THE EFFECT OF CROP LOAD AND EXTENDED RIPENING ON WINE QUALITY AND VINE BALANCE IN VITIS VINIFERA CV. CABERNET SAUVIGNON

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ABSTRACT

Crop load reduction and extended ripening are two practices commonly required for wine grape growers with intention to improve wine quality; however, both cause significant yield loss. Studies on crop load have been conflicting and limited studies exist on extended ripening—warranting further research. The aim of this study was to investigate the interaction of crop load and extended ripening on yield components, wine and fruit composition and to increase understanding of the synchronization of flavor ripeness with sugar ripeness through optimal vine balance. In 2005, 2006 and 2007 a commercial vineyard of clone 8 Cabernet Sauvignon located in Paso Robles, CA was adjusted to four crop levels post fruit set. Each crop level was harvested at five target °Brix levels from 22.5-28.5 °Brix and fermented into wine. Yield components, growth, wine and fruit composition, and wine sensory were measured and assessed on all replicated treatments. A second experiment was conducted in 2006-2007 to investigate the effects of crop load and late season irrigation on extended ripening.

Grapevines exhibited self regulation in growth and yield component compensation. Yield components were reduced from both crop thinning and extended ripening. Pruning weight per vine increased in treatments thinned to lower crop loads in all three seasons, indicating changes in vegetative growth from the crop thinning. Consequently, the light environment within the fruiting zone was effected. Average berry weight, cluster weight and berries per cluster were inversely related to crop load. Extended ripening increased wine color density and anthocyanins each year. Additionally, the lowest crop loads consistently had the lowest color density.

Results from the descriptive analysis characterized the wines, and showed opposing differences between treatments harvested early (22.5-24.0 °Brix) versus those which underwent extended ripening and were harvested at the 27.0-28.5 °Brix target. Consumer acceptability ratings and

expert grading demonstrated that in general, wines from higher °Brix levels in all crop load treatments were preferred. However, the best wines were from treatments with the combination of higher crop load and higher target °Brix at harvest. These results suggest that wine quality can be improved with extended ripening, although significant yield is lost. Additionally, lowest crop load does not always produce highest wine quality. Crop thinning had a detrimental effect on wine quality by disturbing the natural balance of the vine, increasing vegetative growth and negatively affecting the light environment within the fruiting zone. Furthermore, crop thinning did not improve wine quality enough to justify the associated economic losses. Extended ripening proved to be an effective remediation tool for increasing wine quality; however, extended ripening to a target °Brix of 28.5 is not always necessary for well balanced vines. Increased irrigation late in ripening maintained significantly more berry weight and yield relative to the control, and had limited effects on wine quality—although careful monitoring is suggested to avoid wine quality reduction.

STATEMENT

This thesis contains no material which has been previously accepted for an award of any degree or diploma in any University and, to the best of my knowledge and belief, contains no material previously published or written by another person except where due reference is made in the text.

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LIST OF ABBREVIATIONS

Abbreviation	*Explanation and Units*
°	degrees
A	Absorbance
AOV	analysis of variance
BB	bud break
CD	color density—Absorbance (420nm) + Absorbance (520nm)
cl	crop load (clusters/vine)
$CuSO_4$	Copper sulfate
DAA	days after anthesis
DAV	days after veraison
DI	double the standard irrigation
EMS	error mean squares
ETc	evapotranspiration rate of the crop—grapes
ETOH	ethyl alcohol
g	grams
g/L	grams per liter
ha	hectare
HCL	Hydrochloric acid
IBMP	3-isobutyl 2-methoxy pyrazine
Kc	crop coefficient
kg	kilogram
L	liter
LAI	leaf area index
LSD	least significant difference

LWP	leaf water potential (MPa or negative bars)
µL	microliter
mg	milligram
mL	milliliter
mm	millimeter
MPa	megapascal
MOG	material other than grapes
nm	nanometer
N	normality
NaOH	Sodium hydroxide
ng	nanogram
ng/L	nanograms per liter
$NH4^+$	Ammonium
NOPA	alpha amino nitrogen (ppm)
PAR	photosynthetically active radiation (µmol photons (400-700 nm) $m^{-2}s^{-1}$)
PCA	principle component analysis
pH	measurement of acidity
pml	post malolactic fermentation
ppb	parts per billion
ppm	parts per million
ppt	parts per trillion
P value	statistical significance
QDA	quantitative descriptive analysis
r	correlation coefficient
R^2	coefficient of determination

rpm	revolutions per minute
SD	standard irrigation
SO_2	Sulfur dioxide
t	tonne(s)
t/ha	tonnes per hectare
TA	titratable acidity (g/L)
TSS	total soluble solids
TH	thinned
UN	unthinned
VA	volatile acidity
VSP	vertical shoot position
wt	weight
YAN	yeast assimilable nitrogen (ppm)
Y/P	yield to pruning ratio

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Chapter 1. INTRODUCTION AND LITERATURE REVIEW

1.1 Introduction

Farming for desired flavors, wine quality and economic sustainability through vine balance is an ultimate goal of viticulturists. This should be achieved through best management practices for a vineyard site. For as long as grapes have been grown, it has been known that the best wines come from those vineyards where vegetative growth and crop yield are in balance (Dry *et al.* 2004). Vine balance was defined by Gladstones (1992) by stating, "balance is achieved when vegetative vigour and fruit load are in equilibrium and consistent with high fruit quality." Gladstones (1992) also suggests that ripening should be as rapid and complete as possible for any given climate zone; this favors the best compositional balance with desirable flavor characteristics full of ripeness and least loss of aromatics. The term 'wine quality' is widely used by the wine industry at large; however, is best interpreted as the degree to which a set of inherent characteristics fulfils its requirements (International Standards Organization 2011). This definition conveys an appropriate reference for the term quality throughout this thesis.

Delayed harvest date for extended grape ripening or "hang time" has become a recent practice among wineries with the intent of achieving flavor ripeness regardless of sugar content within the berry (Coombe 2001, Grant 2005, Heymann 2006). Tension among sellers and buyers of contracted grapes has escalated from the insistence of extended ripening. In addition, for years growers have been expected to implement the practice of crop load adjustment, causing direct profit loss, based on the assumption that high yielding vineyards will result in poor wine quality (Keller *et al.* 2005). Minimal research or empirical data exists in support of extended ripening (Grant 2005, Heymann 2006) and crop load studies have conflicting results (Bravdo *et al.*1985).

Seemingly, the topic of “vine balance” is not being thoroughly considered or measured (Dry *et al.* 2004) and the efficacy of certain vineyard practices are not being challenged (Smart 2004). The aim of this research was to investigate, scientifically, the role of crop load and extended ripening on: wine quality and composition, vine balance, vine nutrition and reserves, and the cost benefits of each practice. Both sellers and buyers have a vested interest in understanding what will lead to optimal composition (Coombe 2001) and the synchronization of flavor ripeness with sugar ripeness through better vine balance.

The hypotheses tested were: (1) wine quality is improved by crop reduction; lower crop levels will always produce better wine quality (2) extended berry ripening increases wine quality and (3) vineyard economics are negatively affected by extended ripening and crop load adjustment.

1.1.1 Berry Ripening

In all seeded grape cultivars, berry growth and development is initiated by pollination and fertilization (Mullins *et al.* 1992). Berry enlargement begins at fruit set, defined by Mullins *et al.* (1992) as the transformation of flowers into fruit. After fertilization at set, fruit becomes an active importer of photosynthate, tissues become meristematic and berry enlargement begins (Coombe 1973a) following a double sigmoid curve pattern (Winkler 1974, Mullins *et al.* 1992). The berry undergoes three stages noted as stage I, II and III. Stage I is characterized by rapid cell division, cell expansion, organic acid development, and some seed and pericarp development. Final cell number per berry has been determined approximately three weeks post bloom (Coombe 2001). In stage II, also termed Lag Phase (Mullins *et al.* 1992, Coombe 2001), pericarp growth slows while embryo development is rapid and titratable acids reach a maximum

(Mullins *et al.* 1992). Nourishment is provided to the berry by both the xylem and phloem up to lag phase (Coombe 2001).

Stage III starts with berry softening and color change in pigmented varieties. The term veraison is commonly used and refers to the initial loss of chlorophyll from the berry skin and appearance of anthocyanins (Mullins *et al.* 1992) marking the beginning of the ripening phase (Coombe 1992). Hexose sugars accumulate and titratable acidity decreases. Berry growth accelerates, solely from cell expansion, and reaches its maximum during this phase. Increased enzyme activity also occurs. The ripening stage lasts approximately 35-55 days (Mullins *et al.* 1992).

1.1.2 Ripeness

Ripening has been described as the alteration of physiologically mature fruit from an unfavorable to favorable condition with respect to firmness, texture, color, flavor and aroma (Westwood 1993); a process of senescence controlled by hormones (Mullins *et al.* 1992). The term "ripe" or horticultural maturity has been defined as the optimum developmental, chemical and physical condition for the intended enological use of the fruit (Winkler *et al.* 1974, Westwood 1993, Galet 2000, Bisson 2001). Degrees Brix is the measurement of total soluble solids within the berry, expressed by the amount of solute (sugars and all other dissolved solids) per unit of solvent (water) (Coombe 1992). Berry ripeness has an important role in purchase contracts (Smart 2004, Grant 2005, Heymann 2006) traditionally dictated by sugar content measured by degree (°) Brix. This provided an objective index for harvest date decision-making (Grant 2005).

Crop level, seasonal conditions, heat summation and vineyard management, are important factors in influencing time of ripening for a certain grape variety (Winkler 1974). Degrees Brix development has been more closely correlated to days after flowering than with temperature

summation (McCarthy 1999). McCarthy and Coombe (2000) found that the berry reaches its maximum weight around 20-21 °Brix with maximum berry size decreasing at day 91.

1.2 Berry Composition and Accumulation

1.2.1 Primary Metabolites

The term primary metabolites is used to describe sugar and acids; the compounds essential for life processes (Coombe 2001, Moore and Clark 1995). The most prominent compounds in a grape berry are glucose and fructose although the majority is comprised of water. Malic and tartaric acid are present in much lower concentrations (Coombe 2001). Aside from water, glucose and fructose are the most prominent compounds transported into the berry after veraison (Coombe 1975, Mullins *et al.* 1992).

As berries reach maturity, berry growth reaches a maximum and soluble solid accumulation tends to decline (Christensen *et al.* 1995). Reports show limited compounds actually accumulate in berry flesh; glucose, fructose and malate mainly derive from inflow of phloem sap. Many other compounds such as potassium, phenols, inorganic anions and tartrate are concentrated in the skin (Coombe 2001).

1.2.2 Secondary Metabolites

Primary metabolites are defined as a conserved, universal set of compounds that are metabolites of the major metabolic pathways that occur in cells of most organisms. Secondary metabolites are generally defined as compounds which play specific roles in plants or microorganisms; furthermore each is produced by a restricted group of organisms. Secondary metabolites are defined as having a specific function (e.g. aromas) but which is not essential for cell replication.

The term secondary metabolites describes a diverse group of volatile compounds which are the main determinants of aroma and flavor in grape berries (Coombe and McCarthy 1997, Zoecklein *et al.* 2000) and can be sensed through both the nose and mouth (Lawless and Heymann 1998, Pierce and Halpern 1996). Phenolic compounds play a key role in the flavor of red wines, based on a harmonious balance between positive and negative taste characteristics influenced by the concentrations of various molecules that directly contribute to flavor (Riberéau-Gayon *et al.* 2006). Although only present in trace quantities within the plant, even a slight disproportion will greatly affect winemaking and wine sensory attributes (Riberéau-Gayon *et al.* 2006).

The majority of phenolic compounds are located in the grape berry skin. Phenols contribute to wine color, bitterness and astringency. Anthocyanins, flavonols, caffeic acid and condensed tannins are commonly analyzed phenols (Riberéau-Gayon *et al.* 2006). Anthocyanins are important compounds contributing to red pigments in berries and wine, while tannins are substances capable of binding with proteins and contribute to astringency (Riberéau-Gayon *et al.* 2006).

Recently, the study of secondary metabolites as an objective measurement of grape and wine "quality" in addition to the standard measures of sugar and acidity has become an important composition parameter (Bindon 2004). Secondary metabolism involves all the biochemical pathways derived from primary carbon metabolism in plants; these pathways have no direct role in plant growth (Taiz and Zeiger 1998).

1.2.3 Ripening Effects on Flavor Compounds

Most previous research on aroma compounds was focused on volatile monoterpenes that are bound to sugars and form glycosides. These glycosides are increasing throughout the ripening

process but increase more rapidly during the advanced stages of ripening (Hardy 1970, Wilson *et al.* 1984, and Reynolds *et al.* 1994), after sugar increase per berry has declined (Coombe and McCarthy 1997). Non-anthocyanin glycosides appear to follow a similar development pattern (Coombe and McCarthy 1997). Furthermore, sugar accumulation is known to be more rapid with crop reduction (Ough and Nagaoka 1984, Bravdo *et al.* 1985, Reynolds *et al.* 1994, Guidoni *et al.* 2002, Gu and Wample 2006, Ford 2007) and may explain why a balanced crop load could result in a more simultaneous change in both sugar and flavor compounds.

