to confirm this impact of growing region on sterol composition. In terms of absolute concentrations of sterols, the total sterol concentration found in all cultivars ranged from 685 to 1872 mg/kg. 'Koroneiki' oil was found to contain the lowest concentration of total sterols, ranging from 685 to 1114 mg/kg. Total sterols in 'Frantoio' oil ranged from 899 to 1142 mg/kg, 'Leccino' oils from 1046 to 1547 mg/kg, 'Barnea' oils from 1402 to 1873 mg/kg, 'Picual' from 1402 to 2140 mg/kg and 'J5' from 1124 to 1426 mg/kg. Thus, the sterol composition of each oil was strongly influenced by the cultivar.

5.7.3 Fatty Acid Composition

The fatty acid compositions of the ASE extraction oils are presented in Table 11, expressed as % w/w as methyl esters. All the oils analysed here were ASE extracted. We know that with solvent extraction, higher proportions of fatty acids can be extracted from the tissue. Hence some oils appeared to be over the IOC limits for alpha-linolenic acid (greater than 1%w/w). It is reported that fruit will produce more unsaturated fatty acids in colder climates; saturated fatty acids have higher melting points than unsaturated fatty acids; hence in colder climates the plants are adapted to produce more unsaturated fatty acids to reduce cell rupture (Harwood et al., 1994). 'Frantoio', 'Leccino' and 'Picual' fruit produced oils with higher amounts of alpha linolenic acid in Hawke's Bay and regions south of this. For the same cultivars, higher concentrations of palmitic acid (saturated C16:0)) were found in the extracted oils. 'Koroneiki' produced oil with the highest percentage of oleic acid and there was very little difference between oil extracted from fruit from the north of the North Island and oil from fruit harvested from Marlborough. Other than 'Koroneiki', it was very hard to find differences between the other cultivars.

Table 9. Concentration of sterols in Accelerated Solvent Extraction (ASE)-extracted olive oils. 2010. Mean ± SEM (%w/w) (ND = not detected).

Cultivar	Grower	Region	Harvest	Campesterol	Stigmasterol	Clerosterol	beta.-Sitosterol	Sitostanol	5-avenasterol	5,24 stigmastedienol	7-stigmastenol	7-Avenasterol	Apparent beta-sitosterol
'Frantoio'	Toru Waiwai	Northland	26/04	2.71 ± 0.04	0.65 ± 0.01	0.67 ± 0.00	84.87 ± 0.07	1.50 ± 0.04	7.87 ± 0.02	0.70 ± 0.04	ND	1.03 ± 0.02	96.64 ± 0.04
	River	Auckland	08/06	2.49 ± 0.03	0.84 ± 0.02	0.48 ± 0.13	84.44 ± 0.48	1.97 ± 0.06	8.63 ± 0.01	0.54 ± 0.03	ND	0.60 ± 0.20	96.66 ± 0.04
	Armour	Auckland	25/05	2.58 ± 0.01	0.61 ± 0.01	0.38 ± 0.14	87.82 ± 0.13	0.98 ± 0.01	6.85 ± 0.03	0.43 ± 0.01	ND	0.35 ± 0.00	96.81 ± 0.00
	Simunovich	Auckland	22/06	4.50 ± 0.09	0.93 ± 0.02	0.57 ± 0.04	78.47 ± 0.25	3.47 ± 0.05	11.13 ± 0.06	0.46 ± 0.01	ND	0.47 ± 0.01	94.57 ± 0.11
	Matiatia	Waiheke	10/05	2.97 ± 0.04	0.63 ± 0.02	0.53 ± 0.03	81.70 ± 0.01	2.89 ± 0.16	8.81 ± 0.09	0.49 ± 0.01	0.33 ± 0.05	1.64 ± 0.05	96.40 ± 0.07
	Matapiro	Hawke's Bay	30/06	4.76 ± 0.12	1.10 ± 0.02	0.51 ± 0.02	79.50 ± 0.29	3.35 ± 0.03	10.07 ± 0.10	0.31 ± 0.01	ND	0.4 ± 0.03	94.14 ± 0.11
	The Village Press	Hawke's Bay	26/04	5.05 ± 0.04	2.32 ± 0.04	0.72 ± 0.11	83.11 ± 0.10	4.67 ± 0.14	3.56 ± 0.07	0.32 ± 0.00	ND	0.24 ± 0.01	92.63 ± 0.00
	Olea	Wairarapa	02/05	4.69 ± 0.06	0.91 ± 0.01	0.72 ± 0.00	85.71 ± 0.10	4.46 ± 0.03	2.89 ± 0.06	0.03 ± 0.01	ND	0.31 ± 0.01	94.40 ± 0.07
	Nelson Olives	Nelson	22/06	3.87 ± 0.01	0.96 ± 0.02	0.53 ± 0.15	82.74 ± 0.07	4.69 ± 0.04	6.42 ± 0.07	0.30 ± 0.01	ND	0.48 ± 0.02	95.17 ± 0.04
	Tasman Bay	Nelson	21/06	4.04 ± 0.11	1.10 ± 0.01	0.55 ± 0.14	82.12 ± 0.19	4.80 ± 0.05	6.65 ± 0.00	0.32 ± 0.00	ND	0.42 ± 0.01	94.87 ± 0.09
	Neudorf	Nelson	06/07	3.90 ± 0.16	0.95 ± 0.06	0.62 ± 0.04	83.96 ± 0.05	5.11 ± 0.13	4.99 ± 0.08	0.25 ± 0.02	ND	0.22 ± 0.04	95.15 ± 0.22
	Cairnmuir	Central Otago	22/06	4.30 ± 0.01	0.81 ± 0.01	0.56 ± 0.15	84.14 ± 0.20	3.09 ± 0.03	6.63 ± 0.06	0.31 ± 0.01	ND	0.16 ± 0.01	94.89 ± 0.00
	Poverty Gully	Central Otago	09/06	3.69 ± 0.04	0.59 ± 0.01	0.35 ± 0.03	80.75 ± 0.13	4.92 ± 0.07	9.08 ± 0.20	0.30 ± 0.01	ND	0.32 ± 0.01	95.71 ± 0.03
'Leccino'	Quail Ridge	Northland	30/03	3.20 ± 0.15	1.69 ± 0.07	0.57 ± 0.11	86.33 ± 0.48	1.40 ± 0.03	5.24 ± 0.10	0.50 ± 0.03	ND	1.09 ± 0.09	95.12 ± 0.21
	Toru Waiwai	Northland	26/04	2.79 ± 0.04	1.60 ± 0.03	0.74 ± 0.02	84.34 ± 0.15	0.89 ± 0.01	7.56 ± 0.02	0.56 ± 0.05	ND	1.53 ± 0.05	95.61 ± 0.00
	Matiatia	Waiheke	25/05	1.99 ± 0.03	0.98 ± 0.03	0.69 ± 0.02	80.74 ± 0.13	0.77 ± 0.16	11.84 ± 0.28	0.62 ± 0.00	ND	2.37 ± 0.09	97.03 ± 0.06
	Putiki Grove	Waiheke	09/05	2.18 ± 0.01	1.24 ± 0.03	0.38 ± 0.01	83.25 ± 0.06	0.77 ± 0.14	10.18 ± 0.06	0.64 ± 0.01	0.02 ± 0.00	1.35 ± 0.08	96.58 ± 0.04
	Armour	Auckland	25/05	2.61 ± 0.00	1.53 ± 0.03	0.56 ± 0.15	80.03 ± 0.20	0.95 ± 0.02	12.41 ± 0.08	0.64 ± 0.03	ND	1.26 ± 0.01	95.86 ± 0.03
	Matapiro	Hawke's Bay	21/06	3.47 ± 0.05	1.47 ± 0.07	0.70 ± 0.00	84.31 ± 0.27	1.48 ± 0.16	7.70 ± 0.24	0.44 ± 0.01	ND	0.43 ± 0.01	95.06 ± 0.13
	Olea	Wairarapa	02/05	5.10 ± 0.01	2.60 ± 0.07	0.60 ± 0.01	83.13 ± 0.17	3.36 ± 0.01	4.09 ± 0.10	0.37 ± 0.01	ND	0.74 ± 0.03	92.30 ± 0.09

Cultivar	Grower	Region	Harvest	Campesterol	Stigmasterol	Clerosterol	beta.-Sitosterol	Sitostanol	5-avenasterol	5,24 stigmastedienol	7-stigmastenol	7-Avenasterol	Apparent beta-sitosterol
	Lusatori	Marlborough	08/06	3.81	1.19	0.43	83.93	1.66	7.95	0.37	ND	0.65	95.00
	Nelson	Nelson	22/06	3.67 ± 0.01	1.60 ± 0.10	0.45 ± 0.03	84.17 ± 0.04	1.73 ± 0.04	7.40 ± 0.12	0.44 ± 0.03	ND	0.54 ± 0.03	94.73 ± 0.11
	Tasman Bay	Nelson	21/06	3.76 ± 0.12	0.96 ± 0.06	0.68 ± 0.06	86.57 ± 0.66	3.38 ± 0.17	4.08 ± 0.19	0.27 ± 0.02	ND	0.30 ± 0.06	95.28 ± 0.17
	Cairnmuir	Central Otago	22/06	4.33 ± 0.08	1.68 ± 0.02	0.79 ± 0.00	79.77 ± 0.01	1.90 ± 0.05	10.79 ± 0.03	0.35 ± 0.01	ND	0.39 ± 0.05	93.98 ± 0.11
'Koroneiki'	Quail Ridge	Northland	30/4	4.08 ± 0.04	0.75 ± 0.03	0.29 ± 0.01	82.81 ± 0.03	0.60 ± 0.15	10.75 ± 0.06	0.50 ± 0.03	ND	0.22 ± 0.03	95.17 ± 0.01
	Toruwaiwai	Northland	10/05	3.94 ± 0.05	1.06 ± 0.01	0.40 ± 0.01	79.48 ± 0.20	0.79 ± 0.00	13.39 ± 0.11	0.62 ± 0.06	ND	0.32 ± 0.02	95.00 ± 0.04
	River	Auckland	25/05	4.19 ± 0.04	1.57 ± 0.02	0.67 ± 0.01	78.91 ± 0.15	1.58 ± 0.01	12.33 ± 0.07	0.51 ± 0.01	ND	0.24 ± 0.03	94.24 ± 0.06
	Armour	Auckland	25/05	4.29 ± 0.01	1.59 ± 0.01	0.73 ± 0.01	84.41 ± 0.27	0.82 ± 0.16	7.59 ± 0.08	0.41 ± 0.00	ND	0.16 ± 0.00	94.12 ± 0.03
	Stone Valley	Wairarapa	22/06	4.64 ± 0.04	0.90 ± 0.03	0.18 ± 0.02	83.79 ± 0.42	1.84 ± 0.25	8.04 ± 0.04	0.35 ± 0.03	ND	0.16 ± 0.00	94.46 ± 0.07
	Lusatori	Marlborough	23/06	5.53 ± 0.03	0.90 ± 0.02	0.39 ± 0.03	84.59 ± 0.09	1.66 ± 0.03	6.44 ± 0.01	0.30 ± 0.01	ND	0.20 ± 0.02	93.58 ± 0.01
'Barnea'	Matapiro	Hawke's Bay	24/05	5.44 ± 0.21	0.75 ± 0.11	0.64 ± 0.07	88.42 ± 0.78	1.37 ± 0.12	2.96 ± 0.22	0.24 ± 0.01	ND	0.17 ± 0.05	93.81 ± 0.33
	The Village Press	Hawke's Bay	26/04	6.29 ± 0.02	1.26 ± 0.01	0.62 ± 0.14	87.06 ± 0.23	2.74 ± 0.03	1.64 ± 0.03	0.22 ± 0.01	ND	0.16 ± 0.01	92.45 ± 0.01
	Waimarama	Hawke's Bay	08/06	5.03 ± 0.09	2.24 ± 0.10	0.56 ± 0.10	85.78 ± 0.47	1.18 ± 0.06	4.69 ± 0.13	0.32 ± 0.01	ND	0.20 ± 0.03	92.73 ± 0.19
	Olivo	Wairarapa	24/05	6.32 ± 0.03	1.00 ± 0.05	0.63 ± 0.10	84.7 ± 0.10	4.22 ± 0.01	2.55 ± 0.04	0.22 ± 0.02	ND	0.36 ± 0.02	92.68 ± 0.02
'Picual'	Putiki	Waiheke	0905	2.36 ± 0.02	0.38 ± 0.01	0.50 ± 0.16	86.53 ± 0.15	2.68 ± 0.01	5.63 ± 0.30	0.43 ± 0.02	0.06 ± 0.03	1.43 ± 0.03	97.27 ± 0.03
	Simunovich	Auckland	06/07	3.98 ± 0.08	0.72 ± 0.01	0.59 ± 0.13	87.83 ± 0.00	2.64 ± 0.04	3.84 ± 0.03	0.19 ± 0.01	ND	0.22 ± 0.02	95.30 ± 0.07
	The Village Press	Hawke's Bay	26/04	3.97 ± 0.03	1.35 ± 0.00	0.78 ± 0.08	86.65 ± 0.04	4.97 ± 0.00	1.69 ± 0.01	0.21 ± 0.01	0.03 ± 0.01	0.36 ± 0.00	94.69 ± 0.03
	Tasman Bay	Nelson	26/06	3.90 ± 0.06	1.05 ± 0.07	0.68 ± 0.03	86.04 ± 0.47	3.07 ± 0.06	4.63 ± 0.19	0.28 ± 0.04	ND	0.34 ± 0.03	95.04 ± 0.13
'J5'	Quail Ridge	North Auckland	29/03	3.08 ± 0.04	0.89 ± 0.01	0.57 ± 0.15	85.13 ± 0.15	3.31 ± 0.03	5.29 ± 0.03	0.54 ± 0.04	0.02 ± 0.00	1.18 ± 0.02	96.02 ± 0.05
	Simunovich	Auckland	06/07	3.22 ± 0.13	0.59 ± 0.02	0.47 ± 0.12	85.41 ± 0.12	1.66 ± 0.07	8.08 ± 0.07	0.36 ± 0.00	ND	0.21 ± 0.01	96.19 ± 0.15

Table 10: Concentration of sterols Sterols in Accelerated Solvent Extraction (ASE)-extracted olive oils. 2010. Mean ± SEM (mg/kg) (ND = not detected).

