



ORIGINAL RESEARCH ARTICLE

# Grape berry size is a key factor in determining New Zealand Pinot noir wine composition

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## ABSTRACT

Making high quality but affordable Pinot noir (PN) wine is challenging in most terroirs and New Zealand (NZ)'s situation is no exception. To increase the probability of making highly typical PN wines, producers choose to grow grapes in cool climates on lower fertility soils while adopting labour intensive practices. Stringent yield targets and higher input costs necessarily mean that PN wine cost is high, and profitability lower, in affordable varietal wine ranges.

To understand if higher-yielding vines produce wines of lower quality we have undertaken an extensive study of PN in NZ. Since 2018, we established a network of twelve trial sites in three NZ regions to find individual vines that produced acceptable commercial yields (above 2.0 kg per metre of row) and wines of composition comparable to "Icon" labels. Approximately 20 % of 660 grape lots (N = 135) were selected within a narrow juice Total Soluble Solids (TSS) range of  $22.0 \pm 1.0$  °Brix and made into single-vine wines under controlled conditions.

Multiple Factor Analysis of the vine, berry, juice and wine parameters from three vintages found grape Berry Weight to be the most effective clustering variable. As the Berry Weight category decreased, there was a systematic increase in the probability of higher berry red colour and total phenolics with a parallel increase in wine phenolics and decreased juice amino acids. The influence of berry weight on wine composition would appear stronger than the individual effects of Vintage, Region, Vineyard or vine Yield. Our observations support the hypothesis that it is possible to produce PN wines that fall within an "Icon" benchmark composition range at yields above 2.5 kg per vine, provided that the Leaf Area:Fruit Weight ratio is above 11 cm<sup>2</sup> per g, mean berry weight is below 1.2 g and juice TSS is above 22 °Brix.

**KEYWORDS:** Pinot noir, grape, vine, wine, yield, quality, region, terroir

## INTRODUCTION

Most winegrowers would argue that high-quality Pinot noir wines are predominantly a result of the composition of the grapes.

Making high-quality but affordable Pinot noir wine is challenging in most terroirs and New Zealand's situation is no exception. Pinot noir is also considered by viticulturists and winegrowers to be a difficult variety to produce, being environmentally responsive (Jackson, 2008; Nicholas *et al.*, 2011; Blank *et al.*, 2019), with thin skins, high susceptibility to diseases, and lower anthocyanin concentrations often leading to reduced wine colour intensity (Damberg *et al.*, 2012).

To increase the probability of making highly typical Pinot noir wines, producers choose to grow grapes in cool climates on lower fertility soils while adopting labour-intensive practices. Stringent yield targets and higher input costs necessarily mean that Pinot noir wine cost is high, and profitability reduced, in lower-priced varietal wine ranges. Pinot noir grapes destined for ultra-premium or icon wines are typically expensive to produce because of even more intensive vineyard management (Reynolds *et al.*, 1994; Keller *et al.*, 2005; Uzes and Skinkis, 2016). Maintaining an appropriate balance between vegetative growth and reproductive growth is an important viticultural management decision for improved grape and wine composition. Song *et al.* (2014) investigated the relationship between Pinot noir grapevine vigour and grape and wine composition, where vine vigour was determined by plant cell density (PCD) index obtained from aerial photography. As vine vigour decreased, total soluble solids in grapes, total phenolics and anthocyanins in wines increased while titratable acidity and yield decreased. However, research has demonstrated that the relationship between restricted yield and improved wine quality is not always evident. Reynolds *et al.* (1996) showed yield reduction and/or vertical canopy division reduced canopy density (Scott Henry training) in Pinot noir grown in cool climate conditions improved fruit quality and wine composition (lower titratable acidity (TA) and pH; higher anthocyanins and ethanol) while sensory evaluation demonstrated increases in wine varietal character. These results may, however, have been largely due to increased grape ripeness, which is beneficial in marginal climates but is less difficult to achieve in more favourable situations. In a study in Central Otago, New Zealand, Rutan *et al.* (2018) found that crop thinning increased the phenolic content of Pinot noir wines and influenced their volatile fraction, resulting in higher scores for fruity and sweet and lower for herbaceous and acidic even when all treatments achieved high sugar ripeness. Research by Reeve *et al.* (2016) found a positive effect of crop thinning on Pinot noir berry soluble solids and pH. In a later study by the same authors (Reeve *et al.* 2018) and a study by Mawdsley *et al.* (2018), both studies found that cluster thinning had no consistent effect on Pinot noir berry composition, wine colour or phenolic profile. The growing season had a greater effect on

both berry and wine composition, suggesting an interaction between season and yield, making it difficult to apply standard canopy to yield metrics between seasons. Mawdsley and colleagues concluded that initial yields were at, or below, a balanced crop load, hence thinning had minimal impact on berry or wine composition. A study conducted by Ulmer and Skinkis (2020) compared vine productivity (fruitfulness, yield and berry ripeness) between cane- and spur-pruned Pinot noir vines under cool climate conditions. Results showed pruning method had no effect on vine phenology, yield, berry weight or berry ripeness (TSS, measured as °Brix, pH and TA) at harvest.

Grape berry composition is an important variable influencing final wine composition and quality (Ribéreau-Gayon *et al.*, 2006). Medina-Plaza *et al.* (2021) showed the relationship between berry cell wall composition and phenolic extractability in Pinot noir was mainly site-specific, whereas grape berries grown in the same region exhibited similar phenolic extractability. Where anthocyanin and polymeric pigment content was high in the skins, there were greater amounts extracted in the final wine. Robust small-scale standardised winemaking is, therefore, important for determining the full effects of viticultural factors on wine style and quality.

Furthermore, oenological techniques to improve wine quality may be tested rigorously and cost effectively at small scale. Sampaio *et al.* (2007) demonstrated that research-scale 3.5-kg Pinot noir ferments produced wines with similar fermentation kinetics and composition to those of small commercial-scale 4–5 tonne ferments. However, while skin proanthocyanidin extraction was equivalent, seed proanthocyanidin extraction lagged behind that of the commercial-scale ferment (Sampaio *et al.*, 2007). Sparrow and Smart (2015) compared several different sizes of research-scale fermenters, including the 1.5-L Bodum® coffee plunger as well as 250-mL, 20-L and 0.5-ton “tanks”. Wine phenolic composition was similar across all different ferment sizes; however, the authors noted that wine volatile composition should also be considered, as differences may arise from the different surface area to volume ratios of the vessels (Sparrow and Smart, 2015). Similar temperature gradients between the cap and liquid portions were noted in 50-L and 1600-L Pinot noir ferments by Schmid *et al.* (2009), and provide further evidence that research-scale ferments can be representative of commercial-scale fermentation. Integral to our study has been the development of a cost-effective winemaking methodology for single-vine Pinot noir grape lots to study the transfer, reaction and retention of secondary metabolites in Pinot noir wine made from the vine ideotypes.

Manipulation of a single element of vine management can lead to a wide array of unintended outcomes that affect wine composition. This research studies *in situ* variation of individual vine performance in an attempt to identify and describe the appearance and behaviour of an ideal vine in the context of specific Pinot noir production goals. The wide range of management and biological factors that ultimately determine a wine's composition and its consequent quality

attributes include key decisions, made by winemakers and viticulturists, such as when to harvest the grapes or the duration of maceration. As much as is possible we seek to control factors such as region or vineyard to better focus on the effects of vine yield and vine ideotypes on Pinot noir juice and wine composition. In an earlier publication from our research into New Zealand Pinot noir production (Martin *et al.*, 2020) we have shown that the climatic conditions of two very different vintage years dominate other factors likely to influence grape composition, such as crop load, region and vineyard. We observed large variations in performance of the same vines between seasons, thus excluding factors that are stable between seasons as primary causes. Changes in management of the same vine from year to year, such as differences in retained node number post pruning, appear to be the most likely contributors to within-vineyard variation. Amongst the population of vines in the study we also found significant negative relationships between vine yield and grape quality indicators, demonstrating the quality risk associated with higher yield. Nevertheless, we did also find a small proportion of vines that appeared to meet grape quality benchmarks and achieve acceptable commercial yields (> 1.75 kg per linear metre of trellis) as specified by the programme industry advisory group. The current study extends to the composition of the wines made from the individual monitored vines and adds one additional full season to our dataset.

## MATERIALS AND METHODS

### 1. Vineyard Monitoring

Vineyard, vine selection and vine monitoring methods for the study are fully described in Martin *et al.* (2020). In summary, 20 individual vines in four vineyards in each of three New Zealand Pinot noir growing regions (Central Otago, Marlborough and Wairarapa) have been monitored (Table 1).

The study network represents a maximum total of 12 vineyards and 240 individual vines per vintage.

Within 2 days prior to commercial harvest the monitored vines in each vineyard were hand harvested and yield components were determined. Occasional events such as frost damage or logistical problems prevented data collection at some sites in some years. For four vintages from 2017/18 through to 2020/21, data were collected from an average of 11 of the 12 vineyards per year. The total vine x year population that provided all the vine performance and grape composition information was 880.

Processes for the collection of meteorological information are also fully described in Martin *et al.* (2020). In short, regional data were sourced from weather stations operated by the National Institute for Water and Atmosphere (NIWA). Local weather information for each vineyard was sourced from the nearest Plant and Food Research (PFR)- or NIWA-managed station, which was typically at a similar elevation and within 2 km of the study vineyard. Seasonal and regional variations in weather were compared using climate metrics expressed as accumulated deviations from Long Term Average (LTA) data (Agnew and Raw, 2019). For regional and seasonal comparisons, daily Growing Degree Days above 10 °C (GDD) were calculated for the growing season period from 1 September to 30 April for each vintage year. Seasonal Water Balance (SWB) on any given day was calculated as the difference between average daily ET<sup>o</sup> and average daily rainfall observed over the preceding 90-day period. The accumulated daily SWB was then subtracted from the LTA accumulated SWB to give the SWB deviation. Accumulated Diurnal Variation (DV) was calculated as the sum of the difference between daily maximum and daily minimum temperatures from 1 September to 30 April in each season. The accumulated daily DV was then subtracted from the LTA accumulated DV to give the DV deviation. For vineyard-based comparisons the accumulated growing degrees were calculated for the phenological period from budburst to harvest observed at each vineyard.

**TABLE 1.** Summary details for the vineyard blocks selected for the New Zealand Pinot noir Ideal Vine study. All blocks are Pinot noir Abel clone grafted to 3309C rootstock and trellised to a Vertical Shoot Position canopy. A map of vineyard locations and further information can be found in Martin *et al.* (2020).

Vineyard ID	Region	Sub region	Block area (ha)	Year planted	Row spacing (m)	Vine spacing (m)	Pruning system	Target yield (kg/m)	End-use class
OA	Otago	Bannockburn	0,30	2000	2,20	1,13	2-cane	1,3	Icon
OB	Otago	Bannockburn	0,13	2008	1,60	0,90	2-cane	1,1	Icon
OC	Otago	Bendigo	0,50	1996	1,50	0,90	2-cane	1,1	Icon
OD	Otago	Pisa Range	1,56	2008	2,40	1,50	10-spur	2,3	Affordable
MA	Marlborough	Brancott	0,08	1993	1,50	1,25	2-cane	1,0	Icon
MB	Marlborough	Brancott	0,72	2006	3,00	1,40	2-cane	2,5	Affordable
MC	Marlborough	Waihopai	4,62	2013	1,60	1,25	2-cane	2,0	Affordable
MD	Marlborough	Wairau	0,75	2005	1,80	1,15	2-cane	1,1	Icon
WA	Wairarapa	Martinborough	0,28	2003	2,00	1,20	2-cane	1,0	Icon
WB	Wairarapa	Martinborough	0,67	2009	2,40	1,40	2-cane	1,8	Affordable
WC	Wairarapa	Te Muna	0,30	1998	2,40	1,25	2-cane	1,6	Icon
WD	Wairarapa	Te Muna	0,95	1999	1,60	1,20	2-cane	1,0	Icon



## 2. Leaf area estimation

The theoretical total leaf area of each vine was derived from a canopy RGB photograph. Vines were photographed immediately prior to harvest using either a Nikon® D70 DSLR or a Canon® EOS 500D DSLR camera with a background screen which, depending on region and season, was either red or pale blue. A small white board of known dimensions (250 mm x 250 mm) was typically used to label the vine in the photograph and as a reference to scale the image. Photography was planned to be undertaken in cloudy (diffuse light) conditions to minimise shadowing within the canopy and onto the background screen, although this was not always possible. Batch pre-processing to scale the images was undertaken using imageJ© software.