Aroma accumulation within the grape berry may differ significantly from other processes associated with ripening and was separately named ‘engusting’ by Coombe and McCarthy (1997).

In addition to general chemical composition of a ripe berry, understanding grapevine’s physiological aspects which control amounts and timing of these key substances into the berry is crucial. Therefore, studies which examine changes in berry development during different stages of ripening, including solute transport, synthesis and degradation are necessary to further optimize grape quality (Coombe 2001) and to better understand which mechanisms are involved in synthesis of flavor compounds and the control of berry and flavor composition (Coombe and McCarthy 2000).

1.3 Flavor Compounds: Methoxypyrazines

Methoxypyrazines are grape derived flavor compounds that contribute: vegetative, herbaceous, bell pepper and earthy (Allen and Lacey 1998) aromas to wines of certain grape varieties. Methoxypyrazines occur widely in plants (Hashizume and Samuta 1999) and have been identified in wines made from Cabernet Sauvignon, Sauvignon Blanc (Allen *et al.* 1994, Allen

and Lacey 1998), Cabernet Franc and Merlot (Harris *et al.* 1987, Preston *et al.* 2008). High concentrations of methoxypyrazines can dominate wine aroma and be detrimental to wine quality (Allen *et al.* 1998) or suggest an olfactory defect (Roujou de Boubée *et al.* 2002, Preston *et al.* 2008). Three methoxypyrazines contribute to these aromas i.e. 3-isobutyl-2-methoxypyrazine (IBMP), 3-isopropyl-2-methoxypyrazine and 3-*sec*-butyl-2-methoxypyrazine. All three have very low detection thresholds of 1-2 ppt in water, causing even small levels of methoxypyrazine to have a discernible impact in wine flavor (Allen and Lacey 1998).

1.3.1 IBMP

The most abundant of the methoxypyrazines is 3-isobutyl-2-methoxypyrazine (IBMP), typically present in wine at concentrations between 3.6- 56.3 ppt (Allen *et al.* 1994). IBMP is considered the most significant methoxypyrazine from a sensory and wine quality perspective (Wilkinson *et al.* 2006). The sensory threshold of IBMP has been variously reported as 0.05-2.0 ppt in water (Seifert *et al.* 1970, Koseridis *et al.* 1998) and between 10-16 ppt in wine (Maga 1989, Allen *et al.*1991, Kotseridis *et al.* 1998, Roujou de Boubée *et al.* 2000)—however, this may vary between wine style and variety.

The bell pepper aroma in Cabernet Sauvignon (Noble *et al.* 1995, Chapman *et al.* 2004) and Sauvignon Blanc (Allen *et al.* 1991) has been correlated with concentrations of IBMP. High levels of IBMP contribute to unpleasant green and herbaceous character in wines (Allen *et al*. 1991, Bogart and Bisson 2006). Descriptive analysis has shown vegetal characteristics to be negatively correlated with fruity characteristics (Heymann and Noble 1987, Chapman *et al.* 2005, Falcao *et al.* 2007).

IBMP content in finished wine is largely dependent on the concentration at harvest (Bogart and Bisson 2006). Roujou de Boubée *et al.* (2002) reported that IBMP is mainly located in stems, then in skins and seeds—with little in the flesh. However, viticultural and environmental conditions such as temperature during ripening, berry maturity and sunlight strongly influence the methoxypyrazine concentrations in grapes and resulting wine (Allen and Lacey 1998). The IBMP concentration in Cabernet Sauvignon has been reported to increase after fruit set, peak at or just before veraison, then decline towards harvest (Hashizume and Samuta 1999, Roujou de Boubee *et al.* 2002, Sala *et al.* 2004).

Studies on increased light exposure (Morrison and Noble 1990, and Noble *et al.* 1995) and temperature (Roujou de Boubée *et al.* 2000) showed a reduction in both IBMP concentrations and bell pepper aromas in the resulting wines. Hashizume and Samuta (1999) showed IBMP decreased in the presence of light in ripening grapes. Therefore, pre-veraison cluster exposure appears to be critical to reduce IBMP at harvest. Still, the initial accumulation of IBMP (pre-veraison) has been associated with high vigor sites particularly those with high soil water availability (Sala *et al.* 2005). Wilkinson *et al.* (2006) reported that increased 'vegetative', 'herbaceous' and 'green capsicum' aroma ratings correlated to increased IBMP concentrations in wines derived from vines with increased canopy vigor and cluster thinning. It has been suggested that IBMP may be translocated from the leaves to the berries via the xylem (Ryona *et al.* 2008). Therefore, increased vigor (e.g. increased leaf number) may also increase the accumulation of IBMP. However, increased vigor generally leads to more shading on the fruiting zone—consequently vigor and cluster exposure may be confounded (Ryona *et al.* 2008).

1.3.2 Sensory Evaluation and Methoxypyrazines

The use of ‘vegetal’ as a wine descriptor is complex. Aside from methoxypyrazines there are other compounds which contain sulfur and produce related aromas such as asparagus, cooked corn and rubber (Goniak and Noble 1987, Swiegers *et al.* 2005).

Preston *et al.* (2008) studied ‘vegetal’ aromas, in quantifiable sensory terms using both descriptive analysis and experts’ groupings to identify the criteria used by winemakers for the term vegetal in Cabernet Sauvignon wine. The study showed that, trained panelists were able to distinguish specific qualitative differences between the descriptors of vegetal aroma. However, the contrast between vegetal aromas and nonvegetal/fruity aromas were a main criterion in the characterization of Cabernet Sauvignon wines. The study also noted that sensory differences in vegetal perceptions may be due to several factors including, (1) higher concentrations of chemicals contributing to vegetal aromas, (2) absence of compounds that contribute to fruity aromas, or (3) masking of vegetal aromas by fruity aromas when compounds associated with fruity aromas are present. Finally, Wilkinson *et al.* (2006) studied green characters in Cabernet Sauvignon and correlated perceived green character, in wines ranked by an expert panel, with IBMP concentration.

1.4 Transport and Berry Ripening

Both the xylem and phloem, but especially the xylem, are involved in transport of water and other solutes from fruit set to veraison (Creasy *et al.* 1993, Coombe and McCarthy 2000). Malate is the main solute accumulating at this time. From veraison to 18-20 °Brix xylem flow is inhibited; all water movement into the berry is through the phloem. At this time, water and sugar are the main components of the phloem sap (Coombe 1987, Coombe and McCarthy 2000).

During this time—post veraison—potassium accumulates in berry skin cells in addition to anthocyanins which also accumulate in the skins of colored varieties (Creasy *et al.* 1993, Coombe and McCarthy 2000) via the phloem.

At some point during berry development and aging, the vine ceases transport of sugar to the fruit therefore, Shiraz berries appear to undergo engustment processes in isolation from the rest of the vine (Coombe and McCarthy 1997). Once phloem transport within the grapevine has ended, any further increase in sugar level will be from water loss (i.e. berry dehydration), not continued synthesis and/or translocation of sugar (Coombe and McCarthy 2000). This work supports the statement by Bisson (2001), that extended berry ripening does not appear to be a means for further synthesis of beneficial compounds in the fruit. Although improvement in wine due to extended ripening has been associated with the decline of undesirable compounds—rather than further synthesis of beneficial compounds—there are not any published studies which prove that additional biochemical processes exist with extended ripening and contribute to positive flavors. In addition, non anthocyanin glucosides (red-free G-G) have been shown to increase late season in Shiraz and Muscat Gordo (Gholami 1996, Coombe and McCarthy 2000). Further research is necessary to completely understand berry flavor build up and its association with transport (Coombe and McCarthy 2000).

1.5 Extended Berry Ripening

When fruit has passed the optimal point for enological use, it is termed overripe. Overripeness is characterized by tissue breakdown (Winkler *et al.* 1974, Galet 2000), soft texture, loss of firmness (Westwood 1993), berry shrivel and weight loss (Coombe 1975, Hamilton and Coombe

1992, Galet 2000). These responses are associated with the breakdown of bonds between cell walls and water loss (Mullins *et al.* 1992).

Recent trends in harvest date decisions have shifted towards extended berry ripening; commonly termed "hang time" with the intent of achieving flavor ripeness regardless of sugar content within the berry. Historically, concentration of soluble solids measured in °Brix or Baume, followed up by pH and titratable acidity, were the major influences dictating harvest decisions. Widely described criteria exist for these measurements and many are commonly included in purchase contracts (Grant 2005). In recent years, flavor development during ripening has been acknowledged as an important parameter for harvest decisions (Coombe 2001) in conjunction with the market demand for wines with ripe fruit characteristics that concomitantly have higher alcohol content (Grant 2005). This new direction in wine grape processing, requires a longer ripening period in the vineyard.

1.5.1 Problems with Extended Ripening

The extended ripening or "hang time" practice contributes to a significant yield loss for wine grape growers due to berry dehydration (La Rosa and Nielson 1956, Coombe 1975, Hamilton and Coombe 1992, McCarthy 1997, Bisson 2001, Battany 2005, Grant 2005, Heymann 2006). Extended ripening has created an ongoing debate between viticulturists, winemakers and the industry at large, as to quality benefits gained versus profits lost (Grant 2005, Heymann 2006). In addition to a growing tension between sellers and buyers of contracted grapes, extended ripening poses the following problems within the vineyard: increased pest and disease susceptibility, berry weight loss and yield reduction from berry desiccation and a shortened post harvest period (Grant 2005). Furthermore, delayed harvest commonly introduces negative

consequences including stuck or sluggish fermentations and high alcohol wines (Bisson 2001, Smart 2004).

Although regarded as a high standard, inadequate understanding of the later stages of berry ripening and flavor development inhibits winemakers and viticulturists from reaching a decision on optimal maturity and making prudent harvest decisions (Coombe 2001). In addition, many factors more important than harvest timing decisions such as: irrigation, canopy management, vine balance and pruning level (Smart 2004) are being overlooked. Extended ripening is falsely believed to overcome prior defects in vineyard management (Smart 2004). Delayed berry maturity may be a symptom of an unbalanced vine (Dry *et al.* 2004). Furthermore, there has been little research on the effects of extended ripening on wine quality or vineyard economics. This void presents the opportunity for focused research.

1.5.2 Weight Loss from Extended Berry Ripening

Berry weight loss has been studied; however, there is limited research on berry weight loss at °Brix targets above 24.0 and specific to Cabernet Sauvignon. La Rosa and Nielson (1956) showed weight reduction on Grenache grapes of 4.2 % from the time between 24.7 and 27.0 °Brix, and a reduction of 17.8 % between 27.0 and 28.6 °Brix. A study by Battany (2005) on Merlot, Cabernet Sauvignon and Syrah resulted in berry weight decline of 15.7 %, 25.3 %, and 31.8 % respectively over an eight week period. McCarthy and Coombe (1999) concluded that because phloem transport in Shiraz was greatly reduced after maximum berry weight was achieved, weight loss thereafter was due to berry transpiration.

Loss of berry volume and water appears to be the driving force in accumulation of °Brix above 20-21 in Shiraz berries (McCarthy and Coombe 1999). The study by McCarthy (1997), which

analyzed berry shrinkage in Shiraz, equated an overall average loss in berry size of 0.46 % per day (i.e. 4.6 mg per g berry weight per day).

Caroll *et al.* (1978) associated higher levels of soluble solids with increased wine quality. Most likely, the increase was indirectly related to other changes in fruit composition associated with advanced ripeness rather than just sugar increase within the berry (Bindon 2004).

Therefore, results from these studies suggest that other biochemical changes are occurring in the late stages of ripening which have a positive effect on flavor.

1.5.3 Shortened Post Harvest Period

A post harvest period with functional leaves provides potential for larger yields of ripe fruit—the following season—due to the possibility of additional stored carbohydrates (Howell 2001). However, previous research has showed that post harvest periods are not essential for sustained vine health and productivity (Wample and Bary 1992, Petrie *et al.* 2000, Howell 2001). Grant (2005) stated that the absence of post harvest periods prior to leaf fall may limit crop level over time. In addition, Holzapfel *et al.* (2006) demonstrated that extending the length of the post harvest period by early crop removal over two consecutive seasons, increased yield by 48 % in the third season; and implied that adequate post harvest recovery is essential for maintaining high yield productivity. It has been speculated that a shortened post harvest period may be detrimental to vines compromised by pests, diseases and deficiencies (Grant 2005); thus altering photosynthesis and carbon partitioning between organs (Mullins *et al.* 1992). Most previous research in this topic failed to look at ‘over ripeness’. The majority of related studies terminated at sugar levels between 18-25 °Brix (Grant 2001) although Gu *et al.* (2006) reported that “hang time” had a greater influence on yield and fruit composition than on vine health or vigor.

Moreover, further investigation on the impact of extended ripening and shortened post harvest period on vine productivity and nutrition is warranted.

1.5.4 Impact of Extended Ripening on Must and Fruit Nutrition

Premature arrest of fermentation is one of the most challenging problems in wine production (Bisson 1999). Fermentation rate is considerably influenced by amino acid concentration of the must (Kliewer 1968) with nitrogen being the most important nutrient; necessary for correct yeast growth and obtaining overall wine quality (Hernández-Orte *et al.* 1999). A deficiency could result in a slow or stuck fermentation most likely due to the inhibition of protein synthesis for sugar transport (Ough and Kunkee 1968, Busturia and Lagunas 1986, Bisson 1999, Bisson 2001). Peynaud and Lafon-Lafourcade (1961) reported increases in nitrogen forms that are less easily utilized by yeast as grapes progressed in ripening—thus encouraging slower fermentations.