Cultivar	Grower	Region	Harvest Date	Campesterol	Stigmasterol	Clerosterol	beta.-Sitosterol	Sitostanol	5-avenasterol	5,24 stigmastedienol	7-stigmastenol	7-Avenasterol	Total Sterols
'Frantoio'	Toru Waiwai	Northland	26/04	28.81 ± 0.07	6.87 ± .018	7.08 ± 0.06	902.47 ± 13.25	15.93 ± 0.16	83.71 ± 1.36	7.46 ± 0.34	ND	11.01 ± 0.40	1063.34 ± 14.68
	River	Auckland	08/06	22.43 ± 0.26	7.59 ± 0.17	4.32 ± 0.17	759.43 ± 3.62	4.84 ± 0.55	77.59 ± 0.15	4.84 ± 0.26	ND	5.41 ± 1.84	899.36 ± 0.79
	Armour	Auckland	25/05	29.48 ± 0.43	6.97 ± 0.27	4.38 ± 1.73	1003.45 ± 16.50	11.17 ± 0.14	78.29 ± 1.08	4.93 ± 0.19	ND	4.01 ± 0.10	1142.68 ± 20.44
	Simunovich	Auckland	22/06	44.85 ± 0.91	9.31 ± 0.24	5.69 ± 0.40	782.86 ± 1.84	34.63 ± 0.52	110.99 ± 0.65	4.58 ± 0.03	ND	4.7 ± 0.03	997.61 ± 0.88
	Matiatia	Waiheke	10/05	26.88 ± 0.08	5.68 ± 0.14	4.77 ± 0.20	739.47 ±8.30	26.19 ± 0.45	79.79 ±1.72	4.40 ± 0.02	3.03 ± 0.39	14.88 ± 0.64	905.08 ±10.28
	Matapiro	Hawke's Bay	30/06	41.57 ± 0.55	9.62 ±0.28	4.45 ± 0.15	694.40 ± 11.23	29.30 ± 0.13	87.91 ± 0.22	2.68 ± 0.02	ND	3.51 ± 0.20	873.43 ± 10.94
	The Village Press	Hawke's Bay	26/04	94.57 ± 1.16	43.50 ± 0.51	13.46 ± 2.03	1556.06 ± 8.66	87.42 ± 2.25	66.62 ± 1.09	6.00 ± 0.05	ND	4.58 ± 0.21	1872.20 ± 8.17
	Olea	Wairarapa	02/05	58.60 ± 1.22	11.35 ± 0.25	9.01 ± 0.03	1069.59 ± 8.12	55.69 ± 0.06	36.13 ± 1.04	3.72 ± 0.19	ND	3.86 ± 0.03	1247.96 ± 10.88
	Nelson Olives	Nelson	22/06	43.59 ± 0.72	10.80 ± 0.40	6.00 ± 1.77	930.66 ± 11.10	52.75 ± 0.27	72.25 ± 0.14	3.37 ± 0.10	ND	5.44 ± 0.14	1124.87 ± 14.35
	Tasman Bay	Nelson	21/06	46.19 ± 1.34	12.54 ± 0.15	6.30 ± 1.64	939.67 ± 0.46	54.90 ± 0.50	76.12 ± 0.17	3.66 ± 0.02	ND	4.84 ± 0.13	1143.92 ± 2.15
	Neudorf	Nelson	06/07	40.77 ± 2.10	9.93 ± 0.70	6.43 ± 0.50	876.03 ± 5.01	53.40 ± 1.91	52.13 ± 1.28	2.56 ± 0.22	ND	2.27 ± 0.48	1043.54 ± 11.43
	Cairnmuir	Central Otago	22/06	44.31 ± 0.67	8.36 ± 0.00	5.76 ± 1.60	866.95 ± 9.46	31.82 ± 0.16	68.31 ± 1.55	3.24 ± 0.19	ND	1.67 ± 0.13	1030.41 ± 13.75
	Poverty Gully	Central Otago	09/06	42.07 ± 0.54	6.76 ± 0.12	3.99 ± 0.28	919.99 ± 0.62	56.04 ± 0.71	103.4 ± 2.53	3.41 ± 0.17	ND	3.63 ± 0.12	1139.29 ± 2.64
'Leccino'	Quail Ridge	Northland	30/03	34.75 ± 1.54	18.32 ± 0.68	6.15 ± 1.20	938.83 ± 10.03	15.22 ± 0.42	56.93 ± 1.20	5.42 ± 0.29	ND	11.85 ± 0.96	1087.47 ± 8.29
	Toru Waiwai	Northland	26/04	31.82 ± 0.64	18.32 ± 0.24	8.42 ± 0.25	963.23 ± 5.17	10.12 ± 0.13	86.31 ± 0.89	6.35 ± 0.57	ND	17.48 ± 0.74	1142.04 ± 8.14
	Matiatia	Waiheke	25/05	22.78 ± 0.19	13.01 ± 0.39	3.94 ± 0.04	870.90 ± 2.97	8.04 ± 1.53	106.48 ± 0.19	6.73 ± 0.18	0.18 ± 0.00	14.08 ± 0.74	1046.14 ± 4.28
	Putiki Grove	Waiheke	09/05	22.23 ± 0.45	10.29 ± 0.38	7.71 ± 0.20	900.22 ± 4.62	8.55 ± 1.79	132.01 ± 2.67	6.90 ± 0.02	ND	26.46 ± 0.93	1115.01 ± 3.87
	Armour	Auckland	25/05	29.96 ± 0.05	17.52 ± 0.40	6.48 ± 1.66	918.45 ± 4.41	10.95 ± 0.21	142.44 ± 0.58	7.39 ± 0.33	ND	14.49 ± 0.11	1147.67 ± 2.62
	Matapiro	Hawke's Bay	21/06	39.50 ± 0.57	16.68 ± 0.82	8.00 ± 0.02	959.05 ± 1.71	16.83 ± 1.88	87.64 ± 2.80	4.97 ± 0.13	ND	4.93 ± 0.16	1137.59 ± 1.58
	Olea	Wairarapa	02/05	78.95 ± 0.64	40.17 ± 1.33	9.35 ± 0.18	1286.27 ± 4.07	51.98 ± 0.14	63.34 ± 1.91	5.74 ± 0.21	0.04 ± 0.04	11.46 ± 0.46	1547.31 ± 7.99
	Lusatori	Marlborough	08/06	50.73	15.90	5.78	1118.00	22.17	105.86	4.95	ND	8.68	1332.08

Cultivar	Grower	Region	Harvest Date	Campesterol	Stigmasterol	Clerosterol	beta.-Sitosterol	Sitostanol	5-avenasterol	5,24 stigmastedienol	7-stigmastenol	7-Avenasterol	Total Sterols
	Nelson	Nelson	22/06	42.82 ± 0.23	18.72 ± 1.02	5.30 ± 0.28	982.53 ± 7.64	20.17 ± 0.56	86.39 ± 1.98	5.08 ± 0.28	ND	6.32 ± 0.34	1167.33 ± 8.49
	Tasman Bay	Nelson	21/06	49.26 ± 1.81	12.56 ± 0.79	8.89 ± 0.77	1132.89 ± 4.57	44.24 ± 2.45	53.5 ± 2.83	3.52 ± 0.23	ND	3.95 ± 0.78	1308.8 ± 8.58
	Cairnmuir	Central Otago	22/06	56.57 ± 1.17	22.00 ± 0.36	10.28 ± 0.03	1041.80 ± 2.12	24.84 ± 0.64	140.88 ± 0.12	4.55 ± 0.18	ND	5.10 ± 0.62	1306.02 ± 2.48
'Koroneiki'	Quail Ridge	Northland	30/4	30.76 ± 0.36	5.65 ± 0.19	2.22 ± 0.05	623.77 ± 1.27	4.51 ± 1.12	80.97 ± 0.24	3.73 ± 0.24	ND	1.65 ± 0.26	753.26 ± 1.77
	Toru Waiwai	Northland	10/05	32.50 ± 0.56	8.77 ± 0.04	3.31 ± 0.08	656.05 ± 1.52	6.55 ± 0.02	110.52 ± 1.41	5.12 ± 0.56	ND	2.64 ± 0.16	825.47 ± 3.95
	River	Auckland	25/05	31.94 ± 0.25	11.96 ± 0.16	5.14 ± 0.08	601.66 ± 1.78	12.01 ± 0.06	94.01 ± 0.42	3.85 ± 0.11	ND	1.86 ± 0.25	762.42 ± 0.80
	Armour	Auckland	25/05	47.79 ± 0.36	17.75 ± 0.19	8.18 ± 0.13	940.53 ± 0.89	9.18 ± 1.80	84.58 ± 1.28	4.52 ± 0.03	ND	1.80 ± 0.04	1114.32 ± 4.67
	Stone Valley	Wairarapa	22/06	31.79 ± 0.53	6.17 ± 0.27	1.25 ± 0.17	574.11 ± 2.36	13.34 ± 1.86	55.06 ± 0.81	2.37 ± 0.26	ND	1.09 ± 0.03	685.19 ± 6.28
	Lusatori	Marlborough	23/06	47.20 ± 0.13	7.66 ± 0.25	3.29 ± 0.20	722.50 ± 6.77	14.19 ± 0.12	55.00 ± 0.34	2.55 ± 0.11	ND	1.69 ± 0.19	854.08 ± 7.08
'Barnea'	Matapiro	Hawke's Bay	24/05	76.28 ± 2.93	10.56 ± 1.58	9.03 ± 1.02	1240.51 ± 12.17	19.22 ± 1.64	41.52 ± 3.02	3.41 ± 0.20	ND	2.39 ± 0.73	1402.92 ± 2.17
	The Village Press	Hawke's Bay	26/04	117.89 ± 0.45	23.65 ± 0.32	11.64 ± 2.55	1631.39 ± 15.27	51.38 ± 0.22	30.69 ± 0.33	4.14 ± 0.13	ND	3.04 ± 0.22	1873.80 ± 12.59
	Waimarama	Hawke's Bay	08/06	74.89 ± 1.74	34.92 ± 1.66	10.05 ± 1.43	1227.95 ± 8.04	17.94 ± 0.85	70.45 ± 2.29	4.68 ± 0.17	ND	3.79 ± 0.52	1444.68 ± 12.44
	Olivo	Wairarapa	24/05	110.91 ± 1.60	17.58 ± 0.66	11.05 ± 1.82	1486.93 ± 12.44	74.10 ± 0.95	44.68 ± 0.28	3.91 ± 0.33	0.10 ± 0.10	6.25 ± 0.39	1755.52 ± 16.68
'Picual'	Putiki	Waiheke	0905	25.32 ± 0.30	4.06 ± 0.07	5.36 ± 1.71	928.02 ± 4.06	28.78 ± 0.01	60.37 ± 3.06	4.64 ± 0.24	0.61 ± 0.32	15.29 ± 0.33	1072.45 ± 2.87
	Simunovich	Auckland	06/07	57.19 ± 1.27	10.36 ± 0.15	8.41 ± 1.86	1261.18 ± 1.74	37.86 ± 0.64	55.09 ± 0.47	2.71 ± 0.18	ND	3.14 ± 0.26	1435.94 ± 2.03
	The Village Press	Hawke's Bay	26/04	84.88 ± 0.12	28.84 ± 0.08	16.76 ± 1.81	1855.05 ± 10.19	106.49 ± 0.65	36.12 ± 0.03	4.44 ± 0.31	0.67 ± 0.22	7.62 ± 0.06	2140.88 ± 12.74
	Tasman Bay	Nelson	26/06	50.80 ± 0.87	13.72 ± 0.91	8.83 ± 0.34	1119.25 ± 4.11	39.98 ± 0.83	60.20 ± 2.61	3.67 ± 0.48	0.05 ± 0.05	4.38 ± 0.41	1300.90 ± 2.39
'J5'	Quail Ridge	Northland	29/03	34.66 ± 0.44	10.06 ± 0.09	6.40 ± 1.65	957.53 ± 3.17	37.20 ± 0.43	59.45 ± 0.42	6.11 ± 0.39	0.18 ± 0.02	13.22 ± 0.25	1124.81 ± 1.73
	Simunovich	Auckland	06/07	45.86 ± 0.83	8.42 ± 0.08	6.72 ± 1.54	1218.47 ± 30.36	23.66 ± 1.56	115.35 ± 3.72	5.12 ± 0.16	ND	3.06 ± 0.19	1426.64 ± 33.55

Table 11. Fatty acid composition of Accelerated Solvent Extraction (ASE)-extracted olive oils from samples harvested in 2010. Mean ± SEM (%w/w methyl esters).