Leaf layer number and leaf area per vine were derived from the RGB images using imAGE.exe© software as per the method described by Hill *et al.* (2011). As many Pinot noir vine canopies were very sparse and because the fruit zone was often highly defoliated, photographs of the vines were manually cropped into three different zones using GIMP© 2.10.8 software.

The first crop zone spanned the approximate perimeter of the vine canopy between adjoining plants and from the fruiting wire to the trim height ( $A_v$ ). This image zone was used to determine the proportion of “green” (Hue range 35–205) vine leaves ( $L_v$ ) in  $A_v$  using the open source imAGE.exe software. The proportion of vine gaps ( $G_v$ ) was estimated as  $(100 - L_v)$ .

Because many Pinot noir canopies were sparse, each image was further cropped (B) to a smaller area of 500 x 500 pixels (approximately 500 x 500 mm) of typical canopy density above the first foliage wire. The aim was to select an image comprising leaves and smaller intra-leaf gaps but excluding large areas of empty space without shoots. This image was used to calculate the proportion of canopy leaves ( $L_c$ ) and canopy gaps ( $G_c = 100 - L_c$ ) and to derive the leaf layer number (LLN<sub>c</sub>) from the formula published by Hill *et al.* (2011). Results from this analysis were also sense checked against unpublished Pinot noir canopy density measurements of LLN using Point Quadrat (Smart and Robinson, 1991), which ranged from 0.7 (very sparse) to 3.3 (very dense).

A third cropped zone ( $C_f$ ) that spanned the fruit zone (from the fruiting wire to the first foliage wire) was undertaken to calculate the proportion of fruit zone leaf ( $L_f$ ) and fruit zone canopy gaps ( $G_f = 100 - L_f$ ) to derive the fruit zone leaf layer number (LLN<sub>f</sub>). Leaf Area per Vine (LAV) was calculated as:

$$LAV = (A_v \times V_c \times LLN_c) + (C_f \times L_c \times LLN_f) - C_f$$

### 2.1. Shoot number and dormant canopy assessments

The monitored vines were also photographed with a background screen after leaf-fall and the images scaled using the same techniques as the pre-harvest photography. From the images, shoots on the monitored vine were manually counted and their lengths assessed using the measuring tool on GIMP 2.10.12. The on-screen diameter of each shoot was categorised into three classes (< 5 mm, 5–10 mm and > 10 mm at the third internode) as per the field-based method described

by Greven *et al.* (2014). The count nodes per vine that did not carry a shoot were recorded as “blind buds”.

## 3. Grape berry analysis

All bunches from each vine were counted and a combined weight taken. If there were more than 20 bunches per vine, a random 20-bunch sub-sample (approximately 2 kg) was selected for berry sampling. When bunch numbers were less than 20, all bunches on a vine were used for berry sampling. Each bunch was dissected to ensure that berries in the centre of, or internal to, the bunch were representatively sampled. Three sub-samples (1 x 50 berries, 1 x 200 g and 1 x 50 g) were taken from each vine. The 50-berry fresh sample was weighed and recounted to calculate mean berry weight. Berries were crushed by hand while in the plastic sample bag and the crushed fruit pressed using manual pressure through a small kitchen sieve with approximately a 1-mm mesh. Juice was centrifuged in a 50-mL tube for 10 minutes at 4600 rpm using a Heraeus® Multifuge 3SR+ and analysed for TSS, pH and titratable acidity. TSS was determined on a hand-held Atago refractometer, while pH and TA were determined on a Mettler Toledo T70 autotitrator. Acid concentration was determined using an end-point titration to pH 8.2. Aqueous sodium hydroxide (0.1 M) was used as titrant and TA was expressed as g/L of tartaric acid equivalents (Iland *et al.*, 2000; Iland *et al.*, 2004). The 200-g (frozen) grape samples were analysed for tannin, colour and phenolics using a modified method originally developed by the Australian Wine Research Institute for their WineCloud™ service:

[https://www.awri.com.au/commercial\\_services/analytical\\_services/the-winecloud/](https://www.awri.com.au/commercial_services/analytical_services/the-winecloud/)

<https://www.awri.com.au/wp-content/uploads/2013/08/sample-prep-guide-grape-portal.pdf>

<https://www.awri.com.au/wp-content/uploads/2014/01/measuring-grape-tannins.pdf>

At the time of analysis, which was within 3 to 6 months of harvest each year, 70 % of the grapes in each sample were allowed to fully thaw at room temperature for approximately 1 hour, while the remaining 30 % were kept frozen until homogenisation. The thawed and frozen samples were combined and homogenised using a 1200 Series Nutribullet® for 2 x 1-minute cycles, to ensure all seeds were thoroughly disintegrated. The homogenate was stirred and 1 g was weighed into a 50-mL centrifuge tube, in duplicate, where 10 mL of acidified 50 %v/v ethanol/Milli-Q water solution was added. The samples were allowed to extract for 1 hour with constant mixing via a Chiltern® rotating wheel. The homogenate was centrifuged Heraeus® Multifuge 3SR+ at 4600 rpm for 5 minutes. Once clarified, 1 mL of the supernatant was placed in a new 50-mL tube and diluted with 10 mL of 1.0M HCl to make a total volume of 11 mL. Samples were incubated for 1 hour in a dark room. Absorbance readings expressed in Absorbance Units (AU) were taken of each duplicated sample at 280 (Optical Density (OD) 280), 320 (OD320) and 520 nm (OD520) with a

10-mm quartz cuvette using a Thermospectronic® Genesys 10 spectrophotometer.

#### 4. Berry quality benchmarks

The wines produced from the study network of 12 commercial vineyards comprise eight single-vineyard “Icon” wines and four multi-vineyard blend “Affordable” wines (Table 1). Of the eight “Icon” wines, several are considered to be among New Zealand’s best examples of Pinot noir (Jukes and Stelzer, 2019; Parr *et al.*, 2020). The process of establishing annual benchmark ranges for grape berry weight and basic berry composition parameters (TSS, TA, pH, OD280, OD520) is fully described in Martin *et al.* (2020). With additional data, the method was improved such that half the “Icon” vine population was used to derive the specification ranges while the remaining half was included in the analysis dataset. In other respects, the methods to determine whether fruit from an individual vine was “in-spec” were the same as previously published (Martin *et al.*, 2020).

#### 5. Winemaking

Using basic yield and fruit maturity parameters as the selection criteria, grape lots from individual vines that displayed a wide yield range (0.9 to 8.0 kg/vine) but for which berry total soluble solids (TSS) were within a relatively narrow standard deviation range ( $22.7 \pm 0.85$  °Brix) were made into wine. There were many vines with yields lower than 0.9 kg but where there were insufficient fruit to make wine. Winemaking occurred in 2018, 2019 and 2021 but could not be carried out in 2020 because of COVID-19 restrictions. The selected vines for winemaking represented a subset of approximately 20 % (N = 123) of the overall monitored population.

Grapes were hand harvested as single-vine fruit lots from vineyards in Central Otago, Martinborough and Marlborough and transported (chilled) to the Marlborough Research Centre. A standard storage period of 24 hours from harvest date was applied to all grapes before processing, to remove the variable of transport time to Marlborough. Depending on vine yield, all the grapes from a vine or a maximum 2-kg subsample were crushed in a manual crusher (Marchisio Cervino 400–600 kg/H). Rachis were removed by hand and a standard sulphur dioxide (SO<sub>2</sub>) addition (40 ppm) was added as potassium metabisulphate (PMS) at crushing.

Must was loaded into stainless steel fermentation tubes (1.5 or 2 kg capacity) and cold soaked for 3 days at 6 °C. The tube was screw-capped at both ends, the base closed with a pipe-blank cap and the top with an air-lock cap. To effect mixing and extraction, the air-lock cap was swapped for a blank cap, the fully sealed fermenter was then slowly inverted and the upper blank cap swapped for an air-lock cap. A small 40-mL juice sample was collected 24 hours after cold soak began (see Juice analysis). During the cold soak period, tubes were inverted once daily to mix gently. Must was then warmed to 18 °C and inoculated with RC212 yeast (Lallemand, Denmark) (rate 250 mg/L) and fermented at 25 °C. A standard yeast Superfood® (BSM™ Wine Division, Napa, USA) addition of 600 mg/L was made to every ferment.

Where juice yeast available nitrogen (YAN) concentrations were below 250 ppm N, an additional diammonium phosphate (DAP) addition was made to raise YAN concentrations up to a minimum of 200 ppm N. Tubes were inverted once daily to gently submerge the cap during fermentation. Fermentation was monitored by total weight loss on a daily basis. Towards the end of fermentation, TSS (°Brix) were monitored using an Anton Paar DMA 35 portable density meter and when °Brix values dropped below 0, residual sugars were determined by an enzymatic assay kit. When ferments contained less than 2.0 g/L residual sugars, primary fermentation was deemed complete. Ferments were pressed after a standardised 15-day maceration period, which included the 3-day cold-soak duration. Ferments were pressed in a 4-kg compressed air bench press (Custom manufacture, Marlborough) under a cover of argon gas and a pressing regime of 2 minutes at 2 Bar was applied. An addition of 50 mg/L SO<sub>2</sub> (in the form of PMS) was made to the pressed wine. Wine was settled for one week, then racked off yeast lees. No malolactic fermentation (MLF) was undertaken on the small-volume (0.6 to 1.2 L) research-scale wines. Samples were taken for chemical analyses (see Wine analysis). A standard addition of 1 g/L of tartaric acid was made to the pressed wine. SO<sub>2</sub> concentrations were monitored before wines were subsampled and adjusted to maintain a molecular SO<sub>2</sub> of approximately 0.5. Wines were subsampled using the “Wine in Tubes” system (George Michel Wine Estate, Marlborough, NZ) to transfer wine to glass vials (100 mL volume) under inert gas protection to minimise oxygen pick-up. Wine subsamples were stored at low temperatures (0 to 2 °C) for further analytical phenolic and volatile chemistry analyses.

#### 6. Juice analyses

Juice samples taken 24 hours after the cold soak period began were subjected to a range of winemaking analyses. These included: TSS content, acidity (pH) and acid concentration (TA), which were measured in the same way as for the berry samples.

In 2018 and 2019 primary amino acid (PAA) concentrations were quantified by the NOPA (nitrogen by o-phthalaldehyde) method with a reducing agent in basic medium generating a chromogen and measured spectrophotometrically (Dukes and Butzke, 1998). For 2021 samples PAA concentrations were quantified using a Biosystems Y15 auto-analyser (Barcelona, Spain), in duplicate against a five-point standard curve ( $R^2 > 0.98$ ) using a method adapted from the Compendium of International Methods of Wine and Must Analysis (International Organisation of Vine and Wine, 2016) and (Zoecklein *et al.*, 2013).

Free and total SO<sub>2</sub> were determined using Biosystems Y15 auto-analyser. Results were used for winemaking quality/process control and are not reported as treatment-related data.

Reducing sugars (glucose and fructose) were quantified by enzymatic assay based on the reduction of NADP (nicotinamide adenine dinucleotide phosphate). The reaction was monitored at 340 nm using Biosystems Y15

auto-analyser (Barcelona, Spain). Results were used for winemaking quality/process control and are not reported as treatment-related data.

Spectrophotometric assays were run on a Molecular Devices (San Jose, California, USA) Spectramax 384 Plus with a 1cm pathlength cuvette reference correction. Optical density was measured directly in a UV-transparent 96-well microplate at 280 and 520 nm. Samples were centrifuged or filtered before analysis and analyses were carried out in duplicate.