Anthocyanin accumulation and degradation are affected by nitrogen supply in must (Hilbert *et al.* 2003). Degree of fruit maturity and crop level undoubtedly influence concentration of amino acids in grapes (Kliewer 1968). Research by Kliewer (1968) showed arginine, the most dominant amino acid at fruit maturity, to decrease as fruit became ripe to overripe. Again, this study terminated all harvests at sugar levels between 19.2 and 26.2 °Brix, leaving questions on the effects of extended ripening to higher °Brix levels. Due to limited scientific studies on fermentation of fruit above 26 °Brix, amelioration of stuck ferments by way of yeast nutrients remains unknown.

1.6 Crop Load

Crop load refers to the balance of vine capacity to the demand of fruit, carbohydrate (CHO) supply compared to demand (Lakso and Eissenstat 2004) or fruit weight per unit pruning weight or leaf area (Keller *et al.* 2005). Many undesirable responses result when crop load is out of balance.

1.6.1 Measuring Crop Load

Crop load is assessed using many different indices (Dry *et al.* 2004), but commonly using the ratio of crop weight to cane weight. This measurement has shown a stronger correlation between cropping and wine quality (Bravdo *et al.* 1984, Bravdo *et al.* 1985). Pruning brush weight per vine correlated with leaf area also gives an acceptable measurement of overall vine capacity (Bravdo *et al.* 1984) when compared as a ratio to fruit weight per vine.

1.6.2 Crop Load and Vine Balance

The grapevine has an inherent self-regulating mechanism which controls the balance of vegetative and reproductive growth at a particular yield. Yield component compensation dictates that if one yield component is changed, levels of one or more others will change as well (Coombe and Dry 1992). Research by Freeman *et al.* (1979) and Smart *et al.* (1982) clearly showed this phenomenon. The experiment on the interaction of irrigation and pruning level by Freeman *et al.* (1979) indicated that berry weight and bunch weight was significantly affected by both pruning level and irrigation. Additionally, severe pruning increased vine vigor. Smart *et al.* (1982) reported that mean cluster weight, berry weight and sugar accumulation significantly decreased as the number of retained buds increased when studying the interrelations between microclimate and yield expression.

1.6.3 Effects of Crop Load

The work of Jackson and Lombard (1993) reported that low yields lead to higher soluble solids and lower acid levels—therefore resulting in a higher sugar/acid ratio. However, severely over-cropped vineyards are known to delay maturity, causing smaller berries, decreased sugar/acid ratio and decreased color (Winkler 1954). Continued over cropping will reduce vine growth, increase irregular production and cause unbalanced fruit at maturity (Winkler 1954). Theory has suggested that crop level reduction will induce benefits to wine quality by accelerating maturity (Ough 1984). Subsequent research shows adverse effects in wine quality and composition from vineyards at extremely low and/or high crop levels (Kasimatis *et al.* 1977, Cordner 1978, Sinton *et al.* 1978) and that higher crop levels are not always associated with lower quality wine (Weaver *et al.* 1961, Kliewer *et al.* 1983, McCarthy *et al.* 1987, Chapman *et al.* 2004).

Under cropped vines have been reported to cause acid, nitrogen and salt accumulation, generating unbalanced wine flavor composition (Sinton *et al.* 1978). Essential amino acids and total nitrogen were significantly higher in vines with crop reduction compared to vines without (Kliewer and Ough 1970); a result of decreased competition for photosynthate and nitrogenous compounds as crop level was reduced. Consequently, a greater supply is available for the fruit (Kliewer and Ough 1970) which can have negative effects on quality.

1.6.4 Crop Adjustment

Wineries commonly require growers to limit vineyard yields based on assumptions that higher yielding vineyards will decrease wine quality (Keller *et al.* 2005); this has serious financial impacts on growers. Yield per vine, also termed crop level, can be easily adjusted through pruning regime and cluster thinning. Cluster thinning is done by hand or machine (Petrie and

Clingeleffer 2006). Pruning level is regulated by bud number per vine. The use of a sacrificial cane, removed or "sacrificed" at later stages of development is a tool used to manipulate vine balance. Pruning level and cluster thinning are commonly used in both commercial vineyards and research on crop load (Chapman *et al.* 2004). Reducing yield by crop thinning rather than by pruning level gives the added advantage of better estimating final yield by observing the inflorescence size and percent fruit set for that particular season (Petrie and Clingeleffer 2006).

1.6.5 Conflicting Responses of Crop Level Adjustment

Chapman *et al.* (2004) found positive effects in flavor and aroma attributes in wines made from higher yielding vines. As yield increased by 12, 08, 24, 30, 36 and 48 buds/vine at winter pruning, "veggie" characteristics of the wines decreased and fruity characteristics increased.

Bravdo *et al.* (1984) showed significant increases in titratable acidity, must proline and trends toward increased tartaric acid, malic acid and potassium due to crop thinning at the following three levels: (1) unthinned 60-70 clusters/vine, (2) moderately thinned 40 cl/vine, (3) severely thinned 20 cl/vine. Ough and Nagaoka (1984) showed wine aroma intensity was unaffected in Cabernet Sauvignon which was thinned by removing one-third, two-thirds or none (control) of the clusters—two weeks after bloom.

Crop level adjustment can have both direct and indirect effects on fruit quality, yield and canopy growth. Evidence from Bravdo *et al.* (1984) demonstrated that yield components were greatly affected by altering crop level; cluster number, berry size and berry number per cluster increased on moderately and heavily thinned treatments as compared to control vines. Bud dissection analysis confirmed the larger clusters in thinned grapevines through relative size measurement of the anlagen for the forthcoming season. This implies crop level adjustment by crop thinning

affects fruitfulness in subsequent seasons. Whether or not changes due to crop level adjustment are good for quality remains unclear.

1.6.6 Timing of Crop Adjustment

Timing of crop adjustment can have an integral role in affecting changes in the grapevine. Chapman *et al.* (2004) showed few sensory differences in wines made from various cluster thinning treatments; however, concluded that Cabernet Sauvignon aromas and flavors respond to crop manipulation when adjusted early in fruit development. Bravdo *et al.* (1984) showed that crop thinning just after bloom will cause a significant yield reduction only when two thirds or more of the fruit is removed. Jackson and Lombard (1993) reported that crop load adjustment by cluster thinning prior to veraison is appropriate in order to affect maturity. In conflict, Keller *et al.* (2008) implemented the crop thinning treatments at lag phase of berry growth (pre-veraison) and showed no effect on vegetative growth, cluster yield components or advanced fruit maturity on Cabernet Sauvignon.

1.6.7 Importance of Crop Load Research

The relationship between crop load and wine quality has been and continues to be a prominent issue in viticulture research and farming (Chapman *et al.* 2004, Keller *et al.* 2005). Traditional thought for ‘old world’ viticulture and winemaking associates higher quality wines with low yields. Europeans have written this assumption into law requiring low yields of certain appellations (Keller *et al.* 2005). However, scientific reports in this research area have been conflicting (Bravdo *et al.* 1985). The inappropriate use of yield per hectare and berry weight as predictors of vine balance and potential wine quality needs to be addressed and other concepts considered (Dry *et al.* 2004).

The disjointed opinions and results from crop level investigations make it an important topic for continued research. Furthermore, the role between yield and quality is yet to be well defined or documented—limiting the ability to optimize or challenge the concept of increasing yields versus decreasing quality (Lakso and Eissenstat 2004). Evidence regarding the strict yield-quality relationship is limited, inconsistent and mostly concluded from research in cool climates which struggle to ripen crop (Reynolds 1989). Directive studies towards finding balance points of crop level that are harmonious with individual site characteristics rather than subjective levels is necessary to re-adjust modern viticulture practices.

1.7 Sensory Analysis

1.7.1 Definition of Sensory Analysis

Sensory evaluation has been defined as a scientific method used to evoke, measure, analyze and interpret those responses to products as perceived through the senses of sight, smell, touch, taste and hearing (Stone and Sidel 1993). This science comprises techniques used to isolate the sensory properties of foods and provides important information to product developers, food scientists and managers about the sensory characteristics of their products (Lawless and Heymann 1998).

1.7.2 Testing Methods

Descriptive analysis is the most sophisticated tool for sensory science, providing complete sensory descriptions of the products, thus determining which sensory attributes are important for acceptance (Lawless and Heymann 1998). Sensory differences in wines with multiple attributes can be analyzed using descriptive analysis techniques (Noble 1979, De La Presa-Owens and Noble 1995). During the 1970's Quantitative Descriptive Analysis (QDA) was developed, using

data generated from unstructured line scales to describe the intensity of rated attributes (Lawless and Heymann 1998). QDA has many advocates and has been extensively reviewed (Stone *et al.* 1974, Zook and Wessman 1977, Powers 1988, Meilgaard *et al.*1991, Heymann *et al.* 1993, Stone and Sidel 1993).

Discrimination testing determines whether two samples are perceptibly different (Stone and Sidel 1993, Lawless and Heymann 1998) and can be used effectively in wine research for checking differences among fermentation replications (S. Langstaff, 2006 personal communication).

1.7.3 Importance of Sensory Analysis in Viticultural Research

Following vineyard experiments through to sensory analysis of wines is difficult, but necessary if the ultimate goal is to influence wine sensory attributes through vineyard management (Chapman *et al.* 2004, Chapman *et al.* 2005, Heymann 2006). Concluding experiments at fruit chemical analysis is limiting, because very few analyses accurately predict wine sensory properties. This also confines the ability to refute or confirm the validity of wine sensory science (Chapman *et al.* 2005).

The correlation of grape composition with finished wine composition is important, as many flavor and aroma components are present in a precursor or undetectable form. These compounds can be hydrolyzed and more detectable during fermentation and aging (Bisson 2001).

1.8 Conclusion

The aim of the following research was to investigate the interaction of crop load and extended ripening on yield components, wine and fruit composition and to increase understanding of the

synchronization of flavor ripeness with sugar ripeness through optimal vine balance. Previous research on crop load fails to clarify whether or not this practice is justified for the amount of yield/financial loss versus quality gained. In addition, although the practice of extended ripening has gained support by winemakers through anecdotal and speculative evidence, few scientific studies exist which examine ripening past traditional levels i.e. 24-25 °Brix. Furthermore, no studies exist on the interaction of crop load and extended ripening.

Chapter 2: General Materials and Methods

2.1 Experimental Site

Materials and methods described in this chapter were common to most of the data collected on this thesis. Specific materials and methods will be described in the associated chapters.

2.1.1 Vineyard Characteristics

The experimental site was established within a 5.6 ha commercial block of Cabernet Sauvignon, with 3.1 ha of experiment treatments. The site was located at J. Lohr Vineyards in the Paso Robles appellation, within the Central Coast AVA of California (35°42'N latitude, 120°37'W longitude). The climate in this area of the Paso Robles region is semi-arid with average rainfall of 355 mm; the majority of rainfall occurs in the winter months. This area had a mean July temperature of 24.2 °C (75.6 °F) and an average diurnal temperature fluctuation of 22.2 °C (40 °F) during the growing season (J. Lohr weather station Airport Rd.). Soil type is Arbuckle San Ysidro complex 106 (USDA soil mapping 1969) and consists of a sandy loam texture formed primarily from alluvium. The site is at 236 meters (m) elevation and has a 1 % slope across the entire research plot. The vineyard aspect is south west; however, due to its minimal slope is not influential. Three soil pits were dug to six feet deep during year one of the experiment to ensure consistency and water holding capacity of the soil profile throughout the experimental site.

The vineyard was planted in 2001 to Cabernet Sauvignon clone 8 on 1103 Paulsen rootstock in an east-west row orientation. Vines are spaced at 2.1 m between vines by 2.4 m between rows, and trained to a bi lateral cordon. Trellis is as a modified VSP system including 2 sets of wires on the north side, and one wire on the south side of the vine lifted before bloom to allow a partial

south side sprawl. This vineyard block was specifically selected on the basis of vine uniformity and common design features among other vineyards in the region.