				% w/w as methyl esters									
Cultivar	Grower	Region	Harvest Date	Palmitic acid C16:0	Palmitoleic acid C16:1	Heptadecanoic acid C17:0	Heptadecenoic acid C17:1	Stearic acid C18:0	Oleic acid C18:1	Linoleic acid C18:2	Linolenic acid C18:3	Arachidic acid C20:0	Gadoleic acid C20:1
'Frantoio'	Toru Waiwai	Northland	26/04	10.41±0.07	0.79±0.02	0.03±0.00	0.08±0.00	1.59±0.03	76.50±0.21	9.32±0.30	0.74±0.02	0.28±0.01	0.27±0.01
	River	Auckland	08/06	8.86±0.00	0.6 ± 0.01	0.02±0.00	0.07±0.00	1.58±0.00	79.90±0.05	7.61±0.03	0.78±0.02	0.29±0.00	0.27±0.00
	Armour	Auckland	25/05	9.39±0.02	0.61±0.00	0.03±0.00	0.08±0.01	1.57±0.00	77.92±0.05	8.91±0.03	0.88±0.01	0.31±0.01	0.30±0.00
	Simunovich	Auckland	22/06	8.20±0.01	0.02±0.00	0.02±0.00	0.07±0.00	0.89±0.00	81.06±0.02	8.16±0.00	1.09±0.00	0.14±0.01	0.28±0.00
	Matiatia	Waiheke	10/05	10.26±0.04	0.18±0.00	0.01±0.00	0.04±0.00	1.80±0.03	77.42±0.01	9.42±0.07	0.43±0.00	0.22±0.01	0.18±0.00
	Matapiro	Hawke's Bay	30/06	8.72±0.00	0.09±0.01	0.01±0.00	0.06±0.00	1.13±0.02	80.52±0.03	8.58±0.03	0.46±0.00	0.18±0.01	0.20±0.00
	The Village Press	Hawke's Bay	26/04	9.56±0.00	0.70±0.01	0.03±0.00	0.10±0.00	1.31±0.01	77.03±0.05	9.59±0.03	1.07±0.04	0.28±0.00	0.33±0.00
	Olea	Wairarapa	02/05	9.05±0.01	0.67±0.00	0.03±0.00	0.08±0.00	1.44±0.01	78.79±0.01	8.41±0.01	1.00±0.00	0.27±0.00	0.27±0.00
	Nelson Olives	Nelson	22/06	8.26±0.00	0.50±0.01	0.02±0.00	0.06±0.00	1.78±0.00	78.86±0.04	8.96±0.03	1.00±0.01	0.30±0.00	0.25±0.00
	Tasman Bay	Nelson	21/06	8.38±0.01	0.57±0.00	0.02±0.00	0.06±0.00	1.69±0.00	78.82±0.01	8.97±0.02	0.94±0.00	0.29±0.00	0.25±0.00
	Neudorf	Nelson	06/07	7.98±0.00	0.47±0.00	0.02±0.00	0.06±0.00	1.76±0.00	79.38±0.00	8.70±0.01	1.08±0.00	0.30±0.00	0.26±0.00
	Cairnmuir	Central Otago	22/06	8.30±0.00	0.52±0.01	0.02±0.00	0.06±0.00	1.64±0.01	78.96±0.06	8.99±0.04	0.98±0.02	0.28±0.00	0.24±0.00
	Poverty Gully	Central Otago	09/06	8.41±0.05	0.46±0.01	0.03±0.00	0.07±0.00	1.83±0.01	78.02±0.10	9.65±0.06	0.99±0.00	0.30±0.00	0.24±0.00
'Leccino'	Quail Ridge	Northland	30/03	11.98±0.00	0.92±0.00	0.04±0.00	0.05±0.00	1.85±0.02	77.06±0.01	6.86±0.02	0.6±0.00	0.29±0.00	0.34±0.01
	Toruwaiwai	Northland	26/04	10.94±0.03	0.98±0.02	0.02±0.00	0.08±0.02	1.59±0.01	77.37±0.01	7.72±0.04	0.83±0.05	0.24±0.01	0.23±0.01
	Matiatia	Waiheke	25/05	10.28±0.01	1.20±0.00	0.03±0.00	0.05±0.00	1.86±0.00	77.06±0.07	8.45±0.04	0.65±0.04	0.24±0.00	0.18±0.00
	Putiki Grove	Waiheke	09/05	11.27±0.11	1.13±0.04	0.03±0.00	0.04±0.00	2.00±0.02	72.10±0.25	12.36±0.15	0.68±0.00	0.21±0.01	0.18±0.03
	Armour	Auckland	25/05	9.69±0.03	0.87±0.01	0.02±0.00	0.07±0.00	1.44±0.01	78.75±0.02	7.82±0.01	0.85±0.01	0.24±0.00	0.25±0.00
	Matapiro	Hawke's Bay	21/06	9.85±0.03	0.92±0.02	0.02±0.00	0.06±0.00	1.67±0.01	77.20±0.06	8.82±0.01	0.99±0.00	0.27±0.00	0.21±0.00
	The Village Press		26/04	10.74±0.02	0.78±0.01	0.02±0.00	0.06±0.00	1.68±0.01	75.91±0.03	9.18±0.02	1.14±0.00	0.26±0.00	0.23±0.00
	Olea	Wairarapa	02/05	9.86±0.00	0.64±0.01	0.03±0.00	0.06±0.00	1.80±0.00	76.56±0.00	9.46±0.00	1.09±0.00	0.27±0.00	0.24±0.00
	Lusatori	Marlborough	08/06	9.84±0.00	0.64±0.01	0.02±0.00	0.06±0.00	1.81±0.00	76.27±0.00	9.69±0.01	1.16±0.00	0.28±0.00	0.22±0.00
	Nelson	Nelson	22/06	9.24±0.01	0.69±0.02	0.03±0.00	0.06±0.00	1.95±0.00	78.36±0.10	8.17±0.05	1.00±0.06	0.28±0.00	0.22±0.00
	Tasman Bay	Nelson	21/06	9.78±0.01	0.64±0.00	0.02±0.00	0.07±0.01	1.9±0.01	76.97±0.14	9.27±0.01	0.9±0.01	0.23±0.01	0.23±0.01
	Cairnmuir	Central Otago	22/06	9.37±0.01	0.61±0.00	0.03±0.00	0.08±0.00	1.73±0.00	77.10±0.02	9.56±0.01	1.04±0.01	0.26±0.00	0.21±0.00
	Poverty Gully	Otago	24/05	9.62±0.18	0.58±0.06	0.09±0.02	0.07±0.01	1.87±0.04	78.66±0.09	7.95±0.18	0.59±0.01	0.28±0.01	0.28±0.00

Cultivar	Grower	Region	Harvest Date	% w/w as methyl esters											
				Palmitic acid C16:0	Palmitic acid C16:0	Palmitic acid C16:0	Palmitic acid C16:0	Palmitic acid C16:0	Palmitic acid C16:0	Palmitic acid C16:0	Palmitic acid C16:0	Palmitic acid C16:0	Palmitic acid C16:0	Palmitic acid C16:0
'Koroneiki'	Quail Ridge	Northland	30/4	7.84±0.02	0.53±0.00	0.02±0.00	0.06±0.00	2.23±0.00	80.26±0.03	7.35±0.06	1.06±0.01	0.37±0.00	0.28±0.00
	Toruwaiwai	Northland	10/05	7.75±0.00	0.66±0.01	0.02±0.00	0.06±0.00	1.84±0.01	80.92±0.00	7.29±0.01	0.90±0.00	0.32±0.00	0.24±0.00
	River	Auckland	25/05	7.65±0.02	0.66±0.01	0.02±0.00	0.06±0.00	1.69±0.00	82.18±0.00	6.30±0.01	0.91±0.00	0.29±0.00	0.23±0.00
	Armour	Auckland	25/05	7.39±0.00	0.57±0.00	0.02±0.00	0.05±0.00	2.72±0.00	79.24±0.09	8.32±0.07	1.02±0.03	0.43±0.00	0.25±0.00
	Watchdog	Waiheke	9/05	8.33±0.02	0.65±0.01	0.01±0.00	0.04±0.00	2.34±0.01	78.64±0.05	8.57±0.05	0.83±0.00	0.36±0.00	0.22±0.00
	Stone	Wairarapa	22/06										
	Valley			7.16±0.01	0.51±0.01	0.03±0.01	0.04±0.00	1.90±0.01	82.40±0.01	6.52±0.01	0.90±0.00	0.31±0.00	0.23±0.00
	Lusatori	Marlborough	23/06	7.15±0.01	0.45±0.00	0.02±0.00	0.04±0.00	2.07±0.00	80.45±0.02	8.10±0.01	1.09±0.02	0.36±0.00	0.26±0.00
'Barnea'	Matapiro	Hawke's Bay	24/05										
				7.15±0.00	0.46±0.00	0.03±0.00	0.06±0.00	1.92±0.01	78.80±0.02	10.14±0.01	0.97±0.00	0.28±0.00	0.20±0.00
	The Village Press	Hawke's Bay	26/04										
				8.25±0.01	0.57±0.01	0.04±0.00	0.08±0.00	1.64±0.02	75.01±0.06	12.14±0.02	1.66±0.00	0.33±0.00	0.29±0.00
	Waimarama	Hawkes Bay	08/06	7.01±0.00	0.50±0.00	0.03±0.00	0.06±0.00	1.86±0.01	78.92±0.05	10.18±0.04	0.90±0.00	0.31±0.00	0.22±0.00
	Olivo	Wairarapa	24/05	7.00±0.02	0.41±0.02	0.03±0.00	0.04±0.00	2.56±0.01	77.74±0.05	10.63±0.04	0.96±0.01	0.37±0.00	0.25±0.00
'Picual'	Putiki	Waiheke	0905	8.97±0.00	0.79±0.10	0.02±0.00	0.04±0.00	2.86±0.01	79.26±0.02	6.97±0.02	0.64±0.01	0.28±0.00	0.16±0.00
	Simunovich	Auckland	06/07	7.05±0.10	0.49±0.01	0.03±0.00	0.09±0.00	1.69±0.01	83.66±0.15	5.68±0.21	0.76±0.04	0.27±0.00	0.28±0.00
	The Village Press	Hawke's Bay	26/04										
				8.85±0.00	0.75±0.00	0.02±0.00	0.08±0.00	1.34±0.00	80.40±0.01	6.87±0.00	1.18±0.01	0.24±0.00	0.26±0.00
	Olea	Wairarapa	2/05	9.04±0.00	0.60±0.01	0.03±0.00	0.06±0.00	1.99±0.00	78.82±0.04	7.87±0.05	1.07±0.00	0.30±0.00	0.23±0.00
	Tasman	Nelson	26/06										
	Bay			8.99±0.01	0.57±0.02	0.03±0.00	0.06±0.00	2.06±0.00	78.93±0.09	7.75±0.05	1.07±0.03	0.31±0.00	0.23±0.00
'J5'	Quail Ridge	Northland	29/03	11.04±0.00	0.92±0.00	0.02±0.00	0.06±0.00	1.76±0.01	75.54±0.05	9.34±0.07	0.84±0.03	0.27±0.00	0.21±0.00
	Simunovich	Auckland	06/07	7.58±0.01	0.49±0.02	0.03±0.00	0.09±0.00	1.18±0.01	80.28±0.16	8.73±0.12	0.95±0.07	0.28±0.00	0.39±0.00

6. General Discussion

Maturity of horticultural crops is a complex issue. Finding maturity indices (measurable characteristics) that can be carried out simply, repeatedly and cost effectively is challenging. It is also rare that only one measure is used to measure maturity. For example, apple maturity is measured using blush and background skin colour, starch pattern index (measure of starch degradation), internal ethylene concentrations, and soluble sugar content (SSC, measured as °Brix). Kiwifruit maturity is based on seed colour, SSC/°Brix and dry matter, and research is continuing to find other indices that could be useful.

The time of olive harvest varies with cultivar, region, local microclimate, risk of adverse weather conditions (e.g. frost), harvest technique (machine/mechanical v. hand), and the nature of the final oil product desired. The latter takes into account desired flavours (more or less pungent/fruity), healthfulness (sterol, tocopherol, or polyphenol content), storability, and whether the oils will be blended. For example, some portion of the crop could be harvest so as to be very pungent while the remainder is harvested later (to increase yield) then blended with the stronger oils.

Although the maturity index used overseas (external and internal colour) has some correlation with the total oil present in the fruit (e.g. Figure 4), it alone is clearly not enough to be used to make harvest decisions.

While attending the World Oil Congress in Australia, co-authors Woolf and Wong visited Modern Olives and discussed the criteria they use to commence commercial harvest. Their primary decision tool was that of accumulation of dry matter (loss of moisture), which correlated well with oil accumulation.

Ultimately, growers need decision support tools that can give rapid answers on a range of parameters (e.g. total oil, ease of oil extraction, polyphenols). The time to carry out these measurements, and the cost, are important considerations, particularly for small grove owners.

6.1. Year 3; 2010 season

The work carried out this season is the first extensive systematic study on the changes of olive fruit characteristics throughout New Zealand as they mature towards an optimum stage for oil extraction. It also provided an opportunity to measure important phytochemicals such as polyphenols, sterols and tocopherols in at least seven growing regions.

6.1.1   Oil accumulation

Significant regional differences for each cultivar were found:

   a. Oil accumulation and timing of when this occurred over the growing season with oil accumulating being 5 to 15% higher in the North Island than South Island.

   b. Timing of the maximum amount of oil accumulated, and whether there was a plateau, with strong differences as orchard location became further south. Some North Island orchards peaked as early as April, while South Island orchards generally peaked in June.

   c. Variation in the maximum oil yield in the North Island v. the South Island, and more variation within North Island regions. Overall, orchards south of Hawke's Bay responded very similarly.

6.1.2   Measuring maturity

a. The development of colour in the fruit was influenced by climatic conditions surrounding the fruit ripening stage. Thus, a maturity index based solely on this parameter is not recommended for the whole country. In addition, fruit colour development is cultivar dependent. It appears to be a better indicator of maturity for some cultivars such as 'Leccino' than for others, such as 'Frantoio'.

b. Measuring dry matter (or its inverse - the moisture content) and expressing it as a percentage (the standard approach used for avocado and other fruit), has proven not to be a reliable indicator of maturity for New Zealand, as it does not reflect changes in oil content. This is contrary to results from Australia, and is perhaps not surprising, considering the high rainfall that occurs in New Zealand during the harvest period.

c. Expressing dry matter on a per fruit basis (g; solids per fruit) has shown a close relationship to oil content throughout fruit maturity and in addition it can be practically measured by the growers (Section 5.6).

6.1.3   Polyphenols

a. Polyphenols decline with maturity and show a close inverse relationship to oil content (Figure 15).

b. To maximise polyphenol content in an oil (for maximising oil stability (longer shelf life) or increased flavour), one strategy is to harvest earlier. Alternatively, a portion of the crop might be harvested earlier (lower yield, but greater polyphenol content and less risk from frost), then a later harvest for the rest of crop. The oils could then be blended for bottling an oil to a target of flavour and shelf life.

c. A general observation was that most of the commercial harvests were carried out when the oil accumulation had reached a plateau. Thus, harvest could have been carried out a few weeks earlier and yet achieved the same yield, but with higher phenolic concentrations. However, other logistic issues such as weather, labour and ease of fruit removal (for mechanical harvest) must also be considered.

6.1.4   Composition

During oil extraction, as well as recovery of the lipid fraction (i.e. oil) containing the triglycerides, important minor components are extracted into the oil, including phenolics, tocopherols and sterols. Their concentration in the oils may be very low, but their presence imparts important sensory characteristics, enhances the oil's oxidative stability and increases the healthfulness of the oil.

Each triglyceride molecule contains three fatty acids and the overall fatty acid composition of olive oils is used as one measure to help with identification of adulteration. As the olive fruit matures, the oleic acid concentration increases and the palmitic and linolenic acid concentrations decrease (Gutie´rrez et al., 1999). The fatty acid profile of olive oil is distinctive, with a high monounsaturated content, 55-86.5% w/w (oleic and palmitoleic acids), moderate amounts of polyunsaturates 3.5-21% (linoleic and linolenic acids) and saturates 7.5-20% (palmitic and stearic acids). ASE-extracted olive oils from all regions in New Zealand were found to have fatty acid compositions within the expected composition of

olive oils. From this year's sampling, it was difficult to determine any differences in fatty acid composition due to variety.

There were definite regional differences in the palmitic acid (saturate) and linolenic acid (polyunsaturated) concentrations. Fruit grown in regions north of Auckland, including Waiheke, generally had higher amounts of palmitic acid and lower amounts of linolenic acid, this being mostly noticeable in the 'Frantoio', 'Leccino' and 'Picual' cultivars. Fruit will produce more unsaturated fatty acids in colder climates. Unsaturated fatty acids have lower melting points than saturated fatty acids; as the more southern regions are more prone to frosts, the fruit have adapted to this by producing more unsaturated fatty acids, which will not freeze and cause cell rupture (Harwood et al., 1994). It has been observed in other oleaginous fruit such as avocado that if grown in warmer climates, the fruit produce a higher percentage of saturated fatty acids.