Cations, potassium, magnesium, calcium and ammonium, were quantified on an Agilent Capillary Electrophoresis instrument (Agilent Technologies, Santa Clara CA, USA) using a solution of pyridine (10.0 mM), glycolic acid (12.0 mM) and 18-crown-6 (6 mM) (pH 3.5) as a background electrolyte buffer. All samples were diluted 5-fold in a solution containing lithium chloride as an internal standard and frozen at  $-80^{\circ}\text{C}$  for a duration typically between three and nine months. Juice samples were thawed, vortexed to mix and filtered through a 0.22- $\mu\text{m}$  syringe filter prior to injection. All samples were run in duplicate and quantified on a five-point standard curve ( $R^2 > 0.98$ ) (Rovio *et al.*, 2011).

Organic acids, tartaric and malic, were quantified on a Shimadzu Prominence, High Performance Liquid Chromatography (HPLC) (Shimadzu Corporation, Kyoto, Japan) system with an isocratic elution phosphate buffer (25 mM, pH 2.3) on an Allure Organic Acids Restek column (5  $\mu\text{m}$ , 240 x 4.6 mm) at  $30^{\circ}\text{C}$ . All samples were diluted 5-fold in phosphate buffer and filtered through a 0.22- $\mu\text{m}$  syringe filter prior to injection. All samples were run in duplicate and quantified on a six-point standard curve ( $R^2 > 0.98$ ). Organic acid methods were adapted from a published protocol (Shi *et al.*, 2011).

Yeast-preferred amino acid profiles were quantified on a fresh juice sample on an Agilent 1200 Series HPLC (Agilent Technologies) using the manufacturer's method (Agilent Biocolumns, 2018). A standard mix of 17 amino acids was purchased from Agilent. Additional standards (asparagine, hydroxyproline, glutamine and tryptophan) were purchased and prepared separately. All standards and samples contained internal standards sarcosine (100 ppm) and  $\alpha$ -aminobutyric acid (100 ppm). All samples were run both undiluted and diluted 5-fold. Samples were filtered through a 0.22- $\mu\text{m}$  syringe filter prior to injection. All samples were run in duplicate and quantified on a four-point standard curve ( $R^2 > 0.98$ ).

## 7. Wine analyses

Wine samples were subjected to a range of primary and secondary metabolite analyses:

Wine pH, TA, reducing sugars (glucose and fructose) and optical densities were determined using the same protocols as for juice analysis. Alcohol was measured using an Anton Paar wine alcolyzer (Anton Paar, Austria). All measurements were taken in duplicate with  $< 0.02$  v/v % variation.

Colour parameters, (total anthocyanins, colour density, hue, total phenolics and  $\text{SO}_2$ -resistant colour) were determined

by the modified Somer's assay (Somers and Evans, 1977; Mercurio *et al.*, 2007). Monomeric anthocyanins were quantified by the pH differential method (Lee *et al.*, 2005) adapted for the analysis of grape juice and wine on a Spectramax 384 Plus platereader (Molecular Devices, San Jose CA, USA). Results were expressed as mg/L malvidin-3-glucoside (M3G) equivalents. Tannins were determined by the methyl cellulose precipitable (MCP) tannin assay using a plate reader (Mercurio *et al.*, 2007).

### 7.1. Monomeric phenolics by HPLC

Monomeric phenolics tannins were analysed at the School of Chemical Sciences at the University of Auckland using a method based on the work of Peng *et al.* (2002) and Garrido-Banuelos *et al.* (2019). The chromatographic runs were performed on an Agilent 1200 series HPLC system, consisting of a G1311A quaternary pump, a G1322A degasser, a G2260A autosampler, a G1316A thermostated column compartment, and a G1315D diode array detector. Data were processed using Chemstation for LC 3D systems (Agilent, version B04.02). This method used a polystyrene/divinylbenzene reversed-phase chromatographic column (PLRP-S, 150 mm x 4.6 mm, 3  $\mu\text{m}$ , 100  $\text{\AA}$ , Agilent) with a binary mobile phase composed by 1.5 % (w/w) phosphoric acid in water (A) and acetonitrile (B). The gradient used was as follows: 5 % to 10 % B from 0 to 16.7 min; 10 % to 11.7 % from 16.7 to 25 min and at this proportion until 35 min; then from 11.7 % to 22 % from 35 min to 55; 22 % to 50 % from 55 min to 62 min and maintained at 50 % until 62 min; then from 50 % to 5 % from 62 to 65 min. Post-run was set at 10 minutes. The flow rate was set to 1 mL/min, the injection volume was 20  $\mu\text{L}$ , and the column temperature was  $40^{\circ}\text{C}$ . Gallic acid, syringic acid, catechin and epicatechin were monitored at 280 nm; hydroxycinnamic acids (quantified using caffeic and coumaric acids) and t-resveratrol at 320 nm; flavonol glycosides (as rutin equivalents) and aglycones (as quercetin equivalents) at 360 nm; and anthocyanins at 520 nm (as malvidin-3-glucoside (M3G) equivalents).

### 7.2. Wine Carbon isotope analyses

Wine samples were analysed for their carbon isotope ratios at the Geochemistry Laboratory of the University of Otago, New Zealand. For  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  measurements, 10- $\mu\text{L}$  aliquots of wine were pipetted into 3.5 x 5 mm tin capsules and the liquid removed by vacuum drying. Samples were combusted to  $\text{CO}_2$  and  $\text{N}_2$ , in an elemental analyser (Carlo Erba NA1100). The isotopic ratios of the respective gases were measured under a continuous flow system using a Delta Advantage Isotope-Ratio Mass Spectrometry (Thermo-Finnigan, Bremen, Germany). Final raw data were normalized to international scales (Carbon – VPDB and Nitrogen – AIR) using two certified reference materials of Glutamic acid (USGS 40;  $\delta^{13}\text{C} = -26.39\text{‰}$ ,  $\delta^{15}\text{N} = -4.52\text{‰}$  and USGS 41;  $\delta^{13}\text{C} = +37.63\text{‰}$ ,  $\delta^{15}\text{N} = +47.57\text{‰}$ ) and an EDTA laboratory quality control material (Elemental Microanalysis Ltd, Cornwall, UK). Isotopic values of carbon and nitrogen for EDTA are  $-38.52\text{‰}$  and  $-0.73\text{‰}$ , respectively.



Time-based instrumental drift was corrected for using the EDTA isotopic values measured between every 12 samples. Linearity correction was performed in a similar manner using EDTA standards. Furthermore, EDTA measurements ( $n = 16$ ) were used for the determination of analytical precision, which was  $\pm 0.20\%$  for  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$ . Categories of vine seasonal water status developed by Brillante *et al.* (2020) were applied to the dataset, albeit that in our study wine isotope ratios were measured rather than juice, as Spangenberg and Zufferey (2019) have shown that and juice and wine carbon isotope ratios are well correlated to one another and to pre-dawn leaf water potentials for a given variety.

## 8. Statistical analyses

Relative Standard Deviations (RSD) were calculated as the Standard Deviation/Sample Mean. Data exploration was initially undertaken using Principal Components Analysis (PCA) on a correlation matrix of the data. PCA analyses were undertaken using Microsoft PowerBI®. To further explore correlations between variables and groups for both datasets, multiple factor analysis (MFA) was performed using FactoMineR in the R statistical software package v4.2.0 (Le *et al.*, 2008). Variables were scaled to unit variance and categorised as groups (latent variables) to account for inter-group differences prior to MFA (Supplementary Table S3). ANOVA was performed using Genstat 20th edition (VSN International, Hemel Hempstead, UK). Comparisons amongst vineyard means were made using Fisher's Protected Least Significant Differences at  $\alpha = 0.05$  (5 % LSD). Regression analysis was undertaken using linear regression functions in Microsoft® Excel 2013 or in Genstat 20 if a  $P$ -value was required to assess regression significance.

## RESULTS

A graphic summary of the weather conditions in each region and each vintage year of the study is presented in Supplementary Figure S1. Commentary on the climatic

conditions of vintages prior to 2021 can be found in Martin *et al.* (2020). While there were inter-regional differences, in general terms, the 2018 vintage was characterised by a very warm spring/early summer followed by a wet and late/summer and autumn. The 2019 vintage was cool in the spring then warm and dry through the middle part of the season. The 2020 vintage was similar to 2019 although cooler and drier especially in autumn.

The 2020/21 season and vintage in Central Otago was "average" but unusual in so far as all three parameters plotted (GDD, SWB and DV) tracked closely to LTA all season. The 2020/21 season in Marlborough was warmer (higher GDD) and drier (lower SWB) than LTA especially from December onwards. Conditions in Wairarapa were similar to Marlborough but high rainfall in November caused the SWD to track above LTA until the end of February.

The spring period from September to November 2020 was warmer than LTA in all three regions and budburst and flowering stages were very advanced in Marlborough and Wairarapa. Despite the overall warmer conditions there were, however, numerous frost events, one of which severely damaged vineyard WA in the Wairarapa. Flowering conditions for Pinot noir in late November were unfavourable in Marlborough and Wairarapa but flowering in Otago was marginally later and warmer. While climatic factors, especially cool temperatures (and patchy frost damage to inflorescences) in the pre-flowering period affected yields, changes in management (bud load and/or shoot number) also contributed to yield variation between vines within a vineyard.

## 1. Harvest data

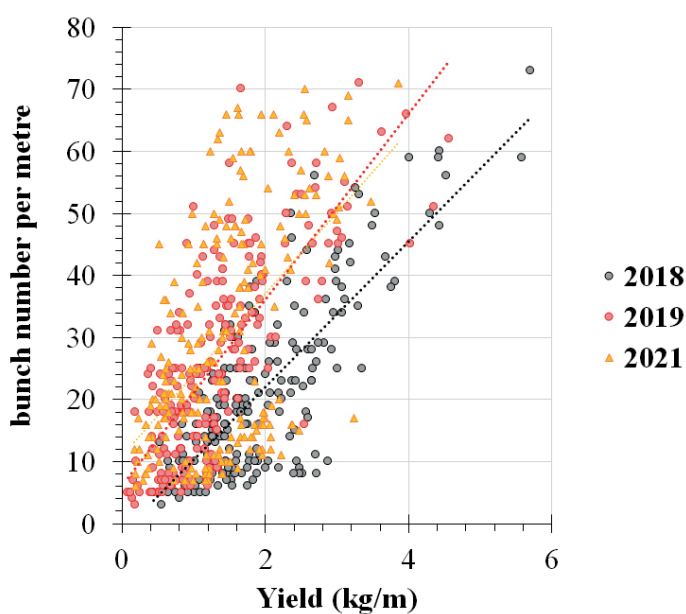
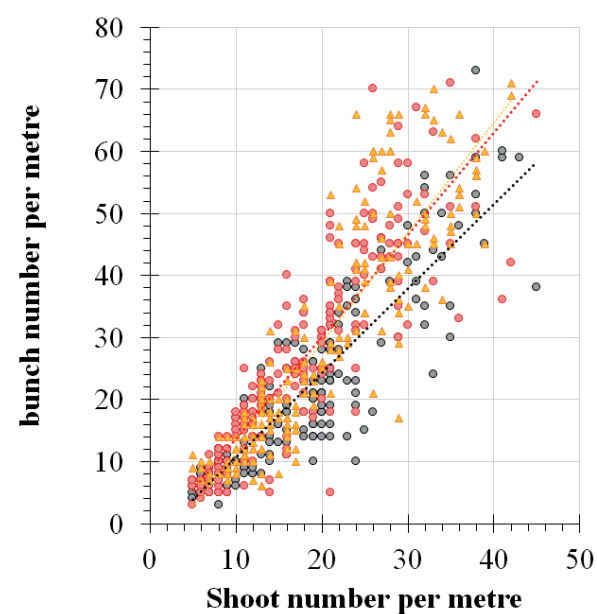
Harvest dates and phenology-based accumulated GDD for each vineyard are presented in Supplementary Table S1. Yield data shown in Table 2, while collected per vine, are expressed as kg of grapes per linear metre of vineyard row (kg/m) to account for differences in within-row plant spacings between

**TABLE 2.** Mean yield for the New Zealand Pinot noir Ideal Vine study network for vintages 2018 to 2021. Numbers in parentheses represent the Relative Standard Deviations. Vineyard details are described in Table 1.