2.1.2 Standard Management Practices

The practices described below were the standard management practices used on the research vineyard; these practices were consistent with other commercial vineyard blocks managed by J. Lohr Vineyards and Wines. Pruning was completed in early January each year; consisting of 7-9 two bud spurs/cordon. Post harvest and pre bloom fertigation of CAN-17 (17 % Nitrogen, 8.8 % Calcium) was applied each year to the entire 5.6 ha block at the rate of 46 L/ha. Potassium thiosulfate (KTS) (17 % Potassium, 16 % Sulfur) was also applied in the pre bloom fertigation at the rate of 18.9 L/acre (46.68 L/ha). Fertilizers were sourced from Buttonwillow Warehouse Co. It should be noted that the fertilizer application of CAN-17 and KTS was applied by drip irrigation over 8 hours—equivalent to 30.28 L/vine of irrigation water. Grapevines did not receive any supplemental irrigation prior to the pre-bloom fertigation. Vine rows were banded with grape pomace based compost at the rate of 4.5 tonnes/ha. The block was drip irrigated with two emitters/vine at a flow rate of 1.9 L/hour. Frost protection with impact sprinklers was used when necessary during early stages of budbreak. The vineyard was irrigated between 60-70 % of ETc for the 2005, 2006 and 2007 growing seasons. Well water was treated with sulfuric acid and gypsum to balance water pH. Pest and disease control sprays were applied for prevention of disease outbreaks and consistent with Department of Pesticide Regulation requirements. Vines were shoot thinned by hand to 2-3 shoots/spur between E-L stages 19-21. A permanent sward of fescue grass and natural vegetation grew in row middles and was mowed at bloom each year. The under vine area was kept free of any vegetative growth by pre emergent herbicide

applications of Chateau by Valent in winter and spot spray treatment of Roundup Ultramax by Monstanto in spring.

2.2 Field Experiments

2.2.1 Experiment 1: Effects of Crop Load and Extended Ripening

The main experiment was a 4 x 5 factorial. The first treatment/factor was crop level. The second treatment/factor was target total soluble solids (TSS) measured in the unit degrees Brix (°Brix) at harvest.

The four crop levels by five target °Brix levels provided 20 treatment interactions. Both crop level and target °Brix were randomized within each of the three field replications. Therefore, a random arrangement of both factors—crop level and target °Brix— existed within each of the three field replications. This set up provided a total of 60 individual treatment plots which were harvested in sets of three (i.e. corresponding treatments replicated three times), each year and made into wine.

The vines were adjusted to four different crop levels by cluster thinning post fruit set at E-L stage 31 (Coombe 1995) also referred to as pea size and approximately 20 days after fruit set. Crop levels were 20, 40 and 60 clusters/vine and an unthinned (control). To ensure that crop level was accurately adjusted and within the same phenological stage each year, it was essential to thin whole row sections of the experimental block, rather than mixed portions of the 150 vine rows (i.e. 60 separate thinning plots) used for the experiment. Furthermore, thinning partial rows to different crop levels would have tripled the labor time for thinning—and grossly exceeded the interval of phenological stage E-L 31.

An additional row or section of a row was reserved within each of the three field replications each year, and is referred to as ‘bonus’. The bonus rows were included to ensure that adequate fruit at each crop level and within each field replication was available for bin fermentation. Uneven fruit mass would have substantially altered the subsequent fermentations between treatments and introduced an additional variable. The bonus rows were thinned in the same manner and phenological stage as the ‘treatment’ rows. The bonus rows were only used in limited instances when fruit weight within a treatment plot did not meet the standard weight of 454 kg due to berry dehydration. The number of vines per bonus row were the same as its corresponding treatment i.e. bonus row of 20 cl was 150 vines, bonus row of 40 cl was 75 vines, bonus row of 60 and unthinned was 50 vines.

Cluster thinning was done by hand and carefully managed to ensure accurate bunch counts for the corresponding treatments. The labor crew was instructed to follow standard management practices for thinning, such as: prioritize the basal bunch and thin the second bunch/shoot, thin bunches from weak shoots and/or thin bunches with poor fruit set. In order to achieve enough fruit for bin fermentation each treatment had a different number of total vines/plot. Plot sizes were as follows: vines adjusted to 20 (cl/vine) consisted of 150 vines, 40 (cl/vine) consisted of 75 vines, and 60 (cl/vine) and Unthinned consisted of 50 vines per plot. Specific vines were designated and marked within the total vines/treatment plot. Within each treatment replication plot, three panels consisting of four vines each were used to analyze: yield component measurements, canopy assessment, pruning weights and vine nutrition. Fruit from these vines was included in the total yield/treatment plot for bin fermentation and were added into the total yield weight per treatment plot to give a comparative measurement of average yield/vine. The experiment layout is schematically presented in Figure 2.1.

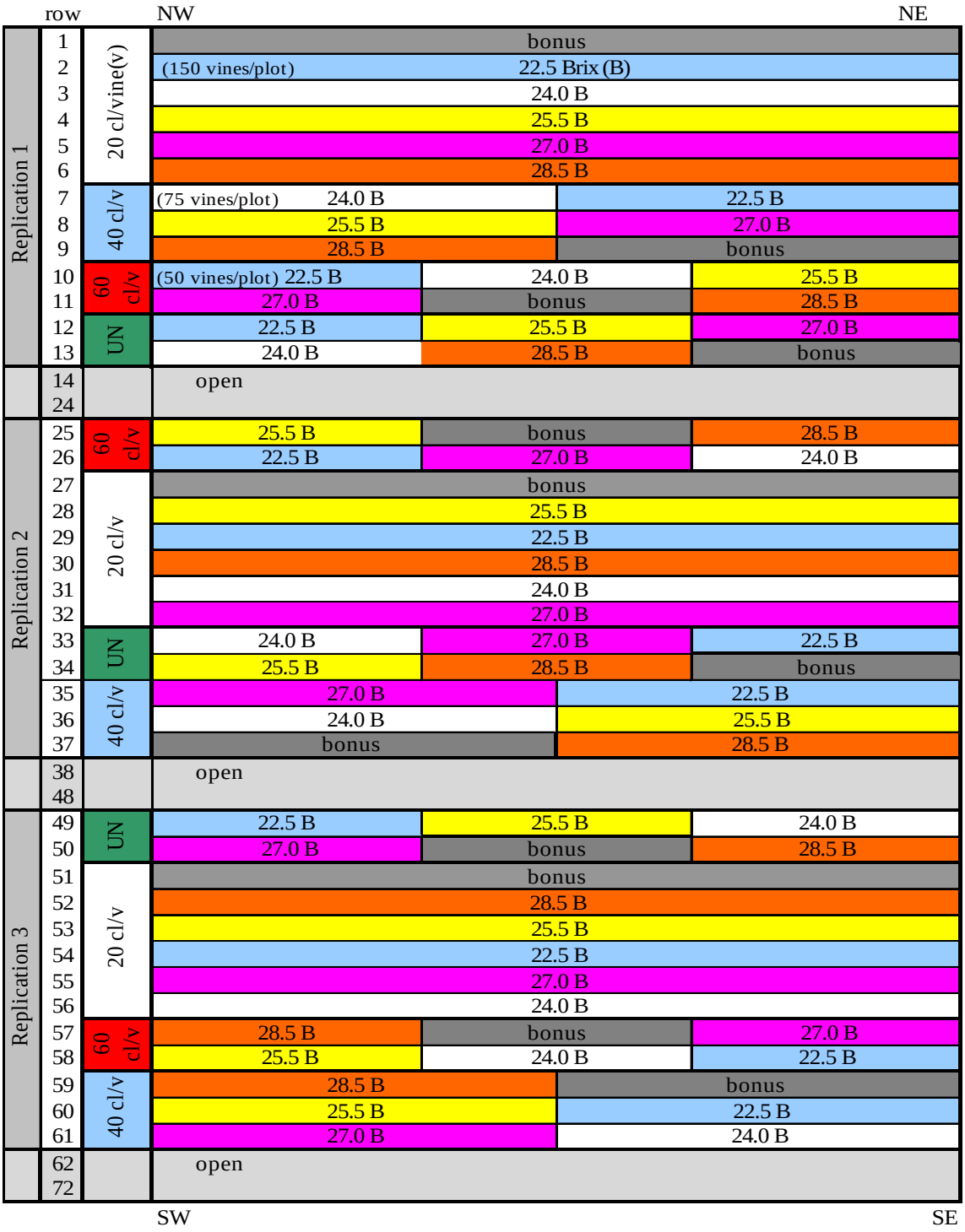


Figure 2.1: Schematic representation of the crop load and ripening treatments across the research vineyard.

The target total soluble solids (TSS) levels were 22.5 ˚Brix , 24.0 ˚Brix, 25.5 ˚Brix, 27.0 ˚Brix and 28.5 ˚Brix. Total soluble solids (TSS) was the measure of ripeness used, and is expressed in the unit (°Brix) —throughout this thesis. In addition, TSS were measured throughout the experiment using an Anton-Paar densitometer calibrated to the unit ˚Brix. The use of °Brix for measuring TSS in grape berries, grape juice and grape must is the industry standard in California. Degrees Brix was monitored by weekly berry sampling in each treatment plot. This data assisted in deciding the harvest date for each treatment and allowed the total cluster number per vine to remain the same between thinning and harvest. Moreover, there was not any whole cluster TSS sampling done on crop adjusted vines. Lastly, the control treatment of both factors was unthinned at 24.0 target °Brix.

There were twenty different harvests per year due to the treatment variables of crop load and target °Brix at harvest. Fruit was hand harvested into plastic MacroBins, fermented, and pressed into neutral barrels. Neutral barrels were barrels previously used for three vintages and thus considered neutral. All barrel characteristics such as cooperage, toast, oak and previous varietal contained in barrels were consistent in all years. A standard winemaking protocol was followed for all bin fermentations. Experiment wine was bottled in late June of each subsequent year. Descriptive analysis was conducted the following July in all three years.

2.2.2 Experiment 2: Crop Load and Late Season Irrigation on Extended Ripening

A second experiment was conducted in 2006 and 2007 in the same general vineyard block as described in 2.2.1 to investigate the effects of crop load and increased irrigation in the late stages of ripening on yield components and fruit and wine composition.

This experiment investigated whether increased irrigation late season mitigates the negative effects of extended ripening. The experiment comprised 8.2 acres and was a complete randomized block design, replicated four times throughout the field. The same standard practices were applied as are stated in 2.1.2. Detailed materials and methods are described in the associated chapter—Chapter 6.

2.3 Phenological Growth and Annual Rainfall

Key phenological dates, growing degree days (GDD) and annual rainfall for the three seasons are presented in Table 2.1.

Table 2.1: Key weather measurements and phenological dates within the 2005, 2006 and 2007 seasons

	weather and phenological dates		
Stage	**2005**	**2006**	**2007**
budbreak	21/3	16/4	28/3
bloom	21/5	2/6	17/5
set (E-L 27)	2/6	8/6	6/6
veraison	10/8	9/8	7/8
# days bloom-veraison	81	68	82
rainfall (mm)	559	385	113
GDD	2690	3408	3447

2.4 Statistical Analysis

ANOVA: Treatments and interactions were analyzed by general two way analysis of variance (ANOVA) using GenStat®10th edition. The treatment structure was crop load x target ˚Brix and the blocking structure by rep. The ANOVA test is widely used in many areas of research and has been used consistently since its introduction by R.A. Fisher in the 1920's to address analyses of agronomic data in agriculture (Palaniswamy and Palaniswamy 2006). The ANOVA tests used in this experiment and derived from the Genstat® program follow the fundamental concept of

ANOVA using the sum of squares for computation of variance, standard deviation, standard error of the mean and the standard error of the difference between the means. The sum of squares (SS) was computed using the following computational formula: $SS = \sum x^2 - (\sum x)^2 / n$

Mean separation was done by least significant difference (LSD) for all significant means and Duncan's multiple range test for means at $p<0.05$. This procedure is referenced in (Palaniswamy and Palaniswamy 2006).

Table(s) set up: The tables in this thesis are set up in the same format as the ANOVA was run: crop load, target °Brix, and crop load x target °Brix (i.e the interaction) for each year and in certain instances a grand mean of all years. More specifically, the means listed for each crop load (e.g. 20 cl) are the average of that specific crop load at each target °Brix level and a total of 15 averaged data points due to the three replications per crop load. Additionally, the means listed for each target °Brix (e.g. 22.5 target °Brix) is the average of that specific target °Brix at each crop level. This provides a total of 12 averaged data points per target °Brix. The interaction is the mean of each specific crop load and target °Brix (e.g. 20 cl x 22.5 target °Brix). The interaction means are composed of three averaged data points (e.g. R1 20/22.5, R2 20/22.5, R3 20/22.5). Although these means were generated in the ANOVA, only their significance is indicated in the tables.

Correlation and linear regression analysis: The correlation analysis was conducted using Microsoft Excel 2007. Correlations among the data sets were initially identified by the correlation coefficients (R values) in correlation matrices generated in Microsoft excel 2007 statistical data package. Subsequently, a simple least squares linear regression was performed to model and analyze the relationship between dependent and independent variables among the data

sets. A linear relationship between two variables x and y can be expressed by the equation: $y = \alpha + \beta x$, (α and β are constants). In the sample data, the equation is: $y = a + bx$ y = dependent variable, x = independent variable, a = the intercept, b = the regression coefficient, also known as the slope of the line indicating the amount of change in y for a unit change in x.

A polynomial form of linear regression (i.e. polynomial regression) was used for some data sets to best represent the relationship between the independent and dependent variables. In many cases the actual equation of the line is presented in the associated figure; however the following represents the general equation of the line used for polynomial regressions: $y = ax^2 + bx + c$

Values for the correlation coefficient (R) were used to calculate the significance of the R^2 value of the regression analyses. e.g. $\sqrt{R^2}$ using standard tables of correlation coefficients at (n-1) 58 degrees of freedom and between 0.05, 0.01 or 0.001 significance. Error bars in graphs represent standard deviation between treatments.