In olive oil produced in colder climates in New Zealand, considerably colder than northern hemisphere olive-growing regions, there is a possibility that oil may be extracted with higher amounts of linolenic acid, which may be outside the IOC limit of $\leq 1.0\%$. The results collected this year show the ASE-extracted oils had more than 1% linolenic acid, but as mentioned earlier, ASE extraction can lead to greater extraction of linolenic acid (20-30% more), more than expected from cold-pressed extraction of EVOO (extra virgin olive oil). The sterol composition presented as a % of total sterols is used also as a method of detecting adulteration. If olive oils have been adulterated with other oils, then the % sterol composition will not agree with standards set by the IOC based on northern hemisphere-grown fruit. ASE-extraction recovers more sterols and it is not known if the sterols are recovered by ASE in the same proportions as they are by cold pressing.

The sterol composition can give an indication of the cultivars from which the oil has been extracted; for example, 'Barnea' was found to have a distinctive sterol composition. 'Barnea' fruit produced oils with campesterol amounts at > 5% for each cultivar and the absolute sterol concentration in the oils was between 1402 mg/kg and 1873 mg/kg. In comparison 'Koroneiki' had sterol concentrations of only 685-1114 mg/kg but the % of campesterol was always found to > 4%. The oils from 'Frantoio', 'Leccino', 'Picual' and 'J5' were not distinctly different in terms of % sterol composition or total sterol concentration (873-1547 mg/kg).The high % of campesterol (>4%) was also found in Australian-grown 'Barnea' and 'Koroneiki' by Mailer et al. (2010).

The oxidative stability of olive oils is influenced by the fatty acid composition and antioxidants present. Oils high in polyunsaturated fatty acids will oxidise faster than those with monounsaturated and saturated fatty acids. Antioxidants present in the oil include the phenolics and tocopherols. The small molecular weight antioxidants are considered to be the most powerful antioxidants present in olive oil but the other phenolics and tocopherols present still contribute to oxidative stability. As shown earlier, the total phenolic contents drop rapidly as the olive matures. As shown in Figure 11, the total phenolic content in 'Frantoio' grown in the North Island dropped to below 1500 mg/kg in May, whereas in the South Island the total phenolics did not drop to the same level until June. For all the cultivars examined in this study, the total phenolic concentration in the oils from fruit harvested close to commercial harvest times were all below 1500 mg/kg (Figures 6 to 9, 11 and 12). Some cultivars did not show a further drop in total phenolics, with the concentration staying constant as oil accumulation had also levelled off, while fruit from some orchards showed a further drop in total phenolics, probably because of rainfall at this time (Section 5.4). Thus, a possible explanation for changes in polyphenol concentrations is that the amounts are defined soon after fruit set (i.e. once fruit cell number has been set), and then

the decline is due to dilution as fruit (and their cells) swell with increased water. This pattern and probable dilution mechanism has been observed in sweet persimmons ('Fuyu').

Most cultivars had similar amounts of phenolics at commercial harvest time, but they had varying quantities of total tocopherols. No regional differences were observed with tocopherol content. 'Frantoio' and 'J5' had the lowest concentrations of tocopherols, 155-290 mg/kg. The other cultivars, 'Leccino', 'Koroneiki', 'Picual' and 'Barnea', all had between 306 and 758 mg/kg, with most between 306 and 515 mg/kg. 'Barnea' from The Village Press had 758 mg/kg total tocopherol content, which was unusually high. Alpha-tocopherol was present in the highest concentration, being > 90% of all tocopherols present, followed by gamma- and beta tocopherols. Delta-tocopherol was only detected in some oils. 'Leccino', 'Picual' and 'Barnea' all produced oils with higher percentages of gamma-tocopherol (3.35-20%) relative to the total, higher than 'Frantoio' and 'Koroneiki' (1.67-3.84%). Various cultivars can be distinguished by their tocopherol composition.

The amount of tocopherols present in olive oils in 2010 was consistent with results obtained in the first and second season of the SFF project (Requejo-Jackman et al., 2009). Concentrations again were higher than reported in some northern hemisphere-produced olive oil (Manzi et al., 1998, Psomiadou et al., 2000).

6.2. All years' overview

Summaries of the research for years 1 and 2 can be found at the end of the Appendix. Key findings over the three-year SFF project have been:

6.2.1 Measures of maturity

- As fruit mature, there is a progressive accumulation of oil, with a levelling off later in the season in most cultivars and growing locations. There was significant variation between olive cultivars, growing regions, and even growers within a region. In general, orchards in the northern regions (Northland and Auckland) developed fruit with higher oil content and at an earlier time than the southern regions, but the fruit were more variable.

- The colour maturity index (as used overseas) was variable for both North and South Island orchards, and patterns of colour development (used for calculating the index) usually did not relate well to the changes in oil content accumulation (particularly for 'Frantoio').

- The dry matter, measured as a percentage (as is carried out in routinely in the avocado and kiwifruit industries) didn't relate well with (%) oil content. However, expressing dry matter on a per fruit basis (which we called "total solids" to avoid confusion), and expressed in grams, correlated very well with total oil content (r^2 of 0.7 to 0.8).

- In general, fruit weight tended to be largest in the northern regions and decreased the further south the orchards were located.

- The differences in climate among the olive-growing areas studied are reflected mainly in their regionally recorded growing degree days (GDD), sunshine and rainfall figures. These differences mean that fruit maturity was achieved at different times during the season, depending on the orchard location, but normally earliest in the northern regions and latest in the South Island regions.

- It should be noted that the level of oil accumulation measured for each cultivar was the total available oil (i.e. extracted by solvent), which will be higher that can be recovered by cold pressed extraction techniques. But it still shows when the fruit has achieved maximum oil accumulation. There are a number of factors that impact on oil yield from cold pressing (e.g. extraction system used, moisture content of fruit, pressure on processer to process more fruit) and the actual commercial yield over the varying maturities will differ from that obtained here with solvent extraction.

6.2.2   Oil composition overview

Because of the excellent grower response to our call to provide samples, we took the opportunity to carry measure oil composition across this wide range of regions and cultivars.

Higher linolenic acid percentages can potentially be found in New Zealand, especially in olives harvested in southern regions, but high amounts were also found in Auckland in Year 1. The higher amounts found in southern regions were due to the colder climate in these regions than in northern regions.

Certain cultivars grown in New Zealand ('Barnea', 'Koroneiki') produce oils with sterols compositions that will fall outside the current IOC standards, but will fall within the new Australia/New Zealand standards proposed. This is mainly because of the higher % of campesterol found in these cultivars. Concentrations that fall outside the IOC standards are not unique to New Zealand, but have been found for the same cultivars in Australia, Argentina and Greece (Mailer 2010; Mailer 2009).

Tocopherol concentrations extracted from New Zealand olive oils are higher than reported from northern hemisphere orchards. This was found for both cold-pressed oils and ASE-extracted oils. The reason for higher amounts of tocopherols in New Zealand oils has not been determined, but could be related to sun intensity and/or UV radiation, since some plants produce tocopherols in response to light stress. The tocopherol composition varied between cultivars, with 'Frantoio' having the lowest amounts and 'Leccino', 'Koroneiki', 'Picual' and 'Barnea' having nearly double the total tocopherol concentration of 'Frantoio'.

Phenolic concentration decreased as the olive fruit matured. This could be due to a dilution effect as the fruit volume increased. The amounts of phenolics were found to level off when oil accumulation also slowed down. But if the fruit absorbed more water because of heavy rainfall, the phenolic concentration would decline, indicating a concentration dilution effect in the fruit. Fruit harvested early in this plateau period of oil accumulation will be more likely to have higher amounts of phenolics extracted into the oil.

Harvesting olives before the phenolic concentration drops too low, before any significant rainfall, will result in an oil with good oxidative stability. All the oils analysed over the last three years had greater than 75% oleic acid present, which is more stable than the polyunsaturated fatty acids. The oils extracted in New Zealand also have high concentrations of tocopherols, which can act as antioxidants. The high monounsaturated consents and high antioxidant capacity of the oil contribute to the healthiness of the olive oils.

In Year 1, oils were only extracted as cold-pressed oils, producing EVOO. In Year 2 and 3, because we harvested fruit very early in the season, cold-pressed extraction of the oil from these fruit was not practical and so ASE extraction was used to recover the oil. In year 2, cold-pressed extraction of the oils was carried out later in the season, when the oil content was higher in the fruit and it became practical to carry out cold-pressed extraction. In year 3,

relatively small samples of fruit were sent by each grower (since nearly 10 kg of fruit is needed to carry out cold-pressed extraction). This meant ASE extraction was the most practical means of obtaining an oil sample for analysis (particularly since samples were harvested from early in the season).

ASE extraction uses hexane (non-polar solvent) to extract the oil from the ground olive tissue. ASE extraction gives a measure of the total oil present in the flesh, which is approximately 40% more than can be extracted by cold-pressed extraction. One would have expected the proportions of each fatty acid to be recovered by the two methods to be the same, but this was not found in Year 2 data. Linolenic acid was found to increase in % after ASE extraction compared with cold pressed extraction. Many of the minor components in the oil, tocopherols and sterols, are more soluble in the solvent hexane, and hence their concentrations in the ASE oil have been found to be 20-30% higher than in cold-pressed oils. Correlation of our collected ASE and cold-pressed data will help in the future to allow comparison of results from oils extracted by different methods.

7. Future research

The large study carried out in Year 3 of this project provides a solid platform of information that would be significantly improved by obtaining at least one more season of data. Significant seasonal differences occur in all fruit crops, and another season's data will improve the correlations and add robustness to the data.

Maturity indices. From research in 2009, measurement of fruit firmness shows potential to be a rapid tool that correlates well with maturity changes in oil content. There are also hand-held systems for measuring fruit firmness, which might be adapted for use on olives. A small project could be carried out that uses a range of measurement techniques over the months prior to harvest for a limited number of cultivars.

Orchard differences. One of the interesting observations in this study has been the very large differences in patterns of maturity between orchards that are relatively close geographically. Good examples of this are Simunovich and River, which are only a few kilometres apart yet, for example, the pattern of oil accumulation for Simunovich is more similar to that of Matapiro (in Hawke's Bay; Figure 4). Whether these differences are due to orchard practices alone, or to microclimate effects, is not known and it would be helpful to understand if orchard practices, such as irrigation, influence harvest timing.

Near Infrared (NIR). There are a wide range of commercial machines available that can rapidly measure samples of olive paste for a number of parameters. These machines, although expensive, are being used widely in many commercial oil businesses, and can measure a wide range of useful quality parameters including oil content, free fatty acids (FFA), peroxide value (PV), moisture content and fatty acid composition. The fact that these measures can be carried out in minutes is an additional benefit. Development of this technology in New Zealand would have a wide range of commercial benefits, and is used routinely in the Australian olive industry.

Ease of harvest. As discussed above, there are a wide range of factors which affect the choice of harvest time. When fruit are harvested by a tree shaker machine, the ease by which fruit can be removed from the tree is a factor which, if known, would add an important decision support tool to harvesting. This can be measured by attaching a force gauge to fruit and measuring the

force required to remove the fruit. This can be described as fruit removal force (FRF). Information on the change in FRF during maturity and how it correlates to oil content will be valuable information for growers and harvesters.

Since the flavour of the oils is a key characteristic, an understanding of the **relationship between maturity characteristics** of olives (especially the phenolic compounds) and their relationship to the compounds responsible for the aroma and flavour, such as n-hexanal ("grassy") or *cis*-2-penten-1-ol ("fruity"), would be valuable. Such a measure could be used such that the optimal harvest point could be based on oil content, phenolic content and "grassy" and "fruity" aroma and flavour. Robust information could provide tools to enable oils to be more consistent from season to season.

Conclusions

This three-year study has provided an extensive information baseline on New Zealand olives in terms of changes in maturity and the chemical makeup of the oils. The range of techniques examined for measuring maturity led to the recommendation that two are most appropriate for use by growers and processors:

- total solids per fruit (g)

- fruit firmness.

The final year of the work sourced olives from a wider range of cultivars, at different maturities and from all significant growing areas in both the North and South Islands. The information obtained is very useful for a number of applications, including development of quality standards, access to markets, and to maximise yield, quality, stability and flavour of oils. This research also highlighted important differences of New Zealand's growing conditions from those of Australia and northern hemisphere growing regions for olives. Hence, recommendations from growing regions other than New Zealand need to be validated under New Zealand's unique growing conditions.

References

Ainsworth, E. A. & Gillespie, K. M. (2007) Estimation of total phenolic content and other oxidation substrates in plant tissues using Folin-Ciocalteu reagent. *Nature Protocols,* 2, 875-877.

Ayton, J., Mailer, R.J., Haigh, A., Tronson, D. & Conlan, D. (2007). Quality and oxidative stability of australian olive oil according to harvest date and irrigation. *Journal of Food Lipids,* 14, 138-156.

Baccouri, O., Guerfel, M., Baccouri, B., Cerretani, L., Bendini, A., Lercker, G., Zarrouk, M. & Daoud Ben Miled, D. (2008). Chemical composition and oxidative stability of Tunisian monovarietal virgin olive oils with regard to fruit ripening. *Food Chemistry,* 109, 743-754.

Beltrán, G., Ruano, M.T., Jiménez, A., Uceda, M. & Aguilera, M.P. (2007). Evaluation of virgin olive oil bitterness by total phenol content analysis. *European Journal of Lipid Science and Technology,* 109, 193-197.

Ben Temime, S., Baccouri, B., Taamalli, W., Abaza, L., Daoud, D. & Zarrouk, M. (2006). Location effects on oxidative stability of Chétoui virgin olive oil. *Journal of Food Biochemistry,* 30, 659-670.

Blekas, G., Vassilakis, C., Harizanis, C., Tsimidou, M. & Boskou, D.G. (2002). Biophenols in table olives. *Journal of Agricultural and Food Chemistry,* 50, 3688-3692.

Boskou, D. (2006). Characteristics of the Olive Tree and Olive Fruit. In Boskou, D. (Ed.), *Olive Oil Chemistry and Technology*, (pp. 13-17). AOCS Press Champaign, IL USA.

Boskou, D., Tsimidou, M. & Blekas, G. (2006). Polar Phenolic Compounds. In Boskou, D. (Ed.), *Olive Oil Chemistry and Technology*, Vol. 2, (pp. 73-86). AOCS Press Champaign, IL USA.

Cinquanta, L., Esti, M. & Di Matteo, M. (2001). Oxidative stability of virgin olive oils. *JAOCS, Journal of the American Oil Chemists' Society,* 78, 1197-1202.

Dutta, P. C., & Normen, L. (1997). Capillary column gas-liquid chromatographic separation of Delta 5-unsaturated and saturated phytosterols. In *2nd Workshop COST916 - Bioactive Inositol Phosphates and Phytosterols in Foods,* (pp. 177-184). Gothenburg, Sweden.

Gawel, R. & Rogers, D.A.G. (2009). The relationship between total phenol concentration and the perceived style of extra virgin olive oil. *Grasas y Aceites,* 60, 134-138.