Vineyard	Mean yield (kg/m)				
	2018	2019	2020	2021	Average
OA	1,62 (0,38)	1,04 (0,34)	1,44 (0,32)	1,79 (0,57)	1,47
OB	1,49 (0,27)	1,04 (0,44)	1,26 (0,52)	1,59 (0,45)	1,35
OC	1,88 (0,36)	0,69 (0,49)	0,72 (0,38)	1,42 (0,43)	1,18
OD	2,54 (0,35)	2,59 (0,46)	2,37 (0,23)	2,40 (0,74)	2,47
MA	1,38 (0,34)	1,40 (0,26)	3,59 (0,38)	1,38 (0,44)	1,94
MB	3,28 (0,42)	1,72 (0,45)	1,97 (0,32)	1,12 (0,48)	2,02
MC	2,06 (0,40)	1,99 (0,33)	2,84 (0,34)	1,88 (0,70)	2,19
MD	1,33 (0,44)	1,05 (0,46)	1,58 (0,56)	0,80 (0,43)	1,19
WA	1,40 (0,28)	0,84 (0,55)	1,31 (0,39)	n/a	1,18
WB	2,09 (0,28)	1,40 (0,48)	3,65 (0,35)	1,89 (0,70)	2,26
WC	1,95 (0,36)	n/a	n/a	0,66 (0,26)	1,30
WD	n/a	0,83 (0,47)	1,63 (0,32)	0,71 (0,38)	1,06

vineyards. Across the four vintages of study (N = 880) the mean yield per metre was 1.67 kg, the range was 0.08 to 6.32 kg/m (81-fold difference) with a standard deviation of  $\pm 0.99$  kg (RSD of 59 %). These statistics describe a highly variable population. Excluding a small proportion of vines (16/880) where a one-off seasonal yield anomaly occurred (e.g. < 0.5 bunches per shoot or mean bunch weights < 30 g), the yield range of “normal” vines was nevertheless 30-fold. Two vines at vineyard MD (MD10 and MD18) had consistently low mean bunch weights (in the range of 13 to 41 g across all years), the causes of which would merit closer investigation.

The differences between the highest and lowest phenology-based GDD between vineyards (Supplementary Table S1 within a vintage and region were typically small, while the GDD range was generally greater between regions within a vintage, and greatest between vintages. These results affirm that seasonal factors other than thermal time can also strongly influence the duration of phenological stages, especially from flowering onwards (Parker *et al.*, 2014).



**FIGURE 1.** Relationships between yield per metre and bunch number per metre (left) and shoot number per metre and bunch number per metre (right) for the New Zealand Pinot noir Ideal Vine study network in vintages 2018, 2019 and 2021.

## 1.2. Exploration of aggregated vine and grape berry data

All vine and berry variables measured across four vintages underwent MFA analysis. Variables were grouped and classification variables (Vintage, Region, Vineyard, Yield and Berry Weight) identified prior to MFA (Supplementary Table S3). Berry Weight (Figure 2) followed by Vintage (Figure 3) were the most effective clustering variables. The Berry Weight classes distributed sequentially along Dim1, with the lowest Berry Weight category scoring positively on Dim1 through to the highest Berry Weight category scoring

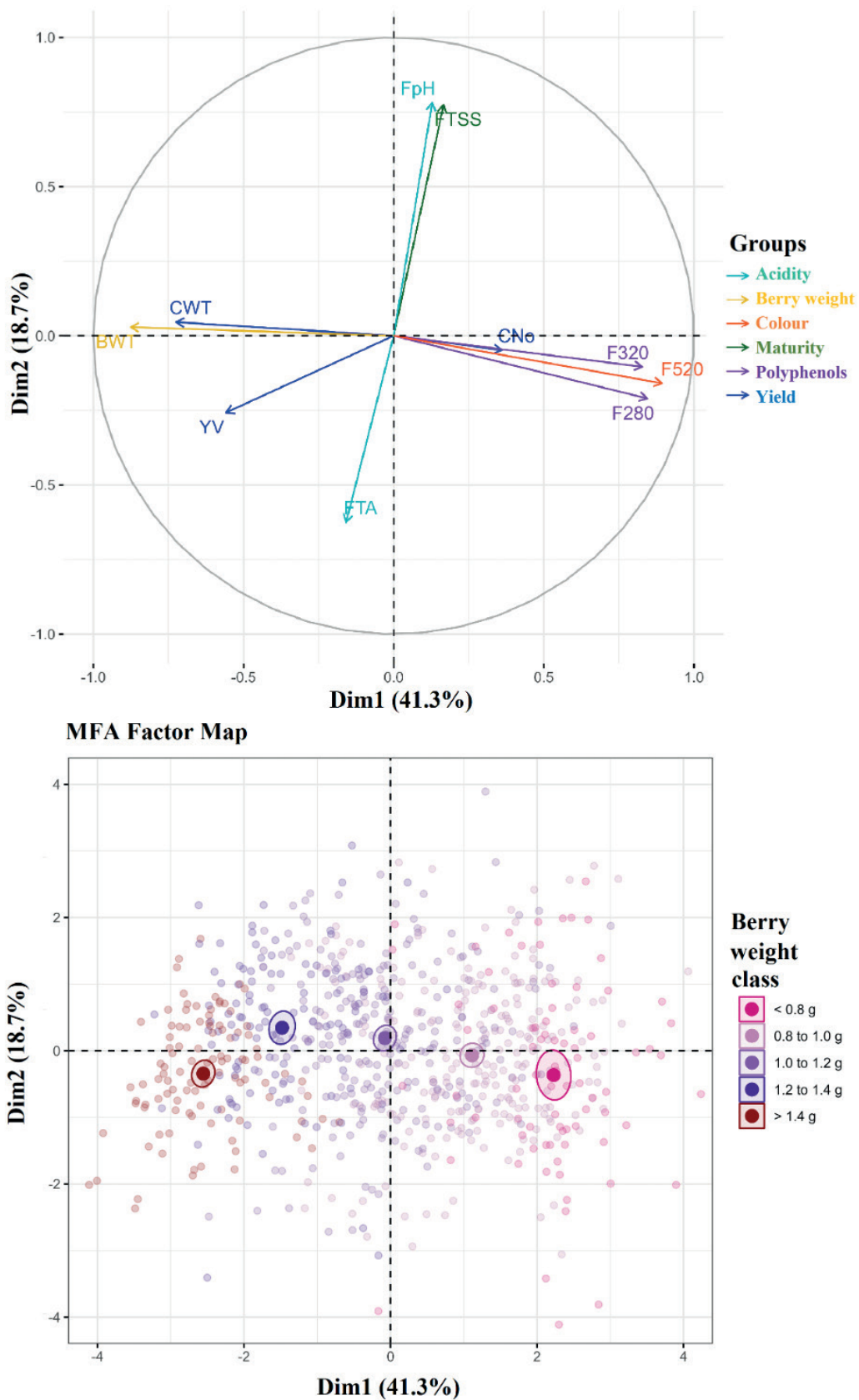
negatively. Berry OD520 (red colour), OD320 and OD280 (total phenolics) vectors were positively loaded on Dim1, indicating higher amounts in smaller berries. There was almost no movement of the clusters in the direction of the Dim2 axis when Berry Weight class decreased, suggesting that highly loaded Dim2 variables (TA, TSS and pH) were not strongly affected by berry weight.

## 1.1. Yield component analysis for all Years

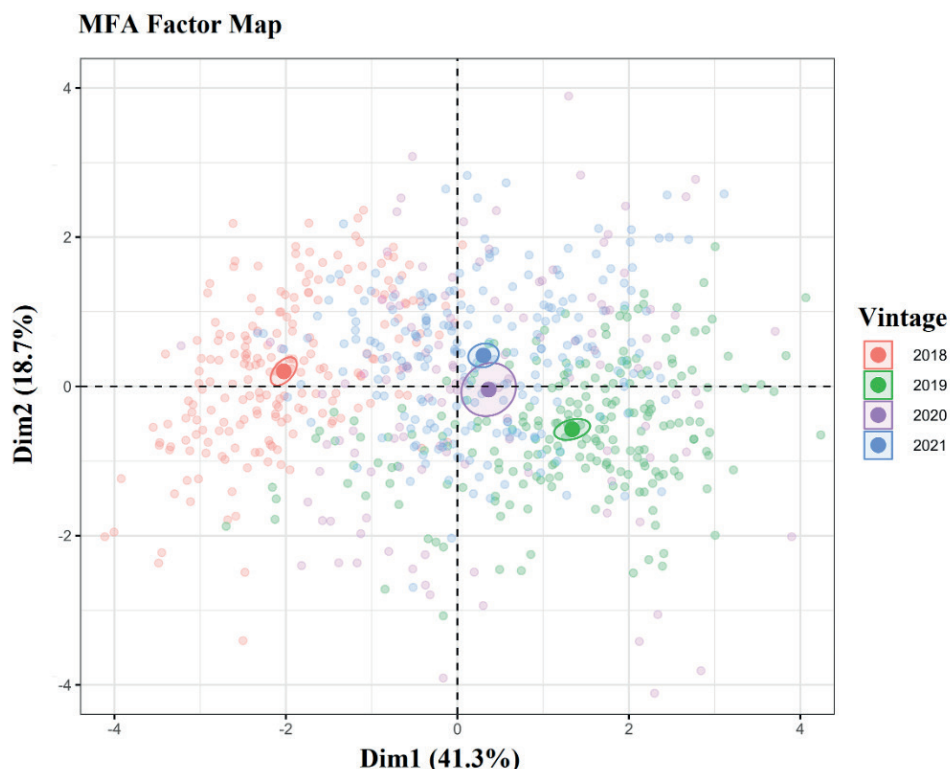
Yield was, unsurprisingly, highly correlated to bunch number and in turn bunch number was highly correlated to shoot number, irrespective of whether values were expressed in yield per vine (data not shown) or yield per linear metre of row (Figure 1). These results are consistent with those obtained in other New Zealand studies of manually pruned vineyards (Grevén *et al.*, 2014). Note that shoot number data were not able to be collected in 2020, thus no 2020 yield information has been included in Figure 1.

A Vintage classification effectively separated 2018 and 2019 data but 2020 and 2021 were closely spaced and centred in the MFA scores plot (Figure 3).

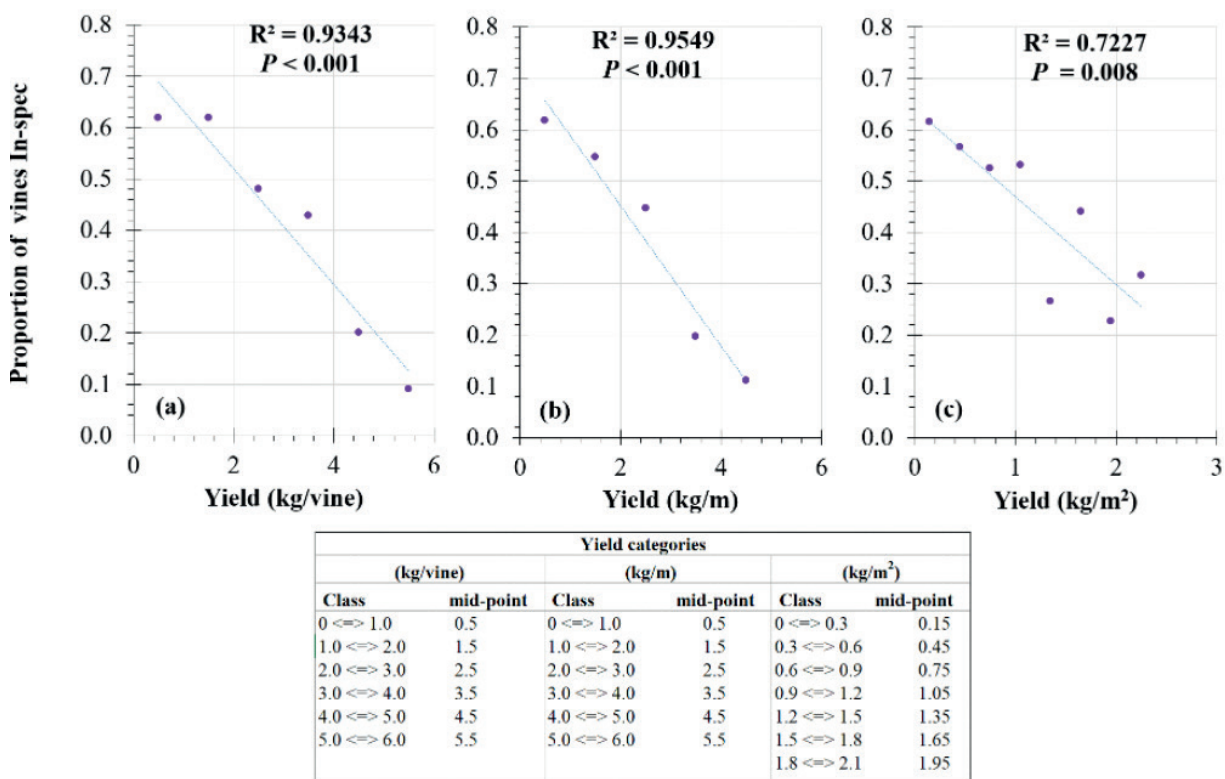




**FIGURE 2.** Multiple Factor Analysis (MFA) of vine and berry parameters of the New Zealand Pinot noir Ideal Vine study network from 2018 to 2021. The upper biplot shows the vector loadings of the measured physiological and chemical parameters and the lower plot shows the scores of vine and berry parameters classified by Berry Weight class. Variable names, groupings and classifications can be found in Supplementary Table S3.