2.5 Hypotheses

The hypotheses tested were: (1) wine quality is improved by crop reduction; (2) extended berry ripening increases wine quality; and (3) vineyard economics are negatively affected by extended ripening and crop load adjustment.

Chapter 3: EFFECTS OF CROP LOAD AND EXTENDED RIPENING ON YIELD COMPONENTS AND VINE GROWTH

3.1 Introduction and Experimental Aims

Crop load adjustment and extended ripening are practices commonly required for California wine grape growers. Both practices are perceived to improve wine composition but have associated problems. In addition, the appropriate definition of ripe fruit is not universally defined and has become subjective. Wine stylistic differences have facilitated varying ripeness targets. Therefore, the timing of harvest and optimal grape ripeness is determined at the point along the ripening continuum which best fits the objective for the wine.

Crop load adjustment contributes to significant financial loss from both overall crop reduction and the cost involved in implementing this practice. Crop load is commonly regulated by pruning to a desired bud number or cluster thinning sometime during the growing season. Both methods of crop load manipulation incur labor costs; although the later is more costly overall. Field workers are paid hourly to hand thin each vine. Any previous vineyard inputs such as fertilizers, irrigation, soil amendments, etc. are wasted or have lost initial purpose when clusters are removed from the vine.

Most studies of yield effects have used cluster thinning (Bravdo *et al.* 1984, Bravdo *et al.* 1985, Reynolds *et al.* 1996), pruning (Freeman *et al.* 1980, Ewart *et al.* 1985, Zamboni *et al.* 1996) or both (Chapman *et al.* 2004) to manipulate crop load. Many of these crop load studies are conflicting and/or inconsistent (Cordner and Ough 1978, Sinton *et al.* 1978, Freeman *et al.* 1980, Bravdo *et al.* 1984, Ough and Nagaoka 1984, Bravdo *et al.* 1985, Ewart *et al.* 1985, Reynolds *et*

al. 1986). Furthermore, these studies are limited and a broader range of crop load investigation is necessary.

In recent years, extended ripening has become a common practice for winemakers to ensure flavor ripeness, especially for the variety Cabernet Sauvignon where vegetative flavors and aromas are commonly present (Allen *et al.* 1994, Noble *et al.* 1995, Allen and Lacey 1998, Chapman *et al.* 2004, Wilkinson *et al.* 2006). Vegetal flavors versus non-vegetal/fruity has been shown to be a key criterion used by winemakers in the classification of Cabernet Sauvignon wines (Preston *et al.* 2008). At high levels, the vegetal/vegetative flavors are considered to lower wine quality (Roujou de Boubée *et al.* 2002, Preston *et al.* 2008). Extended ripening is thought to lessen these flavors in resulting wine and thus is driving the practice in California wine grape production. Extended ripening contributes to significant yield loss due to berry dehydration (La Rosa and Nielson 1956, Coombe 1975, Hamilton and Coombe 1992, McCarthy 1997a, Battany 2005, Bisson 2005, Grant 2005). Although little scientific data exists on this practice, it continues to determine harvest date decisions. The escalating tension among growers and winemakers reinforces the need for this research, with the aim to produce both quantitative and qualitative data that can define the positive and/or negative effects of this practice.

Vine balance plays a decisive role in ripening and final fruit quality, and can be used quantitatively as a predictor of wine style and quality. Historically, it has been noted that the earliest vineyards to ripen were of the highest quality within a region (Dry *et al.* 2004). This contradicts the theory behind the extended ripening practice. The present study puts forth the hypothesis that achieving the best vine balance for a site is the overall goal for predicting and achieving desired wine quality. Additionally, ripening and crop load are strongly linked with vine balance. Although a standard quantitative measure for vine balance and its relation to wine

quality has been debated by viticulturists and researchers in recent years, the ratio between crop weight/vine and dormant vine pruning weight (Y/P) has wide acceptance as an indicator of vine balance. Kliewer and Dokoozlian (2005) reinforced the work of (Bravdo *et al.* 1994, 1995) and showed Y/P ratios of 4-10 and 5-10 on single canopy and divided canopy vines respectively, were well balanced and capable of ripening their crop as well as producing high quality wines.

Excessive shoot vigor and crop load alters the "source-sink" relationship (Ashley 2004) within a grapevine and the increased canopy density and fruit shading can have detrimental effects on fruit quality and yield potential (Dokoozlian and Kliewer 1995). Vegetative vigor can be controlled by crop load. In contrast, over cropping will limit canopy growth and fruit development (Winkler 1954, Jackson and Lombard 1993, Edson *et al.* 1995). Moreover, crop load and vine balance are integral parts of wine grape production.

Nutrition plays a strong role in vine health and the sustainability of a vineyard, both economically and environmentally. Overall vine nutrition is influenced by many factors. The practice of extended ripening has prompted concern among growers regarding its influence on long term degradation of vine nutrition and yield productivity due to a shortened post harvest period. However, Gu *et al.* (2006) reported that "hang time" had a greater influence on yield and fruit composition than on vine health or vigor.

The aims of this study were to:

1. Measure the changes in yield due to crop load, extended ripening and their interaction.
2. Monitor berry development to investigate the effects of crop load and extended ripening on berry weight.

3. Assess the effects of crop load on vine balance through associated canopy measurements and yield components.
4. Evaluate changes in vine nutrition and reserve storage due to crop load and/or extended ripening.

3.2 Materials and Methods

3.2.1 Berry Components

Berry development: Mean berry weight was monitored at weekly intervals, from three weeks after veraison until harvest, to follow changes in berry development among all treatments. The berry collection protocol used in all three seasons is presented in Appendix 1. A 100 berry subsample was taken at random from each collection bag and weighed on a Fisher Scientific accu-2202 scale. The berries were returned to their corresponding bag and crushed by hand for one minute; grape juice was then drained into 200 mL glass beakers to settle. Settled juice was poured into a 50 mL glass beaker and analysis was conducted. Berry composition (i.e. °Brix, pH, and TA) is explained in Chapter 4.

Berry weight at harvest: Berry weights were taken on each treatment (includes the three treatment replications) one hour prior to the harvesting of fruit for fermentation. Berry collection followed the same protocol used for the weekly berry development. One hundred berries were randomly selected from each collection sample and weighed to determine average berry weight. Berry weight was calculated by dividing the total weight for the 100 berry sample by 100. Berry weight data shown is the mean of the three treatment replicates (e.g. mean of R1 20/24.0, R2 20/24.0 and R3 20/24.0).

Berries/cluster: The number of berries/cluster were generated by calculating average harvest berry weight (wt) and cluster weight for each treatment i.e. berries/cluster = average cluster wt (g)/average berry wt (g). The rachis weight per cluster was assumed to be similar among treatments; therefore, yield was not adjusted for rachis weight.

3.2.2 Yield Components

Clusters/vine: Clusters were counted as fruit was harvested from each panel vine and mean ***cluster weight*** (g) calculated as fruit per vine (kg)/clusters/vine.

Yield/vine: Yield per vine (kg) was measured at harvest on the designated panel vines (as explained in 2.2.1) and is referred to as yield/vine *panel*. In addition to the panel vine yield evaluation, a comparative data set was obtained by weighing the total fruit harvested per treatment replication plot (yield/vine *plot*). The fruit was weighed in a Macro Bin on a registered field scale. The yield/vine *plot* was calculated as:

Yield/vine *plot* = (total kg/ treatment ÷ number of vines/treatment).

Tonnes/hectare: Yield, in terms of tonnes (t)/hectare (ha), was calculated from the yield/vine (kg) measurements in both the panel vines (tonnes/ha *panel*) and treatment plots (tonnes/ha *plot*).

Tonnes/ha = (kg/vine x 1922 vines/ha) ÷ 1000 kg/tonne

Second crop: Clusters which grow on the lateral shoots, which grow off the primary shoot, are referred to as second crop (Weaver 1976). The number of second crop clusters and their weight/vine was measured at harvest on the designated panel vines in 2005 and 2007.

3.2.3 Vine Growth Measurements

Mean cane weight: ***Total shoots*** (canes) per vine were counted before pruning and mean cane weight was calculated as: pruning weight (kg)/vine ÷ total canes/vine.

Pruning weights: Pruning was done in early January of each subsequent season and pruning weights were measured to investigate changes in vegetative growth and to calculate the yield/pruning weight ratio **(Y/P)** (fruit.vine^{-1} /pruning.vine^{-1}). The designated panel vines in each treatment were used. Pruned canes from each vine were gathered, tied together and weighed with a Berkley hanging scale.

LAI and PAR: Leaf area index (LAI) and Photosynthetically Active Radiation (PAR) were measured on the vine canopy using an AccuPar LP-80 Ceptometer manufactured by Decagon. Measurements were taken at veraison and harvest, and followed the same protocol—described in Appendix 2.

Percent budbreak: The percent budbreak (% BB) was measured on the designated panel vines in the spring of 2008, after three seasons of treatments to investigate any changes due to crop load and/or extended ripening. The designated panel vines in the 22.5 and 28.5 °Brix targets within each crop load and field replication were used. Percent budbreak was determined using the total count buds per vine and the number of shoots greater than five nodes per vine:

% BB = (shoots ≥ 5 nodes/vine) ÷ (total count nodes/vine) x100

Shoot development: Shoot length was measured on a weekly basis in the spring of 2007 and 2008 to investigate possible changes in the rate of early spring shoot growth due to the previous seasons crop load and/or extended ripening treatments. The #2 positioned vine (second most easterly) of the designated panel vines was used. The first, fifth and last spurs were tagged and

revisited each week for shoot length measurements. The basal shoot was measured unless there was a development problem in which the secondary shoot was then used. Only the 22.5 and 28.5 °Brix levels in each crop load were measured in order to conduct shoot length on the same seven day interval each week. Data collection on all treatments each week was time prohibitive and therefore the two 'extremes' lowest and highest °Brix in each crop load were measured.

3.2.4 Grapevine Nutrition and Shoot Soluble and Insoluble Carbohydrates

Tissue Nutrition: Petiole and blade tissues were collected at bloom of each subsequent season to analyze nutrient status of the vines. Each crop load was tested at the 22.5 and 28.5 °Brix target. Leaves opposite the basal cluster were collected on the north and south side of the vine—blades and petioles were separated. Twenty-five blades and petioles were collected from each of the three treatment replications totaling 75 blades and petioles for laboratory analysis. Analysis was done by California Ag Quest, Fresno, CA.

Carbohydrate Analysis: Carbohydrate reserves were tested in the spring of 2008, after three years of treatments, to test for possible changes in reserve storage due to crop load and/or target °Brix at harvest. Total soluble sugars and starch were measured on each crop load but only at the 22.5 and 28.5 °Brix targets. The first, fifth and last spur were cut at budbreak (E-L stage 3-4) and immediately shipped for analysis. Analysis was done at California State University, Fresno in the Gu lab and followed their standard procedure (Appendix 14).

3.2.5 Statistical Analysis

Harvest components were analyzed by Analysis of Variance (ANOVA), 2-way ANOVA, using GenStat® 10th Edition. There were two factors, crop load and target °Brix and three field replications. Each factor and the interaction of the two factors were analyzed for significant

differences at $p \leq 0.05$ or less. Mean separation was done by LSD for all means at $p < 0.05$, 0.01, and 0.001. Duncan's multiple range test was done only on means at $p \leq 0.05$.

3.3 Results

3.3.1 Berry Components

Berry weight development: Berry development curves for the three seasons are presented in Figure 3.1 a, b, c. Weekly berry weight measurements illustrated an inverse relationship between peak berry weight and crop load in each year. Berry weight decreased as days after veraison (DAV) increased—however the lower crop loads exhibited a steeper decline in berry weight. The number of days required to reach peak berry weight from anthesis (DAA) and veraison are listed in Table 3.1. It should be noted that the overall anthesis date was different each year. Additionally, there were no differences in veraison date between crop loads in any year. On average, 25 DAV were required to reach peak berry weight in the lower crop loads with the 60 cl and UN requiring an average of 32 and 41 DAV, respectively. In 2005 all crop loads generally reached peak berry weight at 34 DAV, although the 20 cl decreased then slightly increased at 41 and 48 DAV, respectively. In 2006 and 2007, the UN consistently required more DAV to reach peak weight, although continued to lose the least amount of berry weight due to extended ripening. The 60 cl and UN averaged lowest in % loss (i.e. 8 % loss) relative to the 20 cl and 40 cl which averaged 16 % and 19 % loss respectively (Table 3.2).