Gutiérrez, F., Arnaud, T. & Garrido, A. (2001). Contribution of polyphenols to the oxidative stability of virgin olive oil. *Journal of the Science of Food and Agriculture,* 81, 1463-1470.

Gutiérrez, F., Jímenez, B., Ruíz, A. & Albi, M. A. (1999) Effect of olive ripeness on the oxidative stability of virgin olive oil extracted from the varieties picual and hojiblanca and on the different components involved. *Journal of Agricultural and Food Chemistry,* 47, 121-127.

Hartman, L., & Lago, R. C. A. (1973). Rapid determination of fatty acid methyl esters from lipids. *Laboratory Practice, 22,* 475-476, 494.

Harwood, J. L., Jones, A. L., Perry, H. J., Rutter, A. J., Smith, K. L. & Williams, M. (1994). Changes in plant lipids during temperature adaptation. In Crossings, A. R. (Ed.) T*emperature adaption of biological membranes*. Portland Press: London, U.K.

IOC (2010). Trade standard applying to olive oils and olive-pomace oils. COI/T.15/NC no. 3/Rev5. I. O. Council. Madrid, International Olive Council: 19p.

Indyk, H.E. (1988). Simplified saponification procedure for the routine determination of vitamin E in dairy products, foods and tissues by high-performance liquid chromatography. *Analyst,* 113, 1217-1221.

Kiralan, M., Bayrak, A. & Özkaya, M.T. (2008). Oxidation stability of virgin olive oils from some important cultivars in East mediterranean area in Turkey. *JAOCS, Journal of the American Oil Chemists' Society*, 1-6.

Kiritsakis, A.K. (1998). Harvest and storage of olive fruit. In, *Olive oil from the tree to the table*, (pp. 41-52). Food and Nutrition Press, INC. Trumbull, Connecticut 06611 USA.

Skevin, D., Rade, D., Strucelj, D., Mokrovcak, Z., Nederal, S. & Bencic, D. (2003). The influence of variety and harvest time on the bitterness and phenolic compounds of olive oil. *European Journal of Lipid Science and Technology,* 105, 536-541.

Tsimidou, M., Papadopoulos, G. & Boskou, D. (1992). Phenolic compounds and stability of virgin olive oil - Part I. *Food Chemistry,* 45, 141-144.

Li, T. S. C., Beveridge, T. H. J., & Drover, J. C. G. (2007). Phytosterol content of sea buckthorn (Hippophae rhamnoides L.) seed oil: Extraction and identification. *Food Chemistry, 101*(4), 1633-1639.

Mailer, R. J., Ayton, J., & Graham, K. (2010). The Influence of Growing Region, Cultivar and Harvest Timing on the Diversity of Australian Olive Oil. *Journal of the American Oil Chemists Society, 87*(8), 877-884.

Manzi, P., Panfili, G., Esti, M., & Pizzoferrato, L. (1998). Natural antioxidants in the unsaponifiable fraction of virgin olive oils from different cultivars. *Journal of the Science of Food and Agriculture, 77*(1), 115-120.

Mateos, R., Cert, A., Carmen Pérez-Camino, M. & García, J.M. (2004). Evaluation of virgin olive oil bitterness by quantification of secoiridoid derivatives. *JAOCS, Journal of the American Oil Chemists' Society ,* 81, 71-75.

Orlandi, F.,& Bonofiglio, T. (2009). Olive flowering monitored in a large area of Italy and local climatic trends. **817:** 161-168.

Orlandi, F., & Ruga L., (2005). Olive flowering as an indicator of local climatic changes. Theoretical and Applied Climatology **81**(3-4): 169-176.

Psomiadou, E., Tsimidou, M., & Boskou, D. (2000). alpha-Tocopherol content of Greek virgin olive oils. *Journal of Agricultural and Food Chemistry, 48*(5), 1770-1775.

Requejo-Jackman, C., Farrell, M., Ogwaro, J., Olsson, S., Beresford, M., Harker, R., McGhie, T., Wang, Y., Wong, M., and Woolf, A. (2010). Effects of Harvest Maturity on Extra Virgin Olive Oil Year 2 – SFF Project #07/1.

Requejo-Jackman C., Farrell M., Olsson S., Beresford M., Harker, R., Wohlers, M., McGhie, T., Comeskey, D., Wang, Y., Woolf, A., and Wong, M. (2009). Effect of Harvest Maturity on Extra Virgin Olive Oil Year 1– SFF Project 07/103

Romero, M.P., Tovar, M.J., Ramo, T. & Motilva, M.J. (2003). Effect of crop season on the composition of virgin olive oil with protected designation of origin "Les Garrigues". *JAOCS, Journal of the American Oil Chemists' Society,* 80, 423-430.

Singleton, V.L., Orthofer, R. & Lamuela-Raventós, R. M. (1998). Analysis of total phenols and other oxidation substrates and antioxidants by means of folin-ciocalteu reagent. *Methods in Enzymology.*

Thompson, R.H., & Merola, G.V. (1993). A Simplified Alternative to the *AOAC* Official Method for Cholesterol in Multicomponent Foods. *Journal of AOAC International, 76*(5), 1057-1068.

Vinson, J.A., Proch, J. & Bose, P. (2001). Determination of quantity and quality of polyphenol antioxidants in foods and beverages, *Methods in Enzymology* (Vol. 335, pp. 103-114).
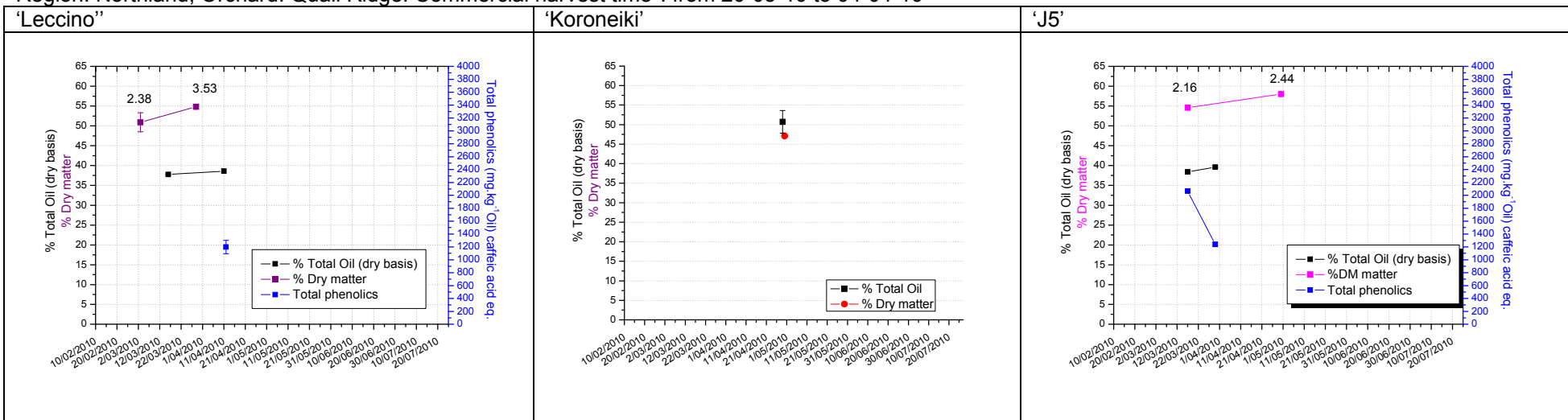
8. Appendix – Individual orchard graphs: %Total Oil, % Dry matter and Total phenolics

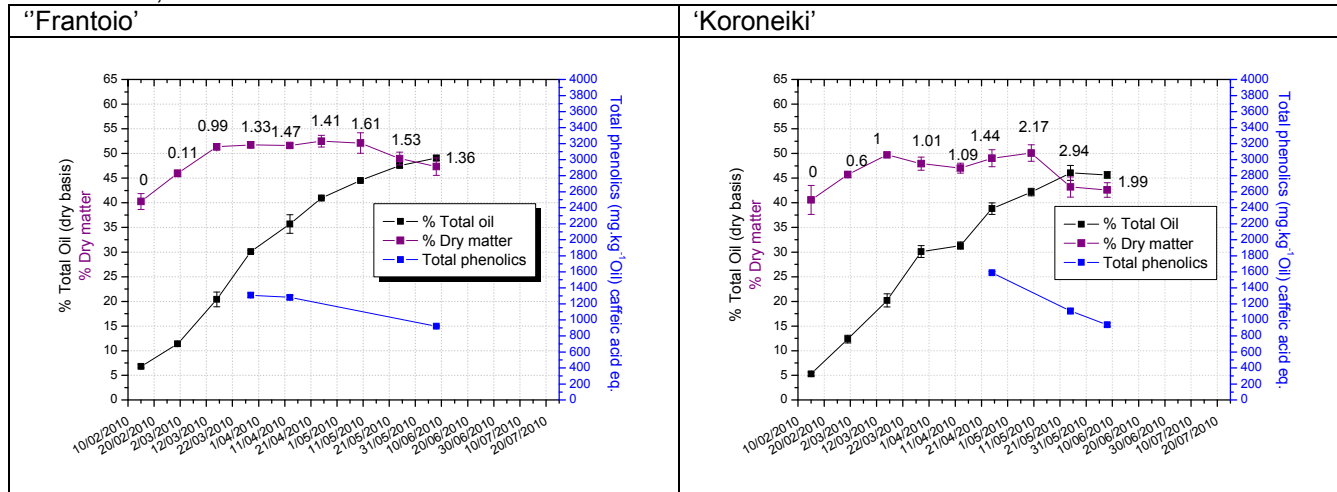Region: Northland, Orchard: Toru Waiwai. Commercial harvest time*: 25-04-10

'Frantoio'	'Leccino'	'Koroneiki'
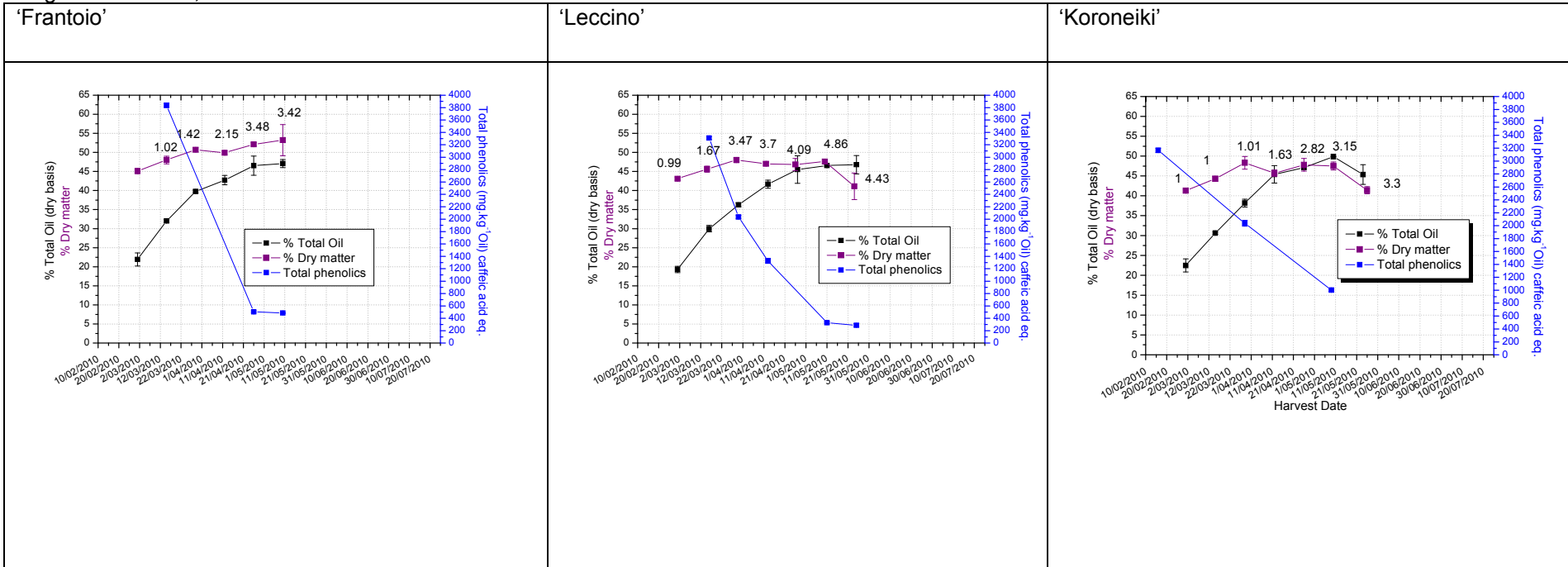
*As advised by grower, or estimated from communications with grower