**FIGURE 3.** Multiple Factor Analysis (MFA) of vine and berry parameters of the New Zealand Pinot noir Ideal Vine study network from 2018 to 2021. The plot shows the scores of vine and berry parameters classified by Vintage year.



**FIGURE 4.** Proportion of vines in the New Zealand Pinot noir Ideal Vine study population (N = 530) for which their berry weight, TSS, TA, pH, OD280 and OD520 values were within their corresponding range for at least five out of six parameters “In-Spec” plotted against the mid-point of the Vineyard Yield class expressed as yield per vine (a), yield per linear metre of vine row (b) and yield per unit of land area (c), to take into account differences in planting densities of the vineyards.

## 2. Berry composition benchmarking

From 50 % of the population of vines that directly contributed to the production of “Icon” wines, we derived benchmark specifications for berry weight and key berry composition parameters (Supplementary Table S2). The berry parameters for the remainder of the dataset were in turn classified according to the parameter ranges established for the Icon subset (Martin *et al.*, 2020). The classification was a simple “yes/no” depending on whether the measured parameter was within range or not. Vines were considered to be within an overall specification “In-Spec” if their berry weight and basic berry composition parameter values fell within their corresponding range for at least five out of six parameters.

The frequency of In-Spec vines was then calculated for each Yield class if the vine number was greater than five. Results are presented graphically in Figure 4. Highly significant negative linear relationships were established between vine Yield class and the frequency of In-Spec vines.

### 2.1. Analysis of vine, grape, juice and wine data for vineyard effects

Over the three study vintages (2018, 2019 and 2021) a total of 123 single-vine fruit lots were made into wine using basic yield and fruit maturity parameters as the selection criteria. The aim was to select vines from within the overall

population that displayed a wide range of yields but for which fruit composition was within a narrow TSS range.

One-way ANOVA using Vineyard as the treatment factor and Region as a blocking term was undertaken on 48 vine, berry juice and wine variables measured in the 2018, 2019 and 2021 vintage years. A total of 23 of the variables differed significantly between vineyards (data not presented). The majority of these variables showed a pattern of one or two vineyards differing significantly at either end of the range of values. However, for 18 of the 23 significantly different variables there was an intermediate grouping of 7 to 10 vineyards that were not different from each other. Furthermore, there was no consistent pattern of which vineyards were significantly different. Each vineyard featured multiple times as either being high or low for a particular variable, making interpretation difficult. Further analysis of the dataset was therefore undertaken to identify correlations between the 20 significantly different variables to determine whether a consistent order or rank in mean values between vineyards for two variables could be found. After setting aside expected correlations such as berry TSS with juice TSS, or correlations between berry, juice and wine acidity, only five variables were found to be both significantly different between vineyards using ANOVA and with highly correlated means ( $R^2 > 0.75$ ). These results are summarised in Table 3.

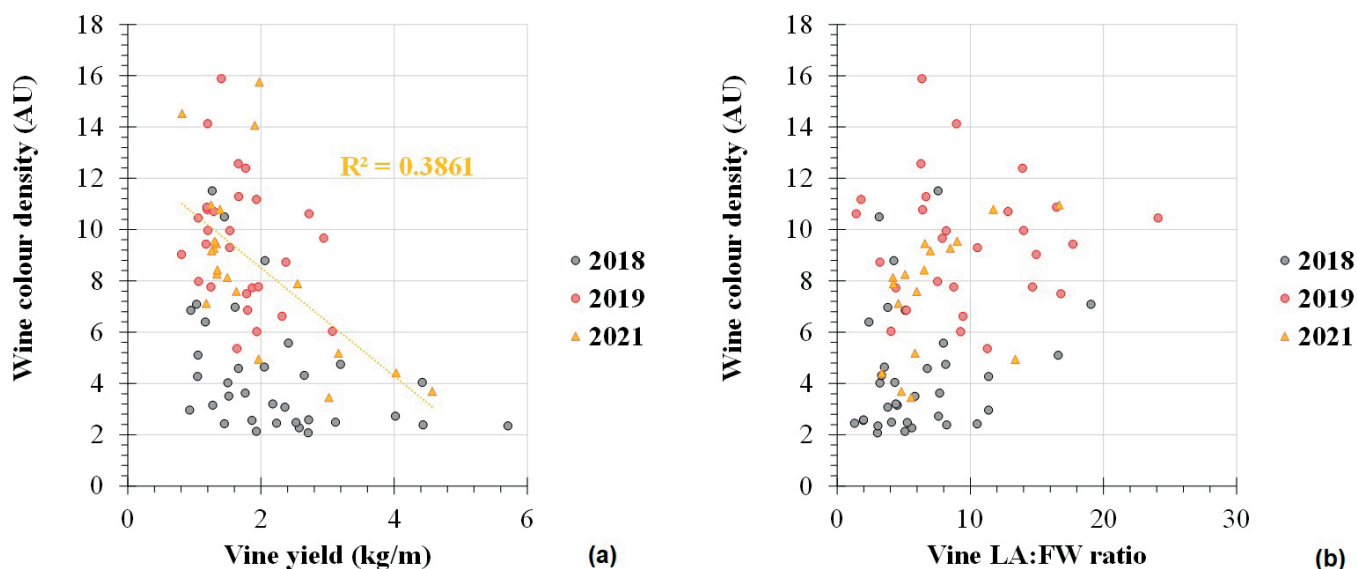
**TABLE 3.** Summary by vineyard of highly correlated and significantly different vine, berry and wine variables across the Ideal Vines study vineyards in 2018, 2019 and 2021. Vineyard details are described in Table 1.

Vineyard ID	Berry weight (g)	Berry OD520 (AU)	Vine Yield (kg/m)	Wine:Marc ratio	Wine Colour Density (AU)	Wine Total Phenolics (mg/L EPG)
MA	1,07 abcde	0,47 abcd	1,36 a	0,35 abcd	8,49 bc	48,83 bcd
MB	1,28 dfg	0,32 a	2,60 c	0,33 ab	5,18 a	42,93 ab
MC	1,13 abcdefg	0,45 abcd	2,06 abc	0,38 bcde	7,26 abc	49,50 bcd
MD	0,90 a	0,60 d	1,37 a	0,44 e	9,55 bc	53,69 bcde
OA	1,11 abcdef	0,49 bcd	1,69 ab	0,36 bcde	6,97 ab	44,52 ab
OB	0,99 ab	0,53 cd	1,55 a	0,41 de	9,42 c	58,14 e
OC	1,04 abc	0,41 abc	1,68 ab	0,40 cde	7,85 bc	48,99 cde
OD	1,32 g	0,33 a	2,68 c	0,30 a	5,48 a	42,18 a
WA	1,07 abcd	0,49 abcd	1,83 ab	0,40 cde	8,77 bc	58,27 de
WB	1,19 cdefg	0,37 ab	2,27 bc	0,34 abc	5,43 a	40,86 ab
WC	1,21 bcdefg	0,44 abcd	1,93 abc	0,37 abcde	7,41 abc	50,63 abc
WD	1,02 abc	0,51 bcd	1,48 a	0,37 bcde	8,93 bc	48,44 bc
Mean LSD	0,22	0,17	0,73	0,07	2,52	9,39
P	0,003	<0.001	<0.001	0,004	<0.001	<0.001
Correlation matrix						
Berry weight (g)		0,79	0,82	0,80	0,82	0,47
Berry OD520			0,76	0,68	0,81	0,52
Vine Yield (kg/m)				0,57	0,81	0,38
Wine:Marc ratio					0,71	0,67
Wine Colour Density (AU)						0,67

Values followed by the same letter down a column are not significantly different.

Abbreviations: AU = Absorbance Units; OD520 = Optical Density @ 520 nm; EPG = Epicatechin gallate; LSD = Least Significant Difference ( $\alpha = 0.05$ ); P = Fisher’s Probability





**FIGURE 5.** Relationships between vine yield per linear metre of canopy (a) and vine leaf area to fruit weight ratio (LA:FW) (b) with wine colour density for the New Zealand Pinot noir Ideal Vine study network in vintages 2018, 2019 and 2021.

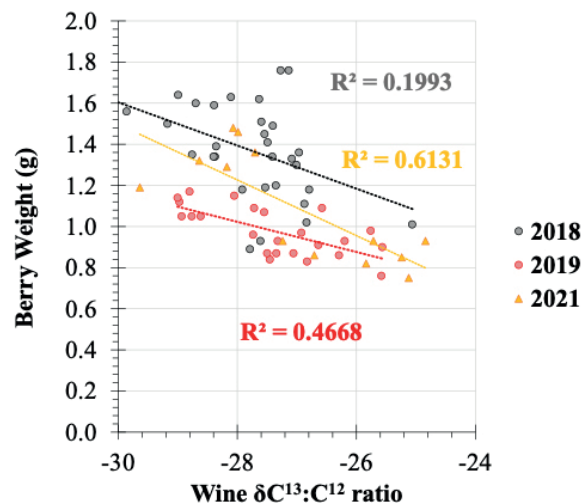
Berry Weight, Yield per metre, Marc:Wine ratio, wine Colour Density and wine Total Phenolics showed relatively consistent ranking of means between vineyards. Vineyard OD had the largest berries, the lowest Marc:Wine ratio, and lower colour and phenolic parameters. Vineyards MD and OB had the smallest berries, the highest Marc:Wine ratios, lower yields, and higher colour and phenolic parameters. These results highlight a key and well-established role for berry size in determining the phenolic content of wines (Abi-Habib *et al.*, 2021; Brillante *et al.*, 2018; Nuzzo and Matthews, 2005; Roby *et al.*, 2004).

Of particular interest in our study was to determine whether relationships existed within the study population between vine performance parameters such as yield (Figure 5a) or LA:FW ratio (Figure 5b) with wine phenolic potential. There was a significant negative correlation between vine yield and wine colour density (Figure 5a) in 2021 ( $R^2 = 0.39$ ;  $P < 0.001$ ) but for the most part the relationships between parameters with a season were either not statistically significant or not meaningful in terms of predicting wine phenolic composition outcomes. A somewhat arbitrary but possibly useful observation, however, was that there were no low-colour wines (colour density  $< 4$  AU) in the sample population when the LA:FW ratio was above  $11 \text{ cm}^2/\text{g}$  (Figure 5b).