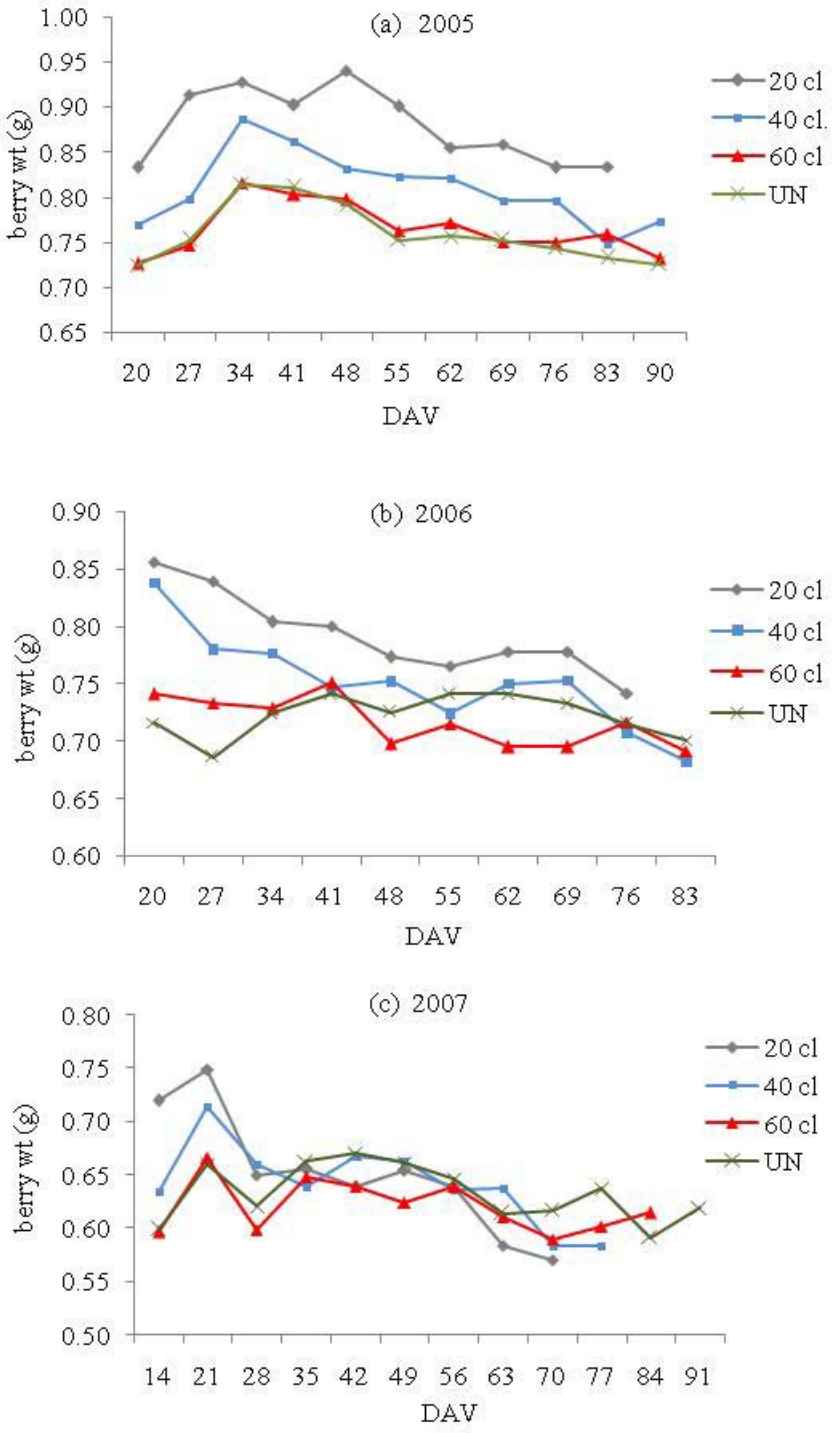


Figure 3.1: Crop load effects on berry weight development during the ripening period in 2005, 2006, and 2007. a, b, c, respectively.

Table 3.1: Days after anthesis (DAA) and veraison (DAV) to reach peak berry weight for three growing seasons.

	DAA to peak berry weight				DAV to peak berry weight			
Crop load	**2005**	**2006**	**2007**	***Average***	**2005**	**2006**	**2007**	***Average***
20	115	88	103	**102**	48	20	21	**30**
40	115	88	103	**102**	34	20	21	**25**
60	115	129	103	**116**	34	41	21	**32**
UN	115	136	145	**132**	34	48	42	**41**
Grand Mean	**115**	**110**	**114**		**38**	**32**	**26**	

Table 3.2: The interaction of crop load and extended ripening on berry weight loss from peak berry weight to final berry weight in each crop load and over three seasons, *based on the berry development curves* .

	% berry weight loss (g)			
	2005	**2006**	**2007**	***Mean 2005-07***
Crop load	**% loss**	**% loss**	**% loss**	**Avg % loss**
20 cl	10	13	24	***16***
40 cl	21	18	18	***19***
60 cl	11	7	7	***8***
UN	11	5	8	***8***
Grand Mean	***13***	***11***	***14***	***13***

Peak weight was between 24.0 and 25.5 °Brix target. Lowest berry weight was at 28.5 target °Brix

Berry weight at harvest: Berry weight was significantly affected by crop load and °Brix at harvest (Table 3.3). An inverse relationship between peak berry weight and crop load occurred in all years. This is indicated by the significant decrease in berry weight as °Brix at harvest increased. The lowest berry weight was consistently found in the 28.5 °Brix target and peak berry weight was generally between the 24.0 and 25.5 °Brix. The average % loss in berry weight from extended ripening regardless of crop load was 11 %, 13 % and 18 % in 2005, 2006 and 2007, respectively. A significant negative relationship was found between berry weight and DAV (Table 3.4), emphasizing that increased time on the vine leads to berry weight loss.

Table 3.3: Effect of crop load and target °Brix at harvest on berry components for three growing seasons.

	berry wt (g) at harvest		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	0.89 d	0.82 c	0.65 c
40	0.84 c	0.73 b	0.60 b
60	0.75 b	0.69 a	0.57 a
UN	0.71 a	0.68 a	0.57 a
	***	***	***
Target °Brix			
22.5	0.80 bc	0.74 bc	0.63 c
24.0	0.83 d	0.77 d	0.62 c
25.5	0.83 cd	0.76 cd	0.62 c
27.0	0.78 b	0.71 b	0.58 b
28.5	0.74 a	0.67 a	0.54 a
	***	***	***
Interaction	**	***	***

means within columns separated by different letters differ significantly at p< 0.001 by LSD

*, **, ***, ns, significant at p< 0.05, 0.01, 0.001, or not significant respectively.

Table 3.4: Coefficient of determination (R^2) and statistical significance level (p value) of the negative linear relationship between berry weight and DAV in three seasons. Significance level is denoted by ***p<0.001

Year	R^2	P value
2005	0.40	***
2006	0.28	***
2007	0.40	***
mean	0.36	***

Table 3.5: The interaction of crop load and extended ripening on berry weight loss from peak berry weight at harvest to the lowest berry weight at harvest i.e. the 28.5 °Brix target in each crop load and over three seasons. Loss is expressed as percent (%) and relative to days after veraison required to reach the 28.5 °Brix target.

	% weight loss berry weight (g) at harvest							
Treatment	**2005**		**2006**		**2007**		***Mean 2005-07***	
Interaction	**% loss**	**DAV**	**% loss**	**DAV**	**% loss**	**DAV**	**% loss**	**DAV**
20 cl	12	81	19	78	28	73	***20***	***77***
40 cl	18	95	17	83	19	83	***18***	***87***
60 cl	12	97	5	89	12	90	***9***	***92***
UN	5	98	9	91	13	93	***9***	***94***
Grand Mean	***11***	***93***	***13***	***85***	***18***	***85***	***14***	***88***

Peak weight was between 24.0 and 25.5 °Brix target. (DAV) is number of days after veraison to reach 28.5 °Brix

Berries/cluster*:* The number of berries/cluster increased due to crop load reduction in 2006 and 2007 (Tables 3.6 and 3.7). The most significant change was in the 20 cl, which increased by 13 % when compared to the control (UN) in both 2006 and 2007. The 2005 data contrasted with this pattern in which berries/cluster were significantly lower in the 20 cl treatment compared with the UN which had the highest number of berries/cluster (Table 3.6). Additionally, the 60 cl had the most berries/cluster when calculated using only peak cluster and berry wt (Table 3.7). Degrees Brix at harvest had no effect on berries/cluster. Statistically, the interaction of crop load and °Brix at harvest showed a significant effect—however, this is likely due to the method for calculating berries/cluster and may not reflect an actual treatment effect.

Table 3.6: Effect of crop load and target °Brix at harvest on berries/cluster for three growing seasons. *Calculated from mean cluster and berry weight.*

	berries/cluster		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	214 a	185 b	174 b
40	226 b	177 b	158 a
60	222 ab	160 a	156 a
UN	239 c	164 a	154 a
	***	***	**
Target °Brix			
22.5	229	172	155
24.0	227	167	161
25.5	215	172	160
27.0	233	173	162
28.5	222	172	165
	ns	ns	ns
Interaction	*	*	*

Means within columns with different letters differ significantly at $p < 0.001$ by LSD. *, **, ***, ns, indicate significance at $p < 0.05$, 0.01, 0.001, or not significant respectively.

Table 3.7: Effect of crop load on berries/cluster for three growing seasons. Berries/cluster are a calculation of peak mean cluster wt (g)/peak berry weight (g).

	berries/cluster *(calculated from peak cluster and berry wt)*			
Crop load	**2005**	**2006**	**2007**	**Grand Mean**
20 cl	223	185	179	**196**
40 cl	232	174	163	**189**
60 cl	237	164	161	**187**
UN	226	158	159	**181**

3.3.2 Yield Components

Clusters/vine: Crop load adjustment had a significant effect on the number of clusters/vine ($p<0.001$) in all three seasons, although °Brix at harvest did not affect clusters/vine (Table 3.8). The UN was significantly higher than other crop loads; however, this difference was not reflected throughout all yield component data. There were no interactions between crop load and °Brix at harvest.

A significant positive linear relationship was found between clusters/vine and DAV. This relationship indicates that as clusters per vine increased, more DAV were required for the fruit to reach the target °Brix at harvest. The coefficient of determination (R^2) and significance levels are presented in Table 3.9. Although there were significant differences at p<0.01 and 0.05, it should be noted that the R^2 values are considerably low.

Table 3.8: Average clusters/vine on crop load and °Brix at harvest treatments for three growing seasons.

	clusters/vine		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	29 a	22 a	24 a
40	50 b	47 b	47 b
60	69 c	62 c	62 c
UN	74 d	66 d	74 d
	***	***	***
Target °Brix			
22.5	55	49	51
24.0	56	48	51
25.5	59	48	52
27.0	54	50	52
28.5	54	51	52
	ns	ns	ns
Interaction	ns	ns	ns

Means within columns with different letters differ significantly at p< 0.001 by LSD. *, **, ***, ns, indicate significance at p< 0.05, 0.01, 0.001, or not significant respectively.

Table 3.9: Coefficient of determination (R^2) and statistical significance level (p value) of the positive linear relationship between clusters/vine and days after veraison to reach target °Brix in each season. Significance level is denoted by **p<0.01, *p<0.05

Year	R^2	P value
2005	0.154	**
2006	0.108	*
2007	0.116	**
mean	0.126	**

Mean cluster weight: Mean cluster weight was significantly affected by both crop load and °Brix at harvest (Table 3.10). Mean cluster weight increased as crop load decreased. The

highest mean cluster weight was consistently in the lowest crop load treatment (20 cl). A significant negative linear relationship was found between clusters/vine and mean cluster weight among all three seasons (Table 3.11). There were no significant differences between mean cluster weight for the 60 cl and UN, even though these treatments had a significantly different number of clusters/vine. Mean cluster weight decreased as °Brix at harvest increased past the 24.0 to 25.5 °Brix target. The most significant differences were between vines harvested at the highest °Brix at harvest (i.e. 28.5 °Brix) and those harvested at lower °Brix.

Table 3.10: Effect of crop load and target °Brix at harvest on mean cluster weight for three growing seasons.

	mean cluster weight (g)		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	191.0 b	149.4 c	111.7 b
40	189.0 b	127.9 b	93.3 a
60	166.3 a	110.7 a	88.8 a
UN	169.0 a	111.2 a	87.8 a
	***	***	***
Target °Brix			
22.5	183.3 bc	126.0 b	97.8 b
24.0	188.5 c	129.8 b	98.4 b
25.5	176.7 b	130.5 b	98.9 b
27.0	181.1 bc	122.8 ab	93.8 b
28.5	164.4 a	115.0 a	88.0 a
	***	***	***
Interaction	ns	ns	ns

Means within columns with different letters differ significantly at $p < 0.001$ by LSD. *, **, ***, ns, indicate significance at $p < 0.05$, 0.01, 0.001, or not significant respectively.

Table 3.11: Coefficient of determination (R^2) and statistical significance level (p value) of the negative linear relationship between mean cluster weight and clusters/vine for three seasons. Significance level is denoted by ***$p<0.001$

Year	R^2	P value
2005	0.36	***
2006	0.74	***
2007	0.49	***
mean	0.53	***

There was a strong negative correlation ($p<0.001$) between DAV and mean cluster weight in all years. The number of DAV and DAA needed to reach peak mean cluster weight varied between the three seasons and between crop loads—within each season (Table 3.12). Generally, all crop loads reached peak cluster weight at or prior to 25.5 °Brix. The average percent (%) weight loss due to extended ripening, between peak and lowest cluster weight, was 14 % (Table 3.13). There were no significant interactions, although the UN had the lowest average % loss in mean cluster weight—including considerably lower losses of 6 % and 9 % in 2005 and 2007, respectively.