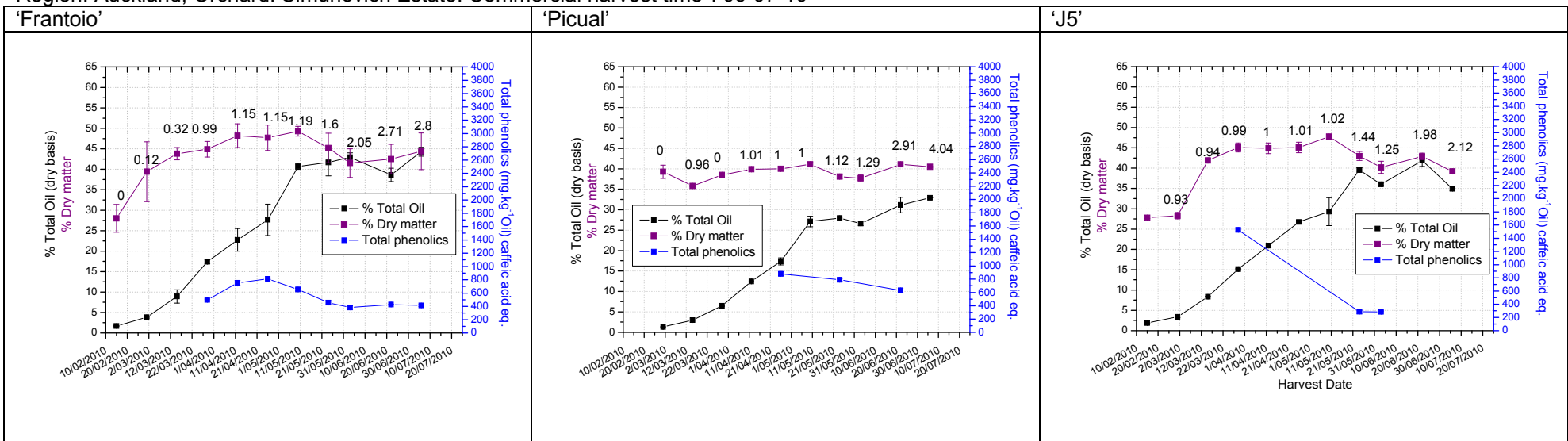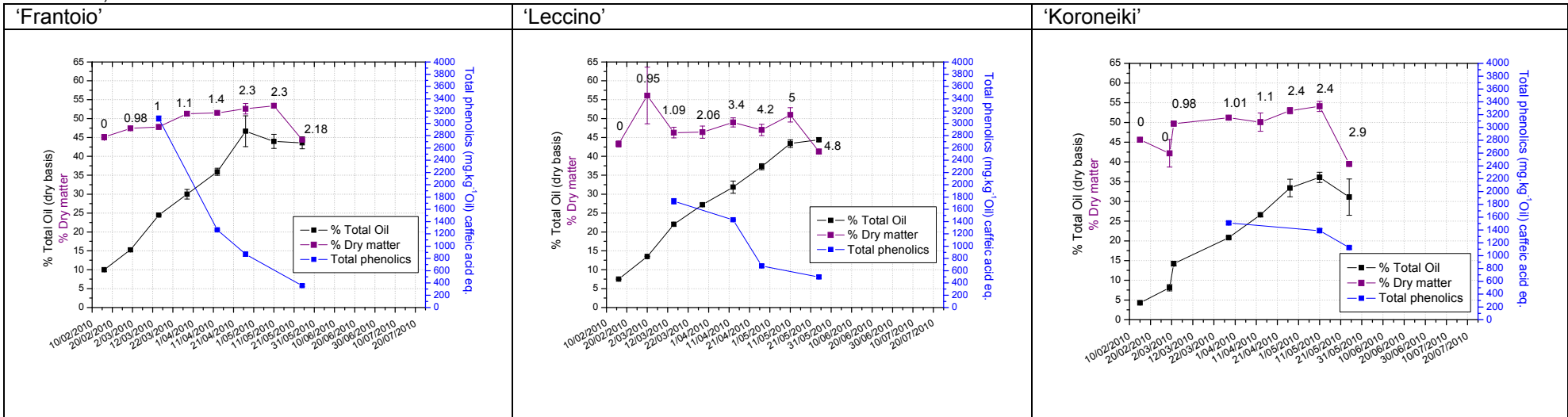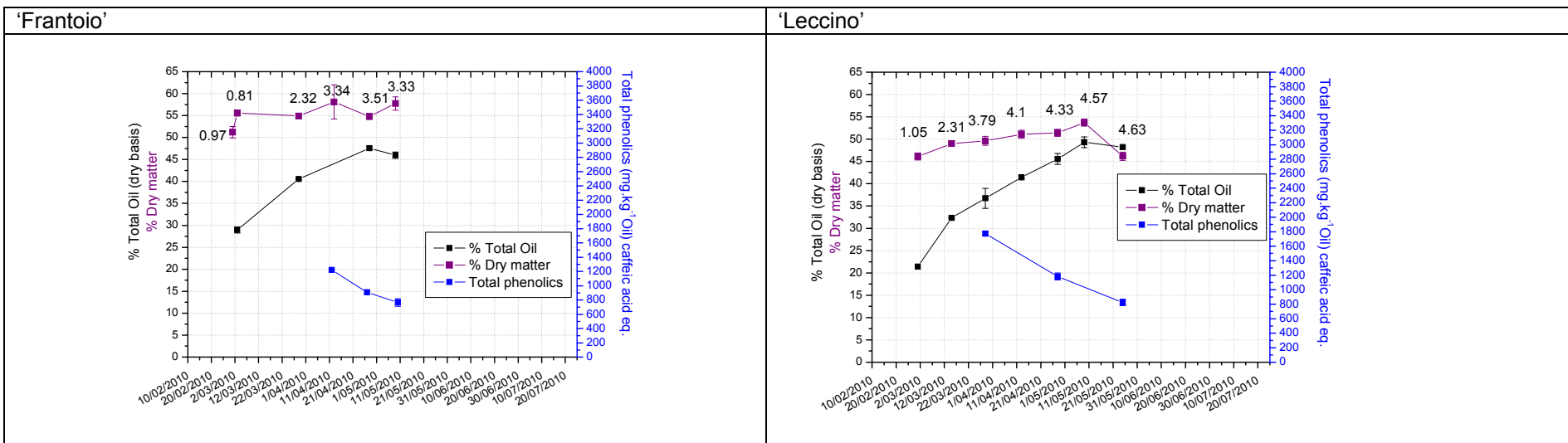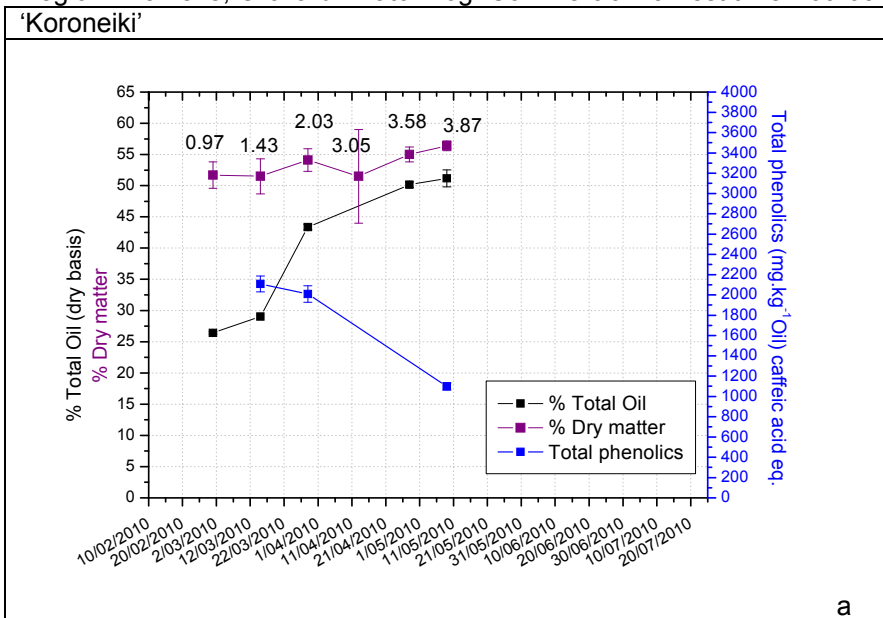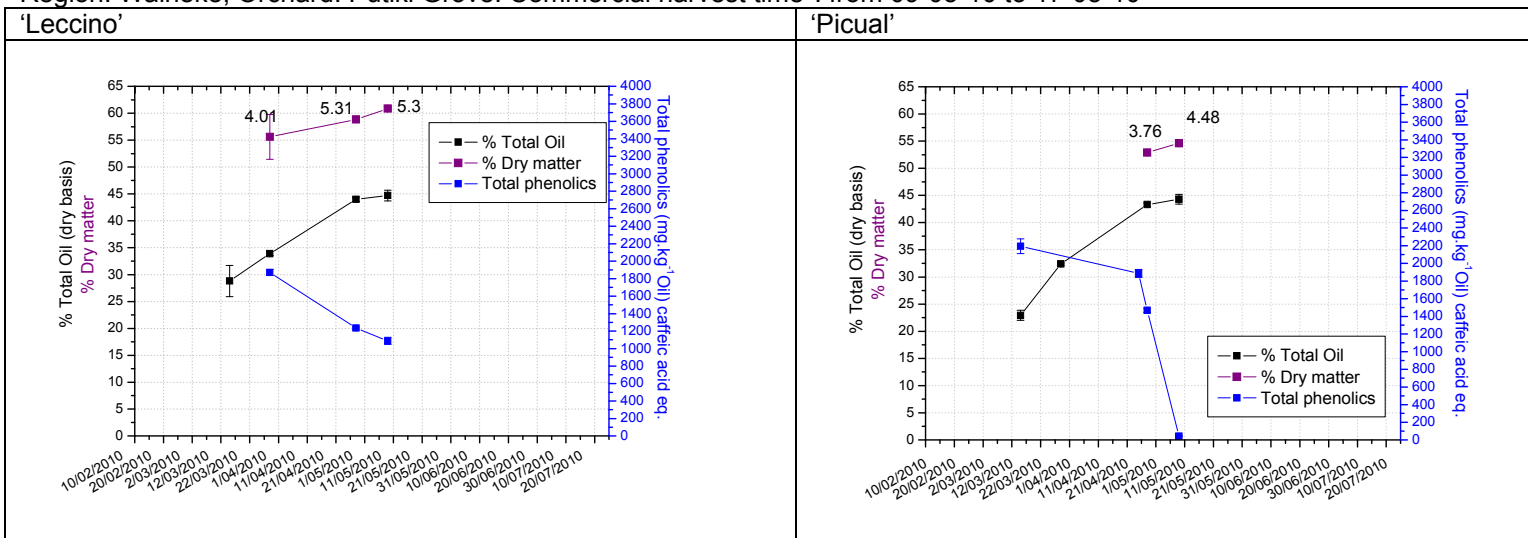Region: Northland, Orchard: Quail Ridge. Commercial harvest time*: from 20-03-10 to 04-04-10

'Leccino''	'Koroneiki'	'J5'

*As advised by grower, or estimated from communications with grower

Region: Auckland, Orchard: River Estate. Commercial harvest time*: 24-05-10 to 01-06-10

''Frantoio'	'Koroneiki'

*As advised by grower, or estimated from communications with grower

Region: Auckland, Orchard: Simunovich Estate. Commercial harvest time*: 06-07-10

'Frantoio'	'Picual'	'J5'

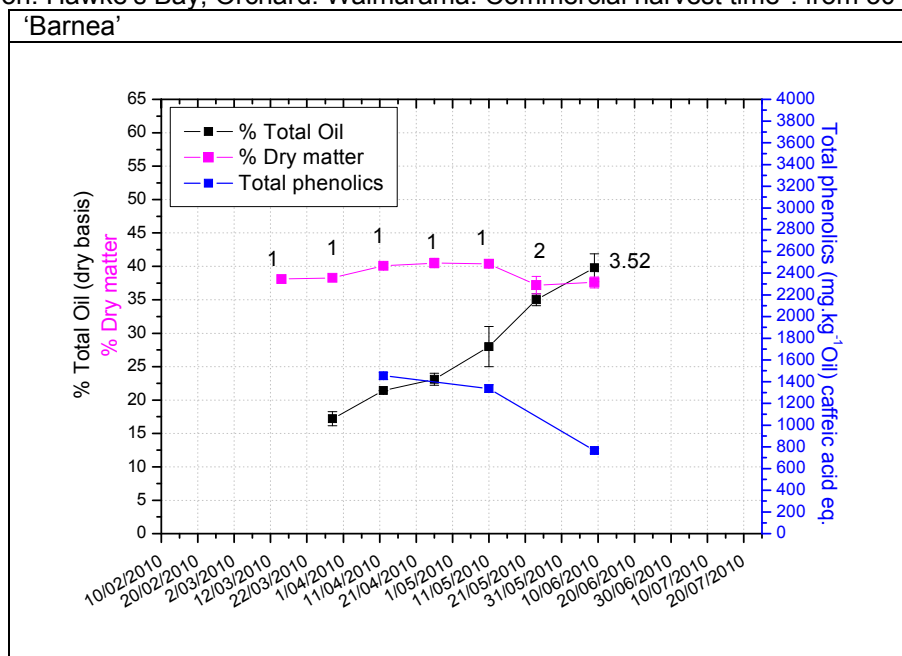*As advised by grower, or estimated from communications with grower

Auckland, Grower: Armour M. Commercial harvest time*: 25-05-10

'Frantoio'	'Leccino'	'Koroneiki'

*As advised by grower, or estimated from communications with grower

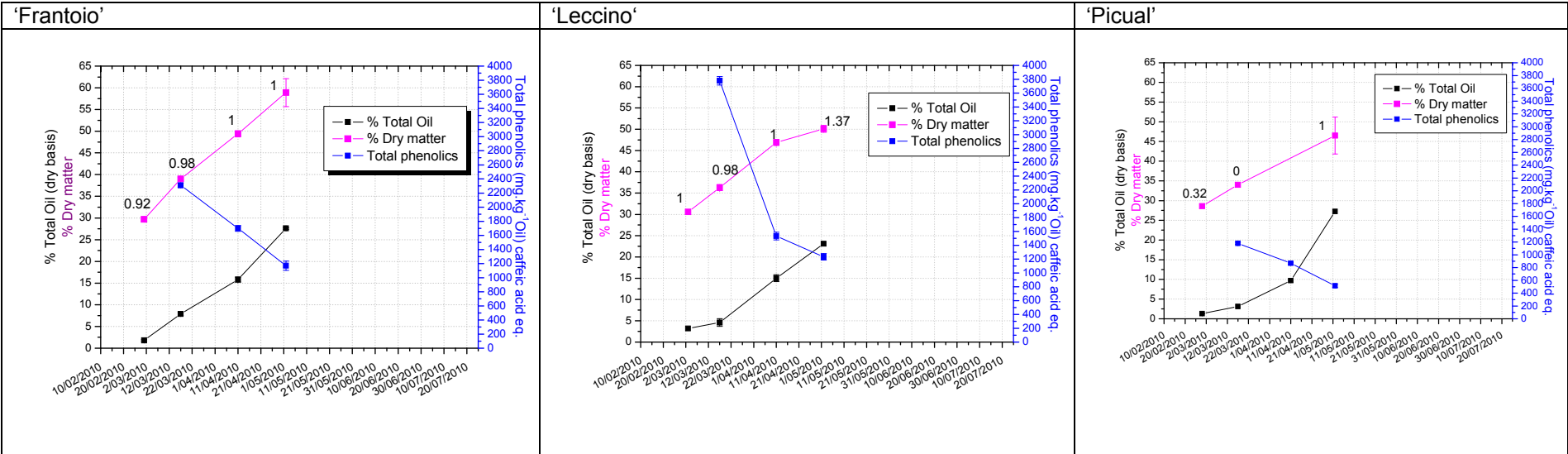Region: Waiheke, Orchard: Matiatia. Commercial harvest time*: from 17- 05-10 to 25-05-10

'Frantoio'	'Leccino'

*As advised by grower, or estimated from communications with grower

Region: Waiheke, Orchard: WatchDog. Commercial harvest time*: 09-05-10 to 17-05-10

'Koroneiki'

a

*As advised by grower, or estimated from communications with grower

Region: Waiheke, Orchard: Putiki Grove. Commercial harvest time*: from 09-05-10 to 17-05-10