### 3. Berry weight and vine water status

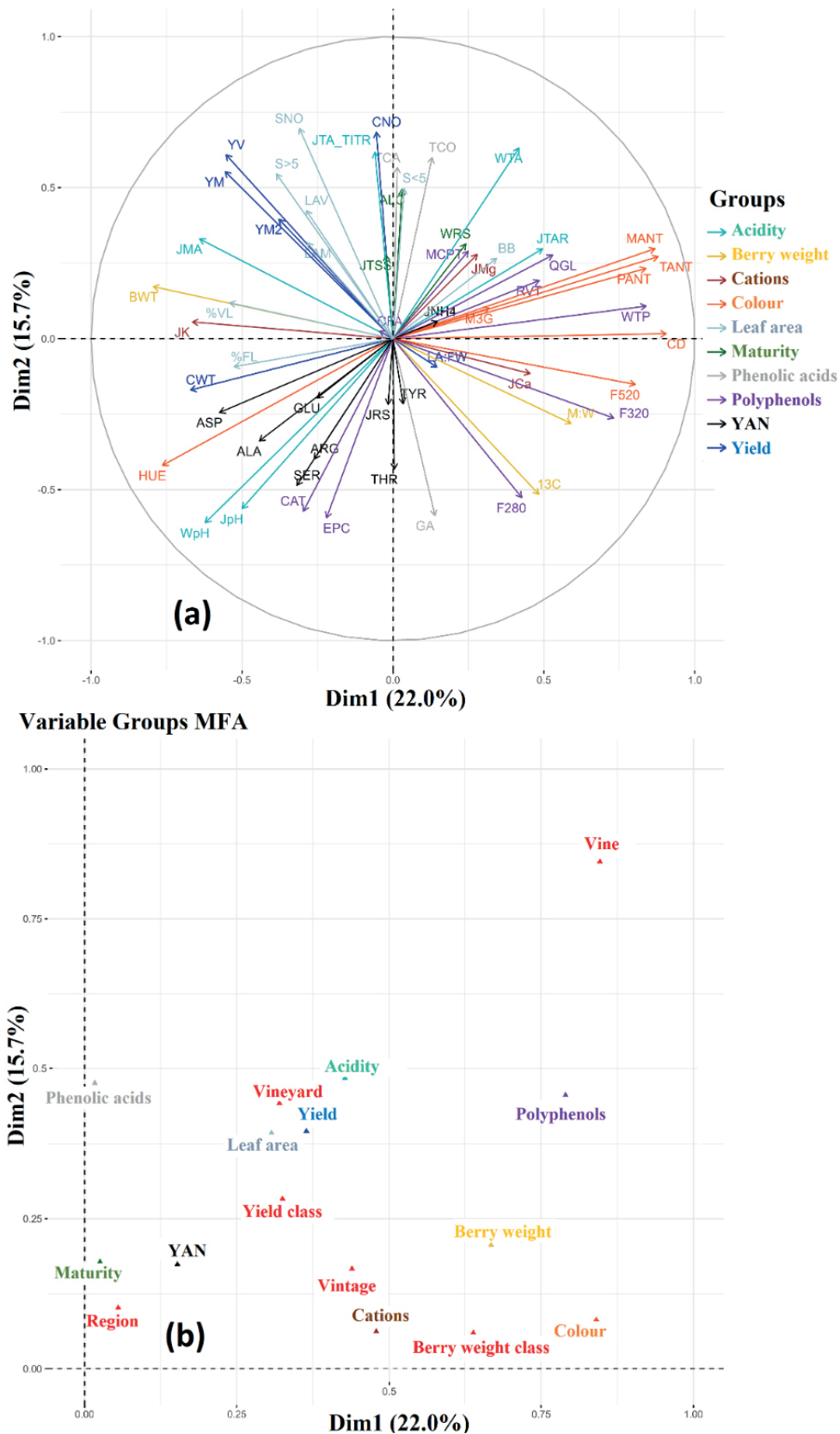
Brillante *et al.* (2020) has shown that for three red varieties in California, a harvest juice  $\delta^{13}\text{C}$  value more negative than  $-27\text{‰}$  corresponds with a mean seasonal stem water potential above  $-1.0$  MPa, maximal stomatal conductance, and minimal water use efficiency indicative of low vine water stress. These results are also consistent with stem water potential values published by Chone *et al.* (2001) for low water stress Cabernet-Sauvignon vines in California and Bordeaux.

Spangenberg and Zufferey (2019) have shown that juice and wine carbon isotope ratios are well correlated to each other and to leaf water potentials for a given variety. In our study we have measured  $\delta^{13}\text{C}$  in wine, and regression plots (Figure 6) show negative linear relationships between  $\delta^{13}\text{C}$  and berry weight that are vintage dependent.

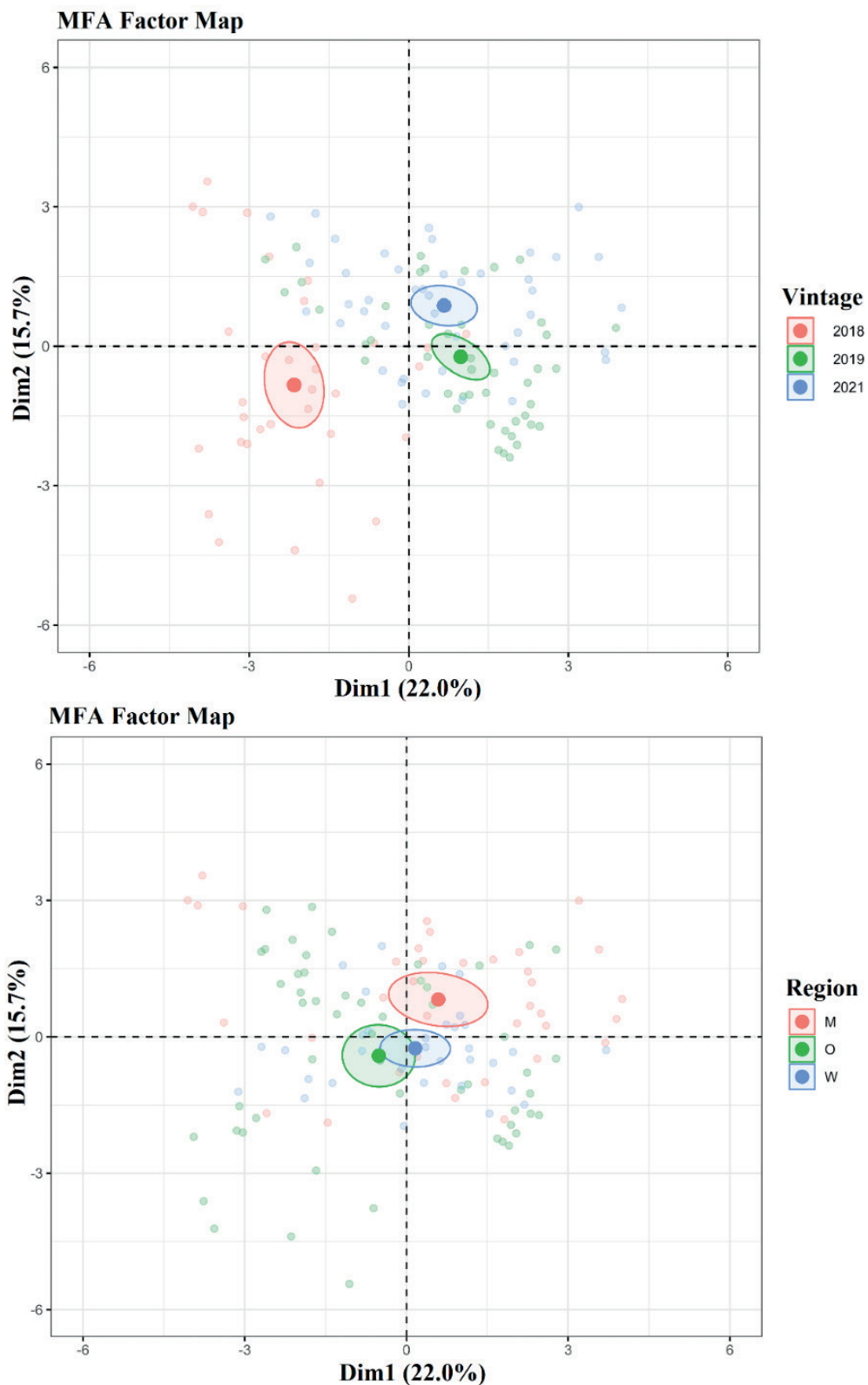


**FIGURE 6.** Relationships between wine carbon isotope discrimination ratio  $\delta^{13}\text{C}:\text{C}^{12}$  and berry weight for the New Zealand Pinot noir Ideal Vine study network in vintages 2018, 2019 and 2021.

The slopes of the regressions are similar between vintages but the berry weight is offset, suggesting that something other than vine water status was having an effect on berry mass. Inter-seasonal differences in seed weight per berry (Friend *et al.*, 2009) are a possible cause, although these data were not available in our study. For a  $\delta^{13}\text{C}$  of  $-28.0\text{‰}$ ,

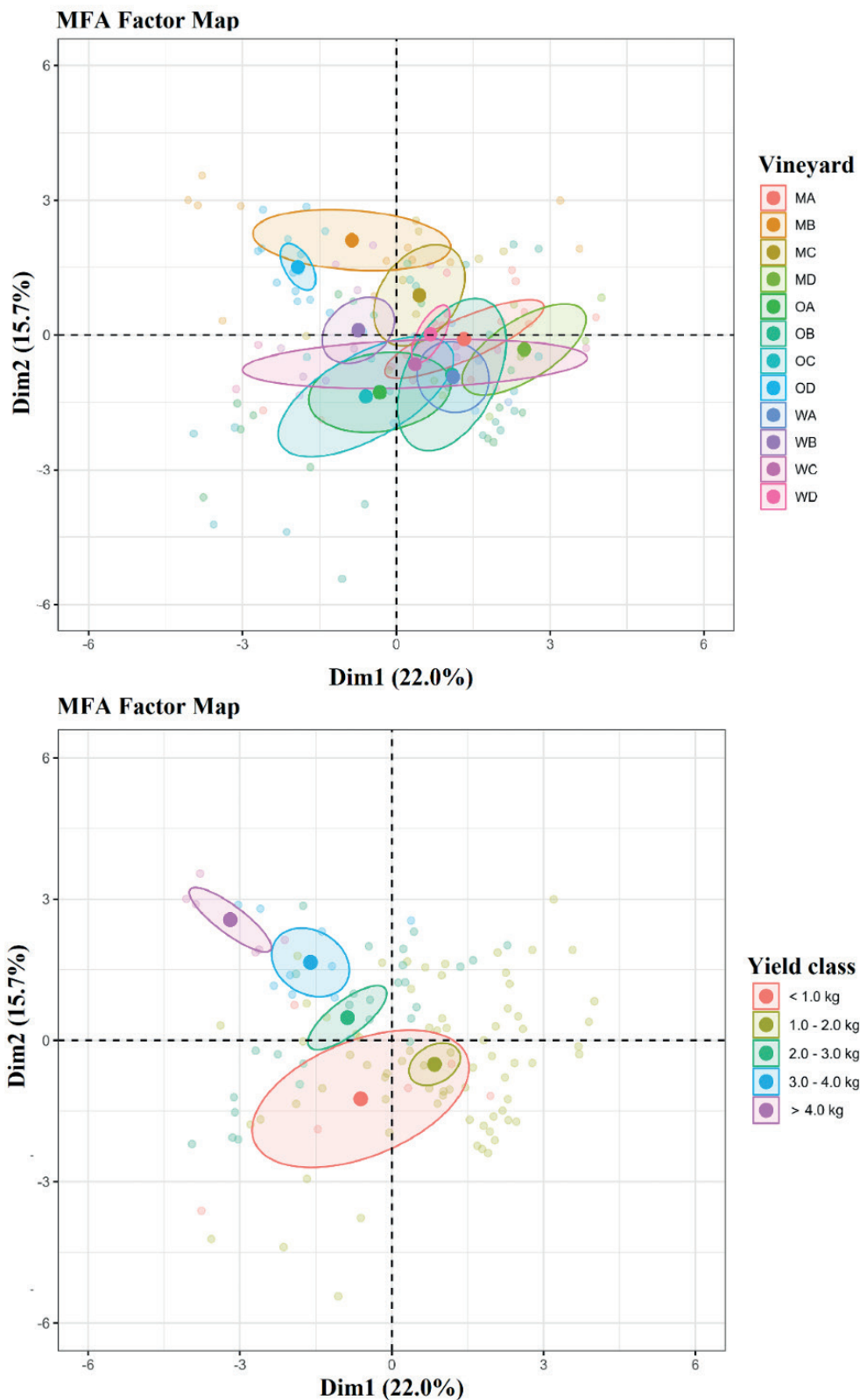


**FIGURE 7.** Multiple Factor Analysis (MFA) of vine, berry, juice and wine parameters of the New Zealand Pinot noir Ideal Vine study network from 2018, 2019 and 2021. The upper biplot shows the vector loadings of the measured physiological and chemical parameters. The lower plot shows the scores of grouped (in non-red colours) and classification variables (in red). Variable names, groupings and classifications can be found in Supplementary Table S3.