Table 3.12: Days after anthesis (DAA) and veraison (DAV) to reach peak mean cluster weight for each crop load for three years.

	DAA to peak mean cluster weight				**DAV to peak mean cluster weight**			
Crop load	**2005**	**2006**	**2007**	***Average***	**2005**	**2006**	**2007**	***Average***
20	121	97	111	***110***	40	29	29	***33***
40	126	120	104	***117***	45	52	22	***40***
60	145	112	116	***124***	64	44	34	***47***
UN	144	136	109	***130***	63	68	37	***56***
Grand mean	***134***	***116***	***110***		***53***	***48***	***31***	

Table 3.13: The interaction of crop load and extended ripening on mean cluster weight loss expressed as % loss within each crop load in three seasons. The percent loss is calculated from peak cluster weight (24.0 or 25.5 °Brix target) to lowest cluster weight at 28.5 °Brix in each crop load and relative to the number of DAV to reach the 28.5 °Brix target.

	% loss cluster wt (g)							
Treatment	**2005**		**2006**		**2007**		***Grand Mean***	
Crop load	**%**	**DAV to 28.5°B**	**%**	**DAV to 28.5°B**	**%**	**DAV to 28.5°B**	**Avg % loss**	**DAV to 28.5°B**
20	18	81	8	78	15	73	***13***	***77***
40	14	95	12	83	19	83	***15***	***87***
60	20	97	13	89	14	90	***16***	***92***
UN	6	98	18	91	9	93	***11***	***94***
Grand Mean	***14***	***93***	***13***	***85***	***14***	***85***	***14***	***88***

Yield/vine panel: The yield per vine from panel sites (yield/vine *panel*) was influenced by both crop load and extended ripening (Table 3.14). There were significant differences for yield/vine *panel* between all crop loads and in each year. On average, adjusting crop load to 20, 40, and 60 clusters per vine reduced yield/vine relative to the UN by 57 %, 26 % and 9 %, respectively. These differences confirm that the different crop loads were successfully implemented each year. The UN had significantly greater yield/vine relative to the other crop loads. Yield decreased as °Brix at harvest increased beyond the 25.5 °Brix target, with the lowest yield consistently at the 28.5 °Brix target—regardless of crop load. Extended ripening to the 28.5 °Brix target reduced overall yield/vine in 2005, 2006 and 2007 by 15 %, 8 % and 10 % respectively, relative to peak yield/vine. There were no significant interactions.

Table 3.14: Effect of crop load and target °Brix at harvest on yield per vine for three growing seasons. *Designated panel vines*

	yield/vine *panel* (kg)		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	5.53 a	3.26 a	2.60 a
40	9.28 b	5.92 b	4.34 b
60	11.55 c	6.82 c	5.53 c
UN	12.38 d	7.27 d	6.51 d
	***	***	***
Target °Brix			
22.5	9.75 b	5.89 b	4.85 b
24.0	10.25 b	5.93 b	4.89 b
25.5	10.15 b	5.97 b	4.88 b
27.0	9.54 b	5.81 ab	4.72 ab
28.5	8.72 a	5.49 a	4.40 a
	**	*	*
Interaction	ns	ns	ns

Means within columns with different letters differ significantly at $p < 0.001$, 0.01 or 0.05 by LSD. *, **, ***, ns, indicate significance at $p < 0.05$, 0.01, 0.001, or not significant respectively.

Yield/vine plot: The yield per vine from each replicated treatment plot (yield/vine *plot*) was greatly affected by crop load in all years (Table 3.15). These data were very similar to the yield/vine *panel* with a subtle difference in the mean separation between the 60 cl and UN in 2005 and 2006. The yield/vine *plot* was done as a check measure against the panel vine data, to ensure that the number of panel sites accurately represented the entire treatment plot. The regression analysis comparing each method of yield evaluation (yield/vine *panel* vs. yield/vine *plot*) had a strong significant positive relationship in all years $p<0.001$ (Table 3.16). Yield/vine *plot* was reduced as °Brix at harvest increased. There was a weak interaction ($p = .051$) in 2006 between crop load and °Brix at harvest.

Table 3.15: Effect of crop load and target °Brix at harvest on yield per vine for three growing seasons. *Treatment plot*

	yield/vine *plot* (kg)		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	5.50 a	3.14 a	2.52 a
40	9.24 b	5.95 b	4.52 b
60	11.94 c	6.97 c	5.59 c
UN	12.26 c	6.98 c	6.52 d
	***	***	***
Target °Brix			
22.5	9.77	5.88 bc	5.02 b
24.0	10.01	5.89 bc	4.97 b
25.5	10.08	5.98 c	4.91 b
27.0	9.59	5.63 ab	4.72 b
28.5	9.24	5.41 a	4.31 a
	ns	**	***
Interaction	ns	*	ns

Means within columns with different letters differ significantly at $p< 0.001$ or 0.01 by LSD. *, **, ***, ns, indicate significance at $p< 0.05$, 0.01, 0.001, or not significant respectively.

Table 3.16: Coefficient of determination (R^2) and statistical significance level (p value) of the positive linear relationship between yield/vine *panel* and yield/vine *plot* in three seasons. Significance level is denoted by ***p<0.001

Year	R^2	P value
2005	0.92	***
2006	0.95	***
2007	0.96	***
mean	0.94	***

Tonnes/hectare: Grapevines which underwent crop load reduction had significantly less yield in terms of t/ha in 2005, 2006 and 2007 (Table 3.17). The 20, 40 and 60 cl averaged 56 %, 25 %, and 9 % less t/ha *panel* respectively, relative to the UN. Moreover, trends in t/ha were consistent with the other yield measurements. Extended ripening reduced yield after the 24-25.5 °Brix target—the most significant loss was at the 28.5 °Brix target. Peak t/ha were consistently between the 24.0 and 25.5 °Brix targets. Extended ripening reduced the overall t/ha *panel* by 13 %, 8 % and 10 % in 2005, 2006 and 2007, respectively.

Table 3.17: Effect of crop load and target °Brix at harvest on tonnes per hectare (t/ha) for three growing seasons. *Designated panel vines*

	tonne/ha *panel*		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	10.98 a	6.28 a	5.01a
40	17.99 b	11.38 b	8.35 b
60	22.21 c	13.11 c	10.62 c
UN	23.80 d	13.97 d	12.51 d
	***	***	***
Target °Brix			
22.5	18.75 b	11.33 b	9.32 b
24.0	19.39 b	11.40 b	9.39 b
25.5	19.50 b	11.47 b	9.37 b
27.0	19.19 b	11.16 ab	9.08 ba
28.5	16.90 a	10.56 a	8.45 a
	**	*	*
Interaction	ns	ns	ns

Means within columns with different letters differ significantly by LSD and Duncan's multiple range test. *, **, ***, ns, indicate significance at p< 0.05, 0.01, 0.001, or not significant respectively.

Second crop: Second crop was measured on the designated panel vines in 2005 and 2007 (Table 3.18). Crop load had a significant effect on second crop incidence and kg/vine in 2007. There was an inverse relationship between primary crop load and second crop. The weight of second crop (kg/vine) decreased as °Brix at harvest increased in 2007.

Table 3.18: Effect of crop load and target °Brix at harvest on second crop incidence and kg/vine for two growing seasons.

	number of second crop clusters/vine		second crop kg/vine	
Treatment	**2005**	**2007**	**2005**	**2007**
Crop load				
20	28	17 a	0.49	0.23 a
40	24	13 b	0.42	0.16 ab
60	28	12 b	0.46	0.12 bc
UN	27	10 b	0.43	0.08 c
	ns	**	ns	**
Target °Brix				
22.5	26	14	0.46	0.23 a
24.0	27	14	0.45	0.20 ab
25.5	28	13	0.50	0.16 b
27.0	26	12	0.41	0.07 ab
28.5	26	12	0.43	0.07 c
	ns	ns	ns	***
Interaction	ns	ns	ns	ns

Means with columns separated by different letters differ significantly at $p < 0.001$ or 0.01 by LSD. *, **, ***, ns, indicate significance at $p < 0.05$, 0.01, 0.001, or not significant respectively.

3.3.3 Vine Growth Measurements

Shoots/vine: Shoots per vine were similar among treatments and years (Table 3.19). There were no significant differences between shoots/vine due to crop load or °Brix at harvest—consequently there were no interactions. The overall average shoots/vine was 41.

Table 3.19: Effect of crop load and target °Brix at harvest on the number of shoots/vine for three growing seasons.

	shoots/vine		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	41.0	41.0	41.0
40	43.0	42.0	41.0
60	41.0	40.0	39.0
UN	43.0	42.0	40.0
	ns	ns	ns
Target °Brix			
22.5	42.0	41.0	40.0
24.0	41.0	41.0	39.0
25.5	43.0	41.0	41.0
27.0	42.0	42.0	40.0
28.5	42.0	42.0	41.0
	ns	ns	ns
Interaction	ns	ns	ns

ns indicates not significant.

Mean shoot weight: Mean shoot weight was affected by crop load and was greatest for the lowest crop load (i.e. 20 cl). The UN had the lowest mean shoot weight in all years measured; however, it was only significantly different from the 40 cl and 60 cl in 2006 (Table 3.20). There were no significant differences in mean shoot weight due to °Brix at harvest and therefore no interactions.

Pruning weights: Pruning weight per vine was affected by crop load reduction in that pruning weight increased significantly ($p<0.001$) for the 20 cl in all years (Table 3.20). Overall, there were few significant changes in the other crop loads. However, in 2006 the 40 cl increased relative to the 60 cl and UN. Degrees Brix at harvest did not affect pruning weights. There were no significant treatment interactions.

Yield/pruning ratio: The yield to pruning weight ratios (Y/P) differed due to crop load and were highly significant between each crop load in each year (Table 3.20). The 20 cl had the lowest Y/P ratios each year, in contrast with the UN which was consistently highest. The Y/P ratios, overall, were different between the three years. The 2006 season had the lowest ratios on average (i.e. 3.0) relative to 6.2 and 5.4 in 2005 and 2007, respectively. There were no significant differences due to °Brix at harvest—hence no treatment interactions occurred.

Table 3.20: Effect of crop load and target °Brix at harvest on canopy growth for three growing seasons.

	mean shoot weight (g)			pruning weights (kg/vine)			Y/P ratio		
Treatment	**2005**	**2006**	**2007**	**2005**	**2006**	**2007**	**2005**	**2006**	**2007**
Crop load									
20	43.71 b	57.71 c	28.33 b	1.83 b	2.38 c	1.15 b	3.1 a	1.4 a	2.3 a
40	36.09 a	47.38 b	22.17 a	1.58 a	2.02 b	0.90 a	6.0 b	2.9 b	4.9 b
60	38.27 a	45.43 ab	21.96 a	1.55 a	1.83 a	0.86 a	7.7 c	3.7 c	6.5 c
UN	35.29 a	43.72 a	20.62 a	1.52 a	1.84 a	0.82 a	8.2 d	4.0 d	7.9 d
	***	***	***	***	***	***	***	***	***
Target °Brix									
22.5	37.53	49.76	23.62	1.58	2.05	1.58	6.4	3.0	5.4
24.0	38.80	49.44	24.53	1.61	2.04	1.61	6.5	3.0	5.5
25.5	38.21	50.22	23.39	1.63	2.08	1.63	6.4	3.0	5.5
27.0	39.70	46.76	22.78	1.67	1.97	1.67	5.9	3.1	5.5
28.5	37.48	46.61	22.05	1.59	1.96	1.59	6.0	3.0	5.2
	ns	ns	ns	ns	ns	ns	ns	ns	ns
Interaction	ns	ns	ns	ns	ns	ns	ns	ns	ns

Means within columns with different letters differ significantly at $p < 0.001$ by LSD.
***, ns, indicate significance at $p < 0.001$, or not significant respectively.

LAI and PAR: Leaf area index (LAI) was measured at veraison (LAI veraison) and harvest (LAI harvest) (Table 3.21). Differences in LAI due to crop load were detected at both phenological stages, but were greater at harvest. The 20 cl had a significantly higher LAI than all other crop loads at both veraison and harvest, and the highest overall mean of 3.6 at veraison.

The LAI (harvest) decreased relative to LAI (veraison) regardless of crop load. LAI (harvest) was affected by °Brix at harvest. In general, LAI (harvest) decreased as target °Brix at harvest increased—significant differences occurred at 27.0 and 28.5 °Brix. There was a significant interaction for LAI (harvest)—however the interaction was not significant for LAI (veraison). The highest overall LAI (harvest) was consistently in the 20 cl and highest overall at treatment 20/22.5. In contrast, the lowest LAI (harvest), was in UN/28.5.

The photosynthetically active radiation (PAR), expressed as % of ambient PAR within the fruiting zone, was measured at veraison and harvest (Table 3.21). Crop load had a significant effect at both phenological stages ($p<0.001$). PAR was lowest in the 20 cl relative to all other crop loads at veraison and harvest. Differences due to °Brix at harvest occurred in PAR (harvest), only. Vines harvested at 28.5 °Brix had the highest PAR (harvest). Overall, PAR increased in all treatments from the veraison to harvest measurement and as °Brix at harvest increased.