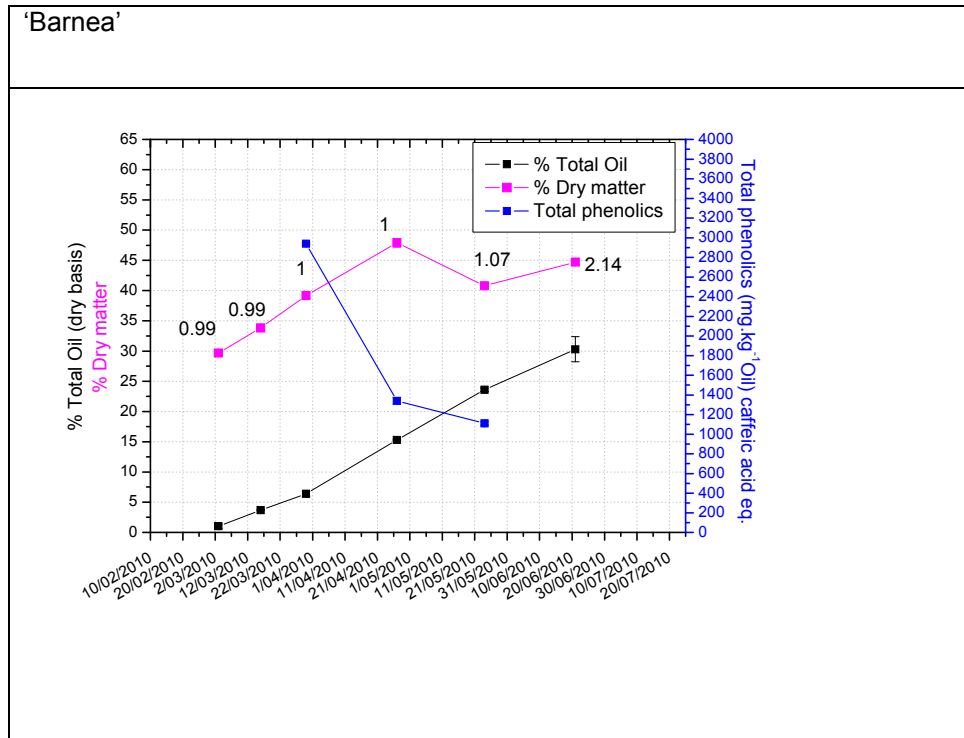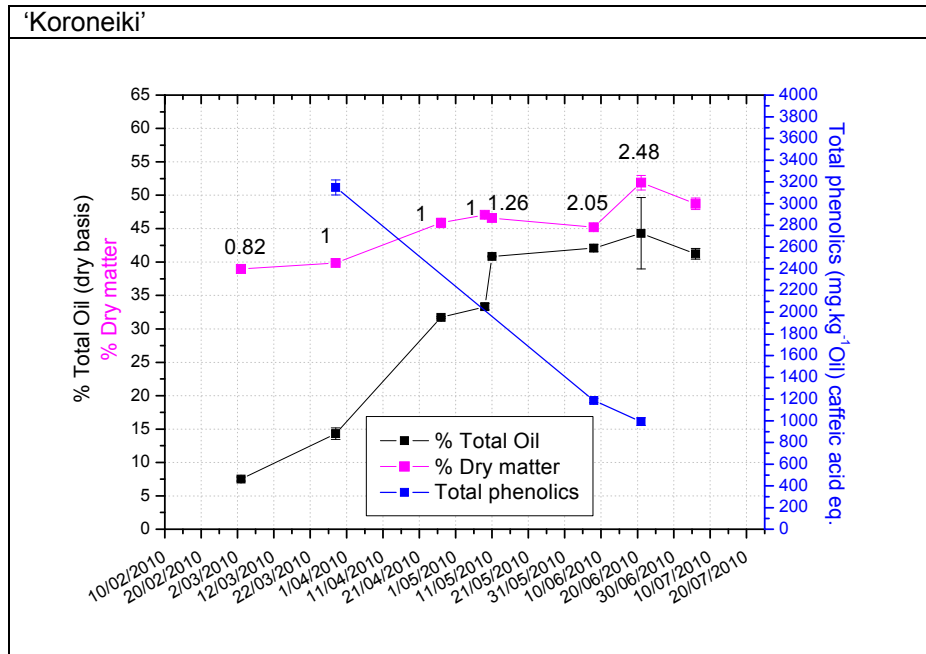| 'Leccino'	'Picual'

*As advised by grower, or estimated from communications with grower

Region: Hawke's Bay, Orchard: The Village Press. Commercial harvest time*: from 26-04-10
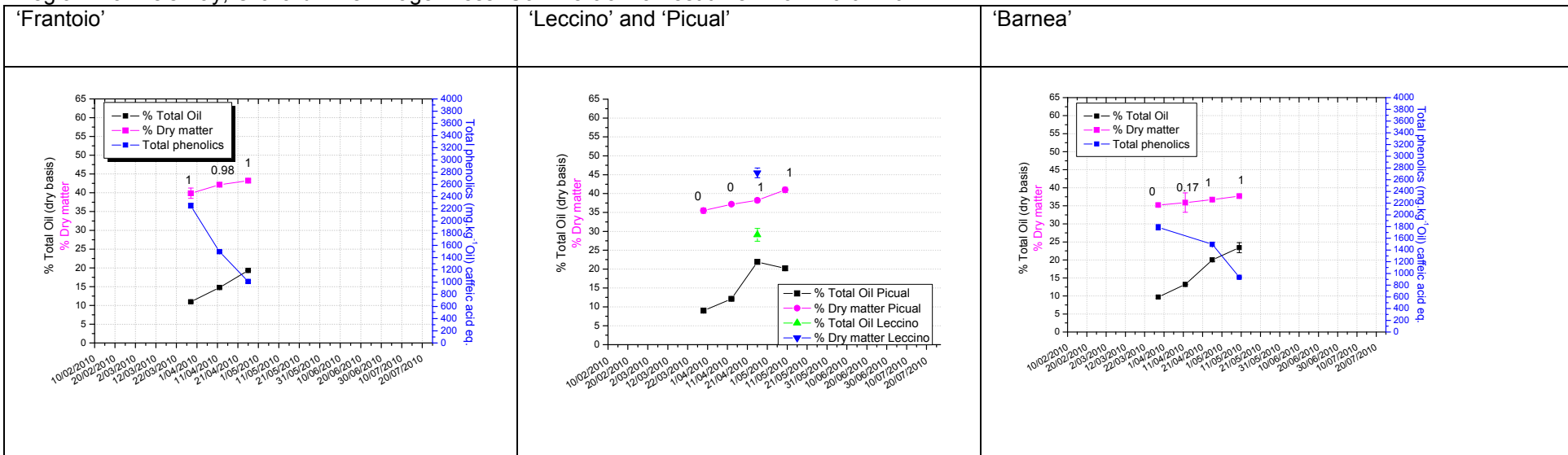
'Frantoio'	'Leccino' and 'Picual'	'Barnea'

*As advised by grower, or estimated from communications with grower

Region: Hawke's Bay, Orchard: Matapiro. Commercial harvest time*: from 21-06-10
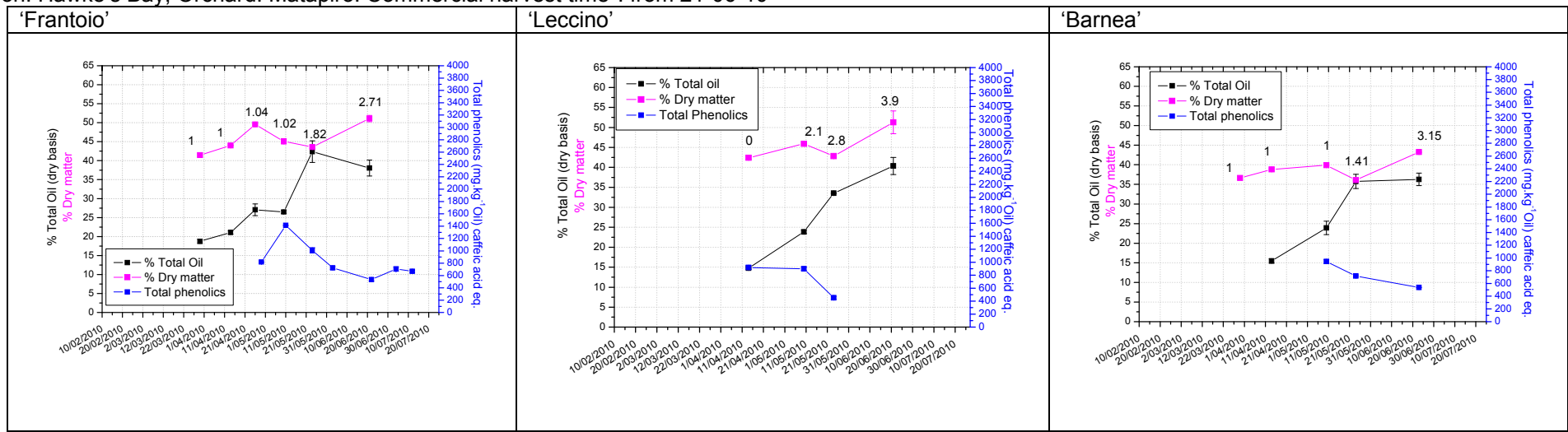
'Frantoio'	'Leccino'	'Barnea'

*As advised by grower, or estimated from communications with grower

Region: Hawke's Bay, Orchard: Waimarama. Commercial harvest time*: from 30-05-10 to 03- 06-10

'Barnea'

*As advised by grower, or estimated from communications with grower

Region: Wairarapa, Orchard: Olea. Commercial harvest time*: Early- Mid June

'Frantoio'	'Leccino'	'Picual'

*As advised by grower, or estimated from communications with grower

Region: Wairarapa, Orchard: Olivo. Commercial harvest time*: 21-06-10

'Barnea'

*As advised by grower, or estimated from communications with grower

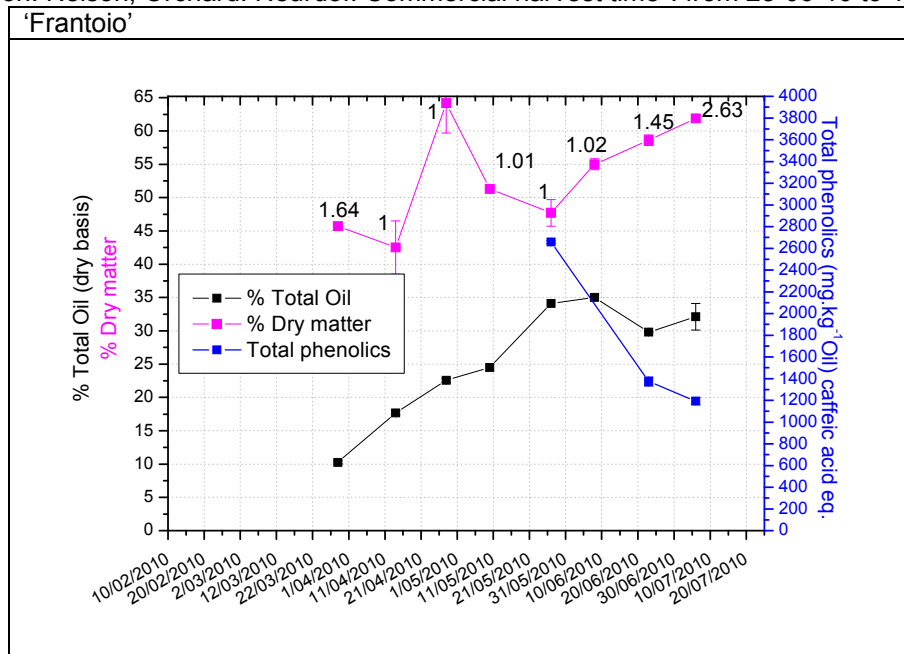Region: Wairarapa, Orchard: Stone Valley. Commercial harvest time*: 25-06-10

'Koroneiki'

*As advised by grower, or estimated from communications with grower

Region: Nelson, Orchard: Nelson Olives. Commercial harvest time*: from 22-06-10
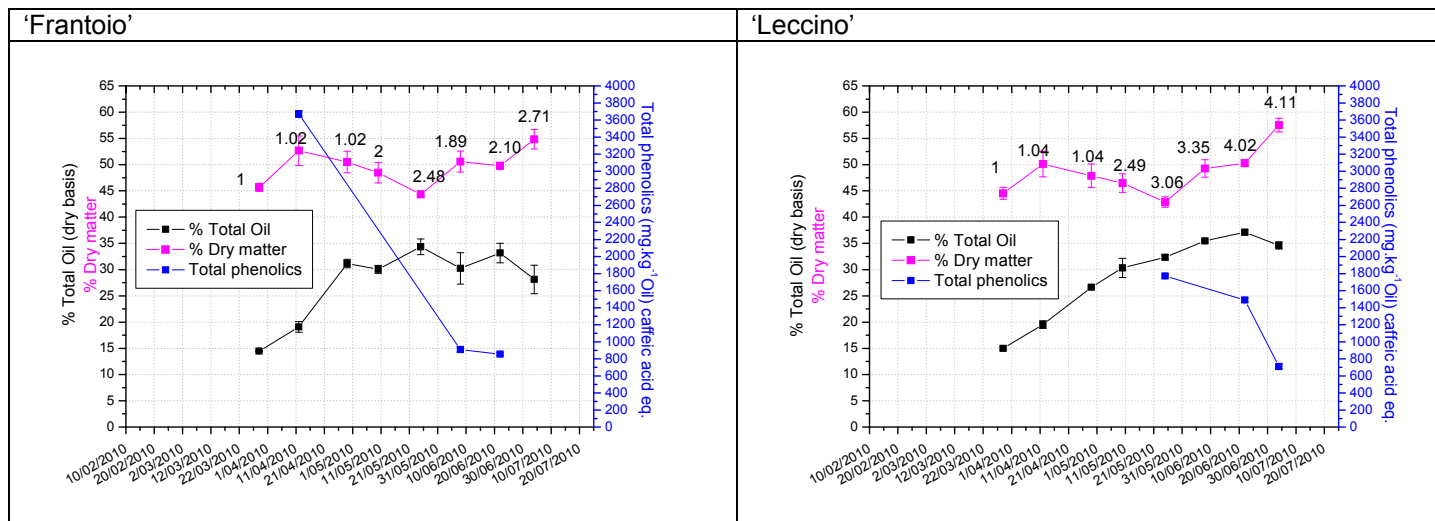
'Frantoio'

'Leccino'

*As advised by grower, or estimated from communications with grower

Region: Nelson, Orchard: Neurdof. Commercial harvest time*: from 28-06-10 to 19-07-10

'Frantoio'