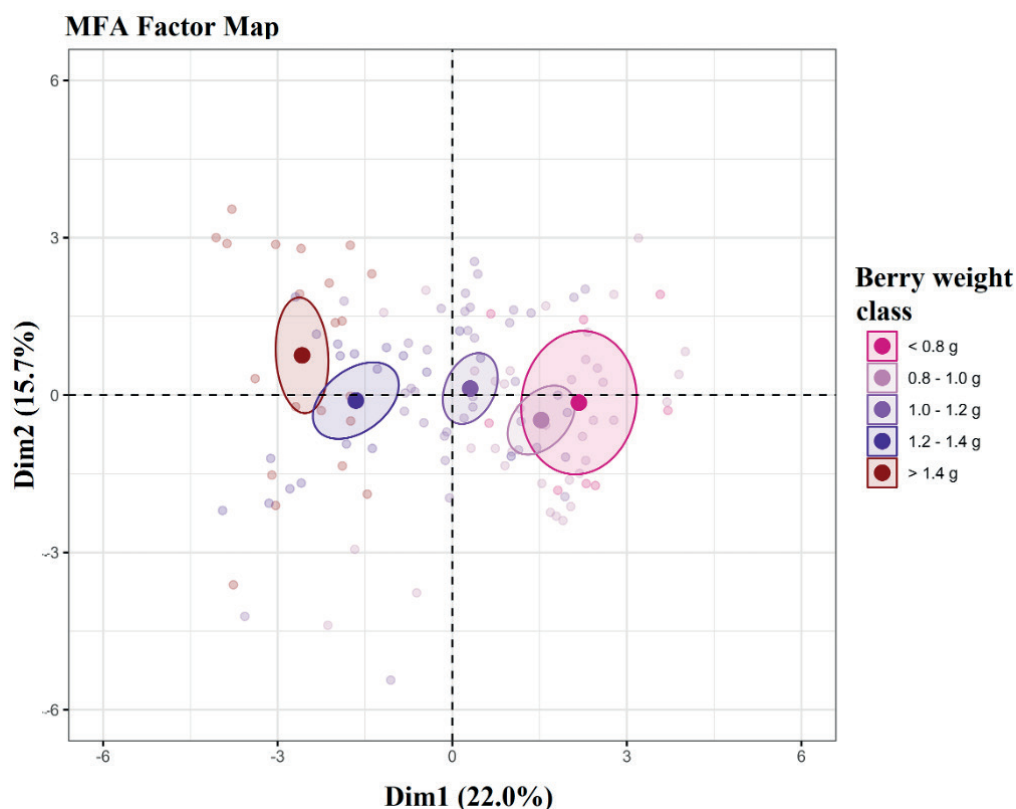


**FIGURE 8.** Multiple Factor Analysis (MFA) of vine, berry, juice and wine parameters of the New Zealand Pinot noir Ideal Vine study network from 2018, 2019 and 2021. The upper plot shows the scores of the measured physiological and chemical parameters classified by Vintage and the lower plot shows the scores of vine and berry parameters classified by Region.





**FIGURE 9.** Multiple Factor Analysis (MFA) of vine, berry, juice and wine parameters of the New Zealand Pinot noir Ideal Vine study network from 2018, 2019 and 2021. The upper scores plot shows the scores of the measured physiological and chemical parameters categorised by Vineyard and the lower plot shows the scores of vine and berry parameters classified by Yield class (kg/m).



**FIGURE 10.** Multiple Factor Analysis (MFA) of vine, berry, juice and wine parameters of the New Zealand Pinot noir Ideal Vine study network from 2018, 2019 and 2021. The plot shows the scores of the measured physiological and chemical parameters categorised by Berry Weight.

the predicted mean berry weight of the overall study population was approximately 1.4 g in 2018, 1.2 g in 2021 and 1.0 g in 2019. The plot also suggests a tipping point at a  $\delta^{13}\text{C}$  above  $-27.0\text{‰}$  where berry weight is reduced as vine water deficit increases, which is consistent with values reported in the literature for viticultural landscapes (Brillante *et al.*, 2020; Guix-Hebrard *et al.*, 2007) and affirm the use of wine measurements to assess vine seasonal water status. Less than 10 % of the wines had a  $\delta^{13}\text{C}$  above  $-26.0\text{‰}$  and the population minimum value was  $-24.8\text{‰}$ , which is entering the moderate to severe water stress range.

#### 4. Exploration of aggregated vine, grape, juice and wine data

All juice and wine compositional data from the 2018, 2019 and 2021 vintages (N = 123) were consolidated, with vine performance and berry composition data. These data included information derived from the grapevine canopy image analysis as well as wine carbon isotope ratio measurements as indicators of vine seasonal water status. Grouped and classified variables (Supplementary Table S3) underwent MFA analysis (Figure 7).

Dim1 loadings, which accounted for 22 % of the variation, were dominated by higher juice amino acids, higher wine pH and wine hue in the negative direction, and by higher berry and wine phenolic indices (OD280, OD320 and OD520) in the positive direction. The vector loadings indicate a partially

antagonistic relationship between wine phenolics and juice primary amino acid metabolisms, which supports other recent findings (Soubeyrand *et al.*, 2014; Soubeyrand *et al.*, 2018; Blank *et al.*, 2022). Negative Dim2 loadings (15.1 %) on Figure 7 showed vectors for vine attributes such as higher shoot and bunch number, malic acid and several phenolic acids. These loadings were opposed by higher  $\delta^{13}\text{C}$ , OD280 and LA:FW.

When wine data were classified by Vintage year (Figure 8a) a similar picture for wine data compared with vine and berry composition data (Figure 3) emerged. The Vintage classification effectively separated 2018 and 2019, while 2021 was centred in the MFA scores plot.

Extension of the clusters towards negative Dim1 scores in the Vintage classification for 2018 indicates larger berries with lower phenolic content and higher hue. The very large GDD deviation from LTA, especially in Otago in 2018 (Figure 1; Supplementary Table S1), is a likely factor. Classification of the MFA scores by Region resulted in centred and partially overlapped clusters (Figure 8b). While it may appear counterintuitive to winemakers that the influence of Region on the measured physiological and chemical parameters was not strong, the MFA analysis indicates that factors such as Vintage, Vineyard or Yield have relatively stronger influences. Wairarapa had a narrower ellipse in both directions of Dim2, indicative of a less variable set of vine, berry, juice and wine parameters, but this is largely an artefact of reduced

Vintage x Vineyard replication (principally because of frost damage) in three of the four vineyards in differing seasons.

No clustering patterns were in evidence when were data were classified by Vineyard (Figure 9a). At first glance it appears that Wairarapa vineyards showed narrower cluster distributions in the direction of Dim2, but as previously discussed, data from Wairarapa spanned less Vintage x Vineyard replication and thus a less variable dataset is consistent.

Yield class did not effectively separate the populations (Figure 9b), which overlapped for classes < 2.0 kg largely because the < 1.0 kg class was highly variable. There was separation of the clustering of the higher Yield classes (> 2.0 kg) in the negative direction of Dim2 and the positive direction of Dim1 scores. Amongst the loadings that characterised these vectors were higher shoot and bunch number per vine.

When data were grouped by Berry Weight class a similar clustering was evident to that observed with MFA of vine and berry data alone (Figure 10). As the Berry Weight class decreased, there was a step-wise and systematic movement of the clusters towards the positive direction of Dim1. This side of the axis is characterised by vectors that are high berry and wine OD520 (red colour), anthocyanins and high wine total phenolics.

## DISCUSSION

### 1. Yields in New Zealand Pinot noir vineyards are highly variable

Vine performance and berry juice and wine composition differed widely between vineyards and vintages. The diversity of seasonal conditions and the range of yield and performance data collected from the same vines across multiple vintages have been instructive. Our results have illustrated the huge vine to vine variation in Pinot noir yield that occurs within, and between, vineyards and across vintages in New Zealand. Vine yield was principally a function of shoot number per vine. Bunch number per shoot also varied considerably but in line with the growers' bunch-thinning practices. These management approaches to crop thinning were the same from vine to vine within a vineyard and season and were comparable for a given vineyard between seasons. While outcomes of thinning practices differentially affect individual vines, our interest to date has been principally in the yield of a given vine at harvest, rather than how that yield had been attained.

Grape yields are strongly affected by seasonal weather conditions, with year-to-year variation exceeding 33 % across an 80-year period in the Czech Republic (Chloupek *et al.*, 2004). Modelling efforts in New Zealand by Zhu *et al.* (2020) identified the importance of maximum air temperatures during the two weeks preceding mid-flowering, both in the year of initiation (affecting bunch number) and in the year of flowering (affecting berry number and bunch mass) in yield formation of Sauvignon blanc. The authors also found that rainfall during the flowering period

negatively influenced berry and bunch mass, while rainfall from fruitset to véraison showed a strong positive effect on the same parameters. At a high level, our data indicate that Pinot noir yield is affected by a similar set of weather and critical period factors, but in-depth analysis of local yield x seasonal weather interactions was beyond our current scope. As we are seeking to identify high-performing individual vines, we have focused our analysis on the relative differences in yield between vines within the same vineyard for the same season (or explored the yield performance relative to those of its neighbours) of the same vine between seasons.

Inspection of the individual vine data from our study population revealed that it was not always the same vines each vintage that performed above or below average. The relationship between Vineyard-normalised vine yield in one vintage and Vineyard-normalised yield from the same vine in any of the other vintages was weak (Martin *et al.*, 2020). Within-vineyard variation in vine yield has been the topic of considerable study especially since the advent of technologies such as on-board yield monitoring, soil EM surveys or aerial/satellite NDVI imaging. In the main, authors have found wide within-vineyard yield variation but spatially stable patterns of relative yield differences between seasons (Bramley and Hamilton, 2004; Kazmierski *et al.*, 2011; Li *et al.*, 2017; Tisseyre and Taylor, 2005). These yield variations are typically well aligned to differences in soil texture or in the conferred canopy density. In our study and on average, 20 % of the monitored vines in a vineyard consistently yielded above the season average while another 20 % were consistently below average yield. These stable spatial patterns are likely to be a function of vine capacity differences that arise from soil, topography or vine health (e.g. grapevine trunk disease) factors.

Only 10 % of vines showed a biennial (alternating high-low or low-high) yield pattern over the 4 years. The remaining 50 % of the vines in each vineyard showed no systematic inter-annual yield pattern, which would appear inconsistent with the previously cited literature which identified stable inter-annual yield patterns. Most authors apply some form of averaging or interpolation to smooth small-scale spatial variation. This process masks vine to vine variation in capacity and variation introduced through uncontrolled factors such as changes to retained bud number, cane breakage or bud damage. Setting an appropriate bud load for a given vine capacity is an established viticultural practice to create a desired ratio of canopy to crop (Ravaz, 1903). The practical realisation of the desired canopy to crop ratio would, however, appear more difficult to achieve. Greven *et al.* (2014) showed that New Zealand Sauvignon blanc yield response to bud loads ranging from 24 to 72 per vine was dynamic over time. In the first year after a change in bud load, yield per shoot did not change with increasing node number, hence the per-vine yield change was proportional to bud load change. In subsequent years the yield per shoot decreased over time, especially with increasing bud number, such that the large first-year yield gains from increased bud loads were progressively lost three years later. The authors



concluded that both node number and the vigour of the canes selected at pruning could be used to set yield potential. New Zealand studies by Bramley *et al.* (2011) and Trought and Bramley (2011) have also shown that spatial soil and canopy density variations do not always directly translate into yield variation, because management factors such as cane selection at pruning may counteract vine to vine differences in capacity.

For many individual vines, inter-annual differences in yield would therefore seem to stem from factors that vary greatly from season to season rather than from more temporally stable attributes (e.g. within-vineyard climate variation, soil physical properties, long-term plant health status, plant genetics). When considered alongside the relationship between shoot number and bunch number variations, inter-annual variation in normalised yield points to differences in bud load as a primary cause of yield variation.

## 2. Lower yields improve the likelihood of achieving an “Icon” berry specification

From the berry quality benchmarking and yield classification process, we have further evidence to support the negative relationship between vine Yield class and the proportion of vines that were within a quality specification based on berry weight and berry TSS, TA, pH, OD280 and OD520 (Martin *et al.*, 2020). Simply put, the lower the yield, the higher the proportion of vines in the vineyard that met the benchmark specification established for “Icon” vines. Our results also support the view that the viticultural practices generally used by New Zealand’s top-end Pinot noir producers, especially with regard to yield management, provide an effective means of ensuring a high proportion of vines meet an arbitrary grape quality specification. Perhaps more importantly however, the results also demonstrate that a considerable proportion of In-Spec vines, that are present in the both “Icon” and “Affordable” vineyards, are also able to meet the same grape quality specification at relatively higher yields. Specifically, of the vines yielding in the range of 1 to 2 kg/m from both the “Icon” and “Affordable” vineyards, 55 % (124/227) met the “Icon” vine benchmark specification. The proportion of In-Spec vines was, however, higher in “Icon” vineyards (75/106 = 71 %) than in the “Affordable” vineyards (49/121 = 40 %) suggesting that site selection and/or vineyard management (i.e. E x M) factors also play a role. The principal drivers for missing specification as yield increased were larger berries and reduced berry OD520. The relationship between these two variables will be discussed further when considering wine composition results. An important finding of this study is that 34 % (103/300) of vines in the “Affordable” vineyard category achieved the “Icon” quality specification without the application of intensive shoot and crop thinning regimes. However, 50 % of “Affordable” vines did not meet an adequate yield target of 2.0 kg/m such that overall only 14 % of vines (41/300) performed optimally. These vines (which were identified in all regions and in all years although not in each region each year) could be considered the outstanding performers or “Ideotype” vines within the study population.

## 3. Grape berry weight is the most influential factor affecting Pinot noir wine composition

The addition of vine performance and berry composition data from 2021 to the dataset from the three preceding vintages changed the hierarchy of influence of classification factors. While Vintage was previously found to be the most effective clustering variable (Martin *et al.*, 2020), Berry Weight became more influential than Vintage with the inclusion of 2021. The 2018 vintage, in which GDD were very high and SWB positive during the ripening period (in all three study regions) appears on the MFA (Figure 3) in an area characterised by larger berries, low berry colour, high hue and reduced phenolic potential. The 2019 vintage was also warm, with above-average GDD, but the SWB was generally negative during the ripening period. This vintage clusters in the positive direction of Dim1 of the scores plot, which is correlated to smaller berries and higher berry colour and phenolics.

Inclusion of juice and wine composition data for a subset of the single-vine population over three vintages (2018, 2019 and 2021) has further strengthened the analysis. A fermenter tube system was used to ferment Pinot noir wines at a scale of 1–2 kg, to accommodate yields of individual vines. Through the narrow fermenter design we have sought to reduce marc/wine surface area and increase the thickness of the cap to slow the rate of phenolic extraction from the marc. We have also sought to slow extraction and minimise oxygen contact in the small volumes by minimising opening times and eliminating plunging operations. This subset of vines made into wine was selected on the basis of a juice TSS of  $22.7 \pm 0.85$  °Brix. Differing from many published prior studies on Pinot noir crop load effects on wine composition, our aim was to mitigate basic berry maturity as a dominant factor (which it clearly is if maturity is insufficient) influencing the vine, berry, juice and wine parameters we measured. Compositional effects on Pinot noir wine are much harder to uncover if vine crop load is within a range that does not limit berry TSS accumulation (Reeve *et al.*, 2016; Reeve *et al.*, 2018; Mawdsley *et al.*, 2018). Our earlier work (Martin *et al.*, 2020) showed that under New Zealand conditions, Pinot noir yield was only rarely a limiting factor in achieving a minimum target berry TSS and typically observed only in cases where high yield was associated with severe vine stress. We consider that our approach aligns well with commercial practice. If berry ripening is slow, a grower will wait longer before harvesting, a luxury that an early-ripening variety such as Pinot noir usually affords in New Zealand. Our results therefore need to be interpreted in the knowledge that the majority of fruit lots made into wine (93 %) achieved a minimum berry sugar ripeness of approximately 22 °Brix.

Additional classifications of Region, Vineyard and Yield were also applied and visualised with MFA scores plots. The effectiveness of the data clustering ranked as: Berry Weight > Vintage > Yield > Region > Vineyard. Inclusion of the juice and wine composition data supported the vine and berry data (Figures 2 and 3) in so far as Berry Weight (which is itself a

yield parameter) accounted for more variation in berry and wine phenolic composition than yield or provenance. Vintage was an effective classification variable with good separation between 2018 and 2019, but 2021 was intermediate and overlapped the earlier Vintages.

Grape berry size is acknowledged as a factor in determining juice composition and final wine quality, particularly in red wines. Sha *et al.* (2018) showed that natural variation in berry size significantly influenced the volatile profiles in Merlot and Cabernet Gernischt grape berries. Mid-sized berries had the greatest aroma compound content, followed by small, with large berries having the least. Walker *et al.* (2005) found Shiraz berries in the smallest mass categories had similar juice composition to larger berries; however, the smallest berries had higher anthocyanin concentration than large berries. Roby *et al.* (2004) also showed the concentration of anthocyanins in the berry was inversely related to berry size. Small-scale wines made from small berries versus large berries showed no differences in wine or sensory properties (Walker *et al.*, 2005). Small berries had a similar skin to fruit ratio as that of large berries; however, the ratio of seed weight to skin weight measured post-fermentation was higher for small berries. Walker *et al.* (2005) concluded that variation in juice and wine composition as a function of berry size showed consistent trends for all seasons, implying that reported instances of improved wine quality from small berries (often associated with pruning treatments or deficit irrigation strategies) were more likely to be due to management effects than to intrinsic developmental differences between large and small berries. Nuzzo and Matthews (2005) also concluded that viticultural practices used to control yield in a vineyard may be more important than the yield or berry size values in determining the quality of the resulting grapes and wines. Two studies on Cabernet-Sauvignon (Roby and Matthews, 2004; Roby *et al.*, 2004), where berries were separated into six berry mass categories, showed that any variation in berry mass had a limited role in determining juice and wine composition. Roby *et al.* (2004) confirmed that the effects of vine water status on fruit composition arise in addition to associated (developmental) differences in fruit size.

In our study the increase in wine colour from 2018 to 2019 (Figure 5) could not be fully attributed to an increase in Marc:Wine ratio as a function of berry weight. Wine colour density was typically 200–300 % higher in 2019 than 2018 whereas the Marc:Wine ratio was only 20–30 % higher in 2019 than 2018. These results indicate that anthocyanin concentrations per unit of berry skin were probably also much higher in 2019 although these were not directly measured in our study. The greatly increased wine colour in 2019 also held true when the same yields were compared between vintages (Figure 5). When considering our study results alongside the literature, they would appear to support the conclusions of Walker *et al.* (2005) that the factors that lead to smaller berries can in parallel lead to higher intrinsic quality potential of the berry in addition to a favourable increase in Marc:Wine ratio.

### 3.1. Berry weight differences were not always related to vine water status

Vine water status in the much drier January to March period of 2019 is a possible driver of the smaller berries and much more highly coloured wines relative to those in 2018. Soil water content has been extensively shown to strongly influence yield, berry size, fruit composition, and overall wine quality (Abi-Habib *et al.*, 2021; Chen *et al.*, 2018; Ferrer *et al.*, 2014; Matthews and Nuzzo, 2007; Ojeda *et al.*, 2002; Roby *et al.*, 2004; Zufferey *et al.*, 2017; Van Leeuwen *et al.*, 2004). Vine water deficits generally lead to smaller berries and changes in fruit and wine composition (Kennedy *et al.*, 2002). Research by Kotsaki *et al.* (2020) investigating grapevine water status in Pinot noir generally showed vine and soil water status as measured by leaf water potential ( $\Psi$ ) and surface soil water content exhibited relationships with yield components (berry weight, cluster number and yield) but not with vine size. Higher leaf  $\Psi$ , and therefore lower water stress in the vines, related directly to larger berry size but without associating directly with higher yields in all cases. Kotaski also found that smaller berries had more anthocyanins, phenols and higher total soluble solids.

The effect of water stress on Pinot noir vines was evaluated by Spangenberg *et al.* (2017) using carbon and nitrogen isotope analysis of wine volatile organic compounds and wine solid residues. Their findings show volatile compounds such as higher alcohols and acetic acid were generally more abundant in wines from mild to severe water-stressed treatments than no or low water stress. This effect was more evident when irrigation restrictions were accompanied by hot and dry summers. Zufferey *et al.* (2017) and Ledderhof *et al.* (2014) reported that a moderate water stress in Pinot noir vines led to more complex and structured Pinot noir wine than those from well-watered vines. However, all the vineyards in our study had drip irrigation installed and very few vines in the overall population displayed visible water deficit symptoms, even in the very dry 2019 year. In addition, the carbon isotope discrimination testing of the wines showed only minor differences in overall seasonal vine water status (Figure 6) between 2018 (mean  $\delta^{13}\text{C} = -27.79\text{‰}$ ) and 2019 (mean  $\delta^{13}\text{C} = -27.25\text{‰}$ ), with both vintage means being below the threshold water deficit value of  $-27.0$  previously discussed. While very short-term vine water deficits that reduced berry weight and increased berry skin colour cannot be completely ruled out, it seems unlikely that the entire study network of 12 vineyards would be concurrently affected in a similar way. Other seasonal environmental factors that can strongly affect berry growth, for example weather conditions during early berry development (Gray and Coombe, 2009; Longbottom *et al.*, 2008), large differences in leaf area during flowering/fruitset (Petrie *et al.*, 2000) or seasonal differences in water flux through the berry (Dai *et al.*, 2010) as a result of differences in bunch zone vapour pressure deficit, present opportunities for more targeted study.

## CONCLUSION

Since 2018 we have studied a network of twelve vineyards in three New Zealand regions in a search to find individual vines that met both yield and quality criteria. Highly significant negative linear relationships were established between vine Yield class and the proportion of vines that met benchmark specifications established for grapes destined for “Icon” wines. The principal drivers for missing specification as yield increased were larger berries and reduced berry colour. Nevertheless 14 % of vines yielding > 2.0 kg/m in the “Affordable” vineyard category achieved the “Icon” quality specification without the application of intensive shoot and crop thinning regimes. These vines were the outstanding performers or “Ideal Vines” within the study population. It was not the same vines each vintage that performed optimally. Inter-annual differences in yield and quality potential at the individual vine level would seem to stem from an interaction between the vine bud load and the vintage conditions.

All vine, berry juice and wine composition variables measured across four seasons underwent MFA using Vintage, Region, Vineyard Yield and Berry Weight as classification variables. Berry Weight followed by Vintage were the most effective clustering variables. As berry weight decreased, there was a proportional increase in Marc:Wine ratio which in turn led to a correlated increase in wine colour density and total phenolics. Indications are, however, that factors that drive formation of smaller berries also lead to an increase in berry quality potential beyond that of the Marc:Wine ratio alone. The effect of berry weight on wine composition is stronger than the previously reported effect of Vintage, which in turn is stronger than Yield, Vineyard or Region effects.

Our work also showed that the majority of vines in the study population do not experience severe water deficits which might otherwise account for differences in berry weight from year to year. Ongoing work is investigating the relative roles of fixed attributes such as vine capacity, and seasonally variable factors such as shoot vigour on berry weight, to develop management options to moderate berry growth while maintaining or increasing yield.

Finally, our results support the view that the viticultural practices generally used by New Zealand’s premier Pinot noir producers, especially with regard to yield management, provide an effective means of ensuring a high proportion of vines meet an arbitrary grape parameter specification. In parallel, however, our observations also support the hypothesis that it is possible to produce Pinot noir wines that fall within an “Icon” benchmark composition range at yields above 1.75 kg/m provided that the mean berry weight is below 1.2 g, juice TSS is above 22°Brix and, ideally, the LA:FW would be above 11 cm<sup>2</sup>/g.

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