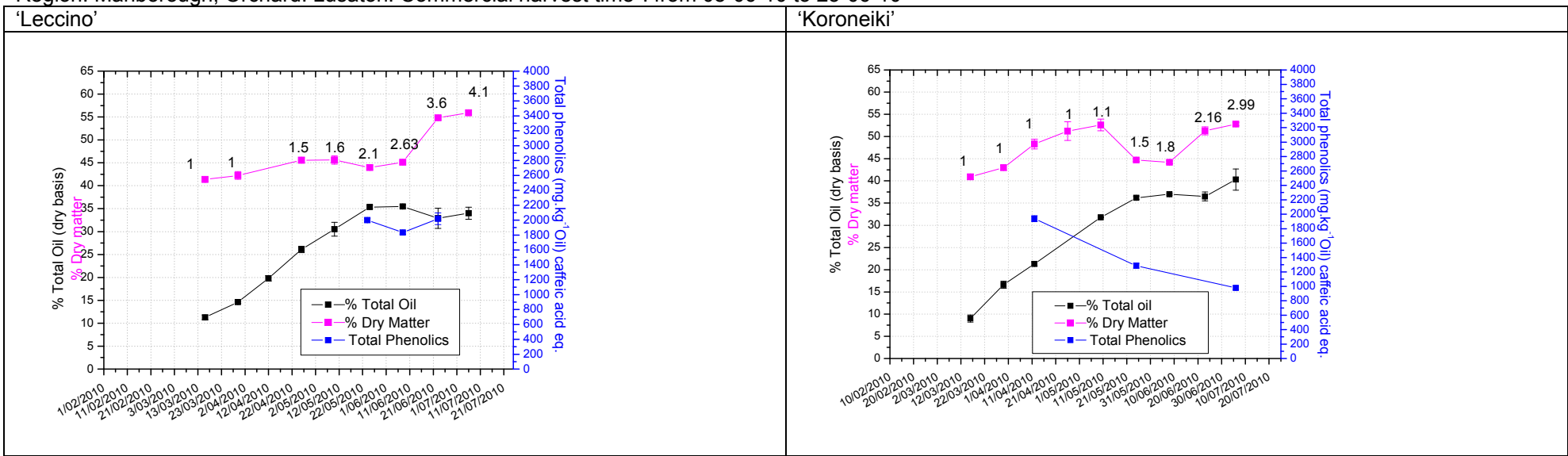*As advised by grower, or estimated from communications with grower

Region: Nelson, Orchard: Tasman Bay. Commercial harvest time*: from 21-06-10

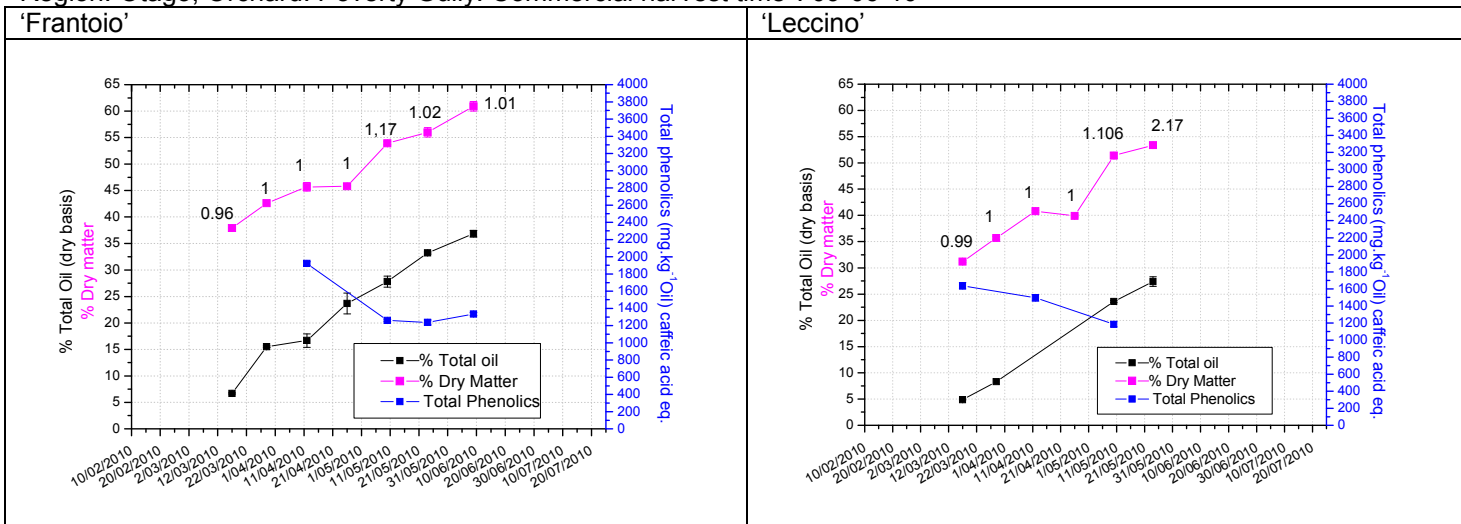| 'Frantoio'	'Leccino'	'Picual'
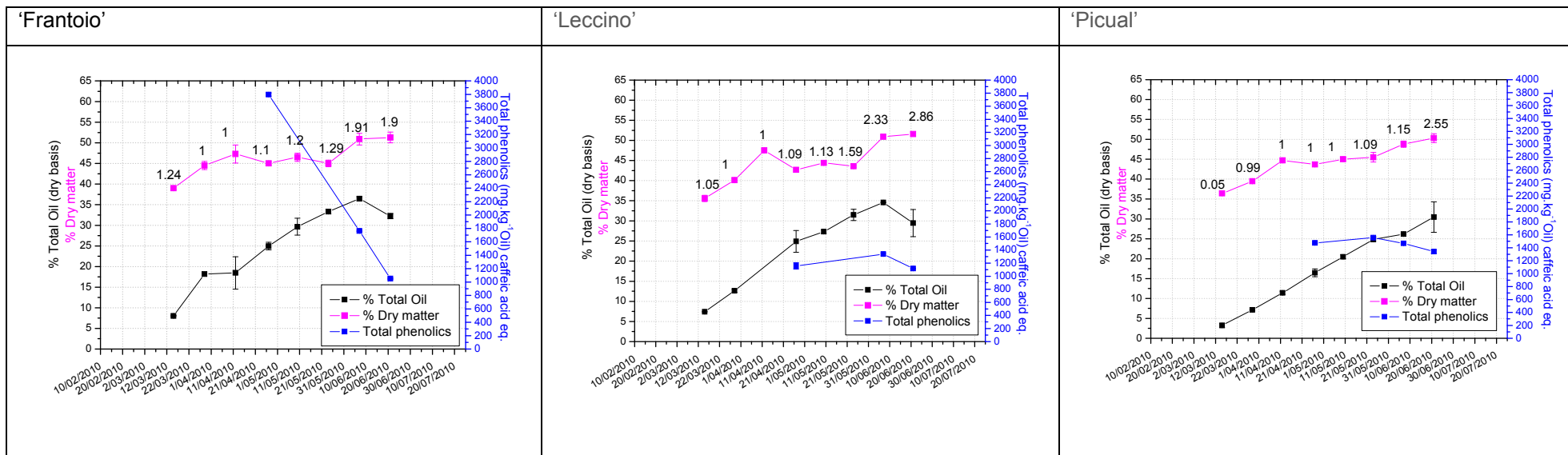
*As advised by grower, or estimated from communications with grower

Region: Marlborough, Orchard: Lusatori. Commercial harvest time*: from 08-06-10 to 23-06-10

'Leccino'	'Koroneiki'

*As advised by grower, or estimated from communications with grower

Region: Otago, Orchard: Poverty Gully. Commercial harvest time*: 09-06-10

'Frantoio'	'Leccino'

*As advised by grower, or estimated from communications with grower

Region: Otago, Orchard: Cairnmuir. Commercial harvest time*: from: 22-06-10 to 05-07-10
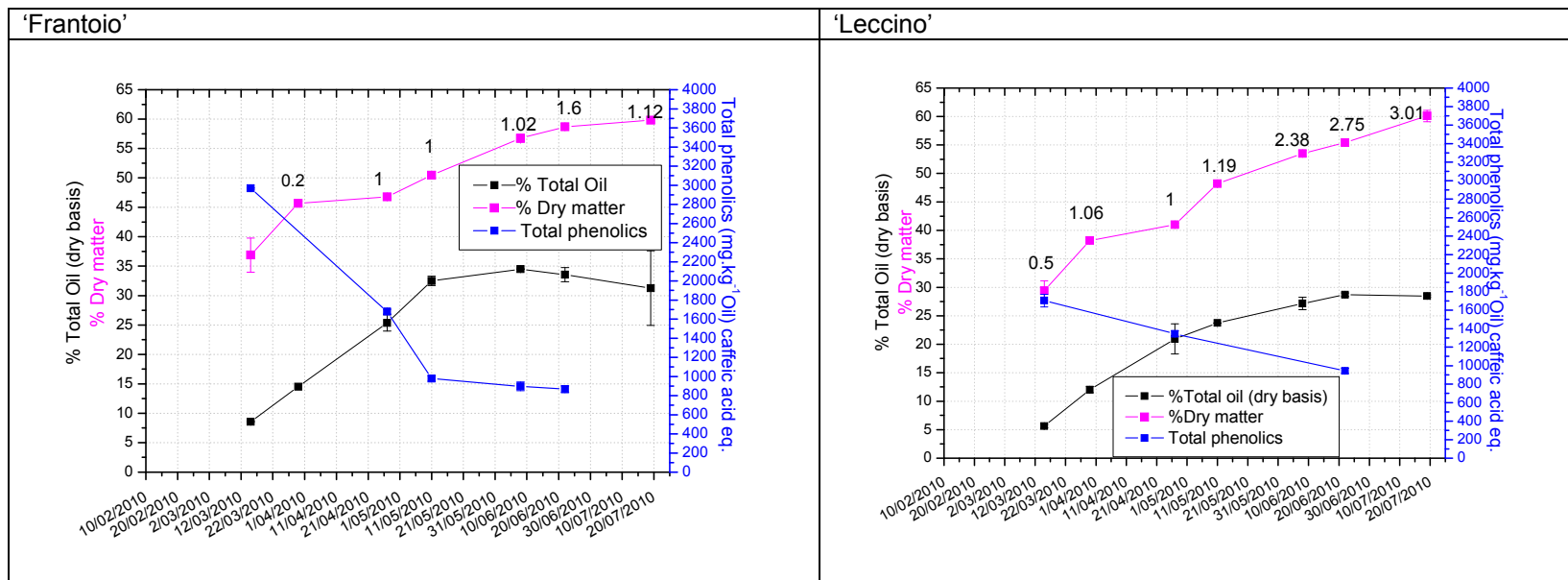
'Frantoio'	'Leccino'

*As advised by grower, or estimated from communications with grower.

Year 1 - 2008 olive season; Auckland, three cultivars, cold pressed extraction

Three cultivars of olives, 'Koroneiki', 'Leccino' and 'Frantoio', were harvested in 2008 at three stages of maturity from four orchards within the greater Auckland region. The fruit were collected at three nominated harvest maturities: early, mid and late harvest, to determine the effect of harvest maturity on fruit makeup as well as on the physical, chemical and sensory profiles of the oil.

Fruit were immediately cold pressed after harvest at the PFR pilot plant facility. All cold pressed oils were found to meet Olives New Zealand extra virgin quality standards. The most important findings were:

- Dry matter percentage showed inconsistency in trends and it seemed to be greatly affected by weather conditions such as rainfall.

- For the period of study, an inverse relationship between fruit firmness and oil content was observed.

- The maturity index, which is based on the colour of the fruit, did not relate well to the oil content i.e., maximum oil content was achieved at different colour maturity indices.

- On the other hand, the phenolic content better related to changes in oil content and maturity. In the early harvests, the total phenolic content was high and similar amounts were found between the different cultivars and the different orchards. This value then decreased in the later harvests and significant differences between varieties occurred.

- Scientific sensory analysis of the cold pressed oils determined for the first time the flavour and aroma profiles of the three cultivars studied. 'Koroneiki' olive oil had the highest amounts of olive fruitiness, rocket aroma and cut grass aroma and flavour attributes. 'Frantoio' olive oils had the highest scores for pungency, and 'Leccino' olive oils were highest in the vanilla toffee and nutty aromas and flavours. While pungency generally decreased from early to late harvest, sensory profiles were influenced more by the individual orchard site and fruit variety.

- In year 1 of the SFF project, all the oils were cold pressed. The fatty acids in all oils were well within the IOC standards, although oil from 'Koroneiki' from Helensville (Amour) was found to contain 0.99% linolenic acid (IOC standard is 1%). 'Koroneiki' oils from Mangawhai (Campbell) had 5.10% campesterol, which was well over the IOC limit of 4%. In this 2008 year, we identified that olive oils produced in New Zealand had higher amounts of tocopherols than

reported in the same cultivars harvested in the northern hemisphere. 'Koroneiki' oil, which was found to have 210-460 mg/kg total tocopherols from the four orchards sampled, was higher than had been reported in 'Koroneiki' oil from Greece (Psomiadou, 2009).

6.2.2   Year 2 - 2009 olive season; Auckland & Hawke's Bay, 'Frantoio'

During 2009, the research was focused on the 'Frantoio' cultivar, as this is one of the main cultivars planted in New Zealand. Two orchards in two regions were selected: South Auckland (Simunovich Olive Estate) and Hawke's Bay (Matapiro Olive Estate). Fruit were collected at several stages of maturity two to three weeks apart, starting at the very immature green stage to the black stage of maturity (from February to July 2009).

Oils were obtained by the accelerated solvent extraction (ASE) method. Fruit were also cold pressed to extract oil at three selected stages of maturity around the main commercial harvest. In addition, cold pressed olive oils and samples of fruit were received from various growing regions in the South Island at commercial harvest time only.

The data were analysed to find which fruit characteristics best reflected the content and quality of the oil. Emphasis was given to the phenolic content of the cold-pressed oils, because these contribute to the unique organoleptic attributes, shelf life and health perception of olive oil.

The main findings were:

- The colour-based maturity index did not accurately reflect the maturity of the fruit grown under New Zealand conditions.

- A strong correlation was found between % oil (in a dry weight basis) and absolute total solids per fruit (in grams), indicating that this way of measuring dry matter could be used as a more robust indicator of oil content than % dry matter content. A relationship was found between dry matter content (% of the total fruit weight), and % oil content (on a dry weight basis). However, this relationship was better at the beginning of the season and became weaker towards the mid season. Dry matter percentage can be easily influenced by extreme weather patterns, reducing its ability to predict oil content to contribute to the right harvest timing decision.

- The inverse relationship found between % oil and fruit firmness found in 2008 was confirmed in 2009 studies. This provided an opportunity to design or adapt from other industries a small device for in-field testing of firmness as an indicator of oil content and maturity.

- Total phenolic content in the oil decreased rapidly in the later stages of oil accumulation. The flavour characteristic of the oil could be dramatically influenced by either harvesting just before the oil content reached a plateau, or waiting until the content had been stable for some time. Less than 10% of the total phenolics available in the whole fruit were transferred into the cold-pressed oil.

- Samples of fruit and commercially pressed oil received from the South island orchards showed concentrations of some compounds to be above the limits established by international olive council.