Table 3.21: Effect of crop load and target °Brix at harvest on leaf area index (LAI) and canopy light environment (PAR) in the fruiting zone at veraison and harvest in the 2007 season. PAR is the amount of photosynthetically active radiation on the fruiting zone.

Treatment	LAI veraison	LAI harvest	PAR veraison	PAR harvest
Crop load				
20	3.6 a	3.3 a	31.2 a	33.8 a
40	3.1 b	2.7 b	38.5 b	36.8 b
60	3.2 b	2.6 b	35.5 b	38.5 b
UN	3.3 b	2.4 c	35.7 b	39.3 b
	**	***	***	***
Target °Brix				
22.5	3.2	3.2 a	35.8	37.2 a
24.0	3.3	3.0 a	34.8	35.7 a
25.5	3.5	3.1 a	33.6	35.1 a
27.0	3.2	2.5 b	36.3	36.5 a
28.5	3.3	1.9 c	35.7	41.2 b
	ns	***	ns	**
Interaction	ns	**	ns	ns

Means within columns with different letters differ significantly at $p < 0.001$ or $p < 0.01$ by LSD.
, *, ns, indicate significance at $p < 0.01$, 0.001, or not significant respectively.

Percent bud break: Percent bud break (% BB), measured in 2008 after three seasons of treatments had no significant differences due to crop load, °Brix at harvest or their interaction (Table 3.22).

Table 3.22: Effect of crop load and target °Brix at harvest on percent budbreak (% BB) in 2008, after three seasons of treatment, calculated from the number of shoots per vine greater than five nodes and total buds per vine.

Treatment	count buds/vine	shoots ≥ 5 nodes	% BB
Crop load			
20	150.0	73.0	49
40	149.0	87.0	59
60	155.0	88.0	57
UN	150.0	87.0	58
	ns	ns	ns
Target °Brix			
22.5	151.0	89.0	59
28.5	151.0	79.0	53
	ns	ns	ns
Interaction	ns	ns	ns

ns indicates not significant.

Shoot development: Shoot development was measured, weekly, in spring of 2007 and 2008—following the previous seasons' treatments (Figure 3.2 and 3.3). There were few differences. Crop load from the previous seasons only caused differences in shoot growth for the later measurements. However, differences were only significant for the 25-May-07 measurement, and included a small difference between the two lower crop loads (i.e. 20 cl and 40 cl) and the UN. The most noticeable difference was at the final measurement on 6-Jun-08, however it was not significant.

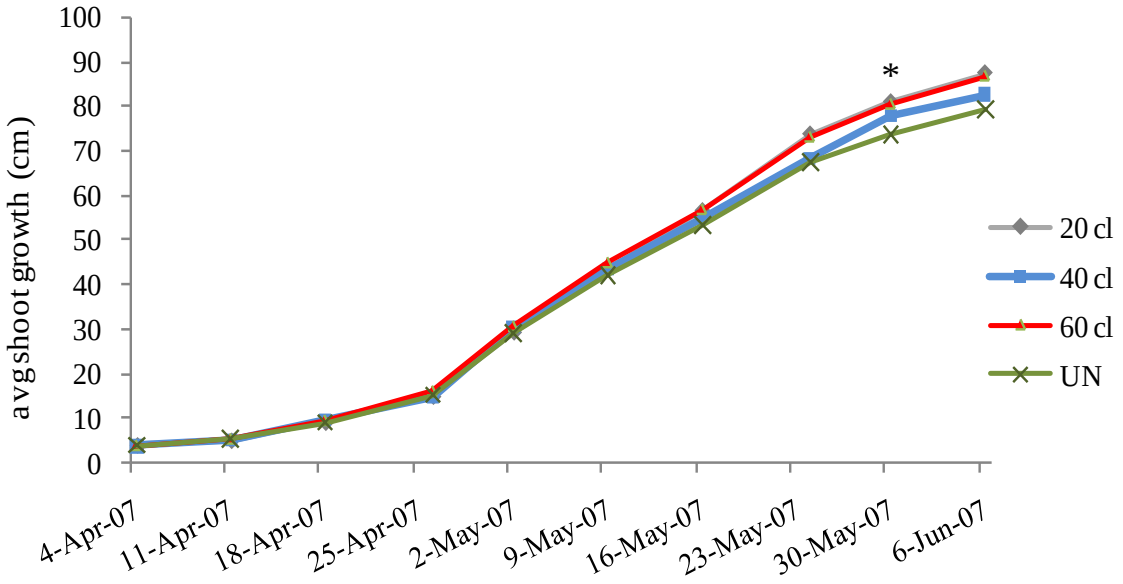


Figure 3.2: Treatment effects on weekly shoot growth in spring 2007 after two years of crop load and extended ripening treatments. * indicates that data was significantly different at $p<0.05$ or otherwise not significant.

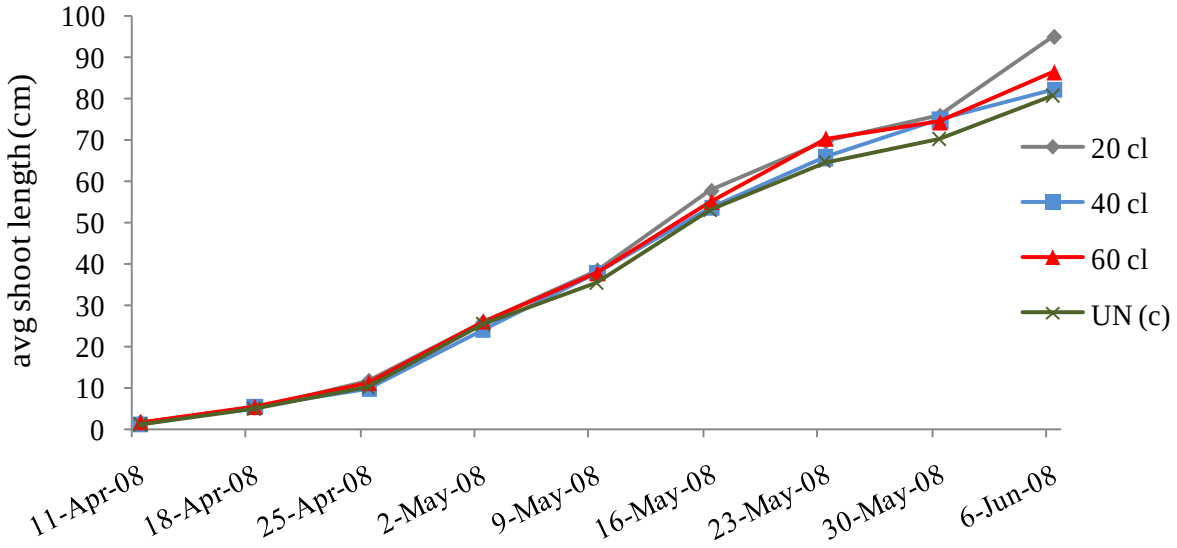


Figure 3.3: Treatment effects on weekly shoot growth in spring 2008 after three years of crop load and extended ripening treatments.

3.3.4 Grapevine Nutrition and Soluble and Insoluble Carbohydrates

Tissue nutrition: Petiole and blade tissues were collected and analyzed at bloom in 2008—after three years of treatments (Table 3.23). There were few consistent trends in the petiole and blade nutrition analysis. Nitrate Nitrogen in both blades and petioles was categorized in the "deficient" range according to the commercial laboratory standards used by California Ag Quest; however, this was most likely a result of nitrate being rapidly reduced by nitrate reductase rather than an actual deficiency. Nitrate is the predominant form of nitrogen taken up by the roots and is carried to the leaves where it is 'reduced' in the chloroplasts for amino acid synthesis (Mullins 1992). In addition, % Total Nitrogen was primarily in the "excessive" or "high" range—suggesting very active nitrate reductase. Potassium (%) had deficiencies in many of the treatments; only 60/22.5 and UN/28.5 were categorized as "marginal". All other treatments had nutrient levels above marginal.

Table 3.23: The effect of crop load and target °Brix at harvest on vine petiole and blade tissue nutrition in spring 2008, following three years of treatments.

BLADES

Treatment	Nitrate Nitrogen ppm	% Total Nitrogen	% Total Phosphorus	% Potassium	% Calcium	% Magnesium	% Sodium	Boron ppm	Zinc ppm	Manganese ppm	Iron ppm	Copper ppm
20/22.5	22	2.87	0.76	0.96	2.40	0.67	0.03	131	52	203	181	246
20/28.5	26	1.15	0.75	0.97	2.25	0.68	0.04	117	55	193	109	317
40/22.5	18	3.64	0.65	0.94	2.11	0.63	0.03	108	57	200	135	252
40/28.5	19	3.53	0.60	0.96	2.17	0.69	0.03	120	54	162	146	267
60/22.5	23	3.36	0.66	1.15	2.58	0.80	0.02	142	91	179	405	304
60/28.5	22	3.42	0.59	0.89	2.14	0.63	0.02	113	50	153	311	191
UN/22.5	20	2.99	0.54	0.86	1.99	0.65	0.03	101	56	143	112	274
UN/28.5	24	2.25	0.65	1.04	2.37	0.72	0.03	126	54	169	158	333

PETIOLES

Treatment	Nitrate Nitrogen ppm	% Total Nitrogen	% Total Phosphorus	% Potassium	% Calcium	% Magnesium	% Sodium	Boron ppm	Zinc ppm	Manganese ppm	Iron ppm	Copper ppm
20/22.5	261	0.92	1.05	2.46	1.91	1.26	0.02	54	24	76	88	31
20/28.5	203	0.89	1.11	2.30	1.95	1.43	0.04	57	32	78	71	39
40/22.5	378	0.92	1.08	2.46	1.64	1.27	0.02	51	65	85	77	45
40/28.5	212	0.91	0.97	2.37	1.77	1.37	0.05	54	23	66	76	36
60/22.5	261	0.91	1.00	2.64	1.84	1.36	0.30	56	23	68	77	38
60/28.5	253	0.90	0.89	2.29	1.69	1.31	0.03	48	21	60	96	27
UN/22.5	277	0.89	0.93	2.36	1.75	1.37	0.02	51	21	60	88	31
UN/28.5	227	0.89	0.97	2.64	1.88	1.40	0.04	54	22	62	82	39

Deficient Marginal Adequate High Excessive

Carbohydrate analysis: The carbohydrate status of the vine, analyzed by testing starch (mg/g) and soluble sugar (mg/g) in spurs after three years of treatments, demonstrated that there were no significant differences due to crop load or °Brix at harvest (Table 3.24). Consequently, there was no significant interaction.

Table 3.24: Effect of crop load and target °Brix at harvest on vine carbohydrates after three seasons of field treatments.

	starch (mg/g)	soluble sugar (mg/g)
Treatment	**2007**	**2007**
Crop load		
20	151.0	56.8
40	149.7	55.6
60	145.2	58.1
UN	142.9	57.4
	ns	ns
Target °Brix		
22.5	147.5	56.4
28.5	146.9	57.6
	ns	ns
Interaction	ns	ns

ns indicates not significant.

3.4 Discussion

3.4.1 The Effects of Crop Load and °Brix at Harvest on Berry Components

Berry weight development: Previous research on berry development reported that peak berry weight in Shiraz occurred near 90 DAA and was more related to DAA than to ripeness, berry size or calendar date (McCarthy 1999, Rogiers *et al.* 2004). In my experiment, the bloom date was inconsistent between years and thus influenced the calculation of DAA to reach peak berry weight. Veraison was within 3 days of a specific calendar date in each year and provides a more comparable analysis of the ripening time required to reach peak berry weight and target °Brix. Notably this demonstrates the grapevine's ability to accelerate phenological development after bloom even though bloom was inconsistent among years.

Crop load influenced the rate of sugar accumulation in terms of DAV to reach both peak berry weight and target °Brix at harvest. The effect of different crop loads on ripening time to reach

peak berry weight was more evident in 2006 and 2007. In 2005, all crop loads achieved peak berry weight at a similar number of DAV and DAA—with the exception of 20 cl. However, the 40 cl, 60 cl and UN required 14 more DAV to reach peak weight in 2005 than 2006 and 2007. In 2006 and 2007, the UN required 28 and 21 more DAV, respectively, to reach peak berry weight—relative to the 20 cl and 40 cl. The 2005 season had above average winter rainfall and yields. This may have aided in the synchronization of peak berry weight among all crop loads at 34 DAV or, this outcome may have been a factor of being the first year of treatments. The increased soil moisture due to above average winter rainfall would have lengthened the duration of time in which the roots depleted the available moisture from the soil profile. Furthermore, the same irrigation regime was used prior to budbreak in each year, and included only 8 irrigation hours as part of the pre bloom fertigation. The work of Keller *et al.* (2006) reported that prior to veraison, berries maintained size until the plant used 80 % of the transpirable soil water. Increased soil moisture may have enabled the hydraulic connection between berries and shoot to be intact longer and may explain the increased DAV to reach peak berry weight in 2005. Additionally, the 20 cl and 40 cl had a greater LAI and larger shoots, but with less clusters to ripen. This would have contributed to faster ripening due to increased photosynthetic capacity, and is supported by the work of Petrie *et al.* (2000) who stated that sugar accumulation is directly related to photosynthetic capacity of the vine. Theoretically, greater leaf area would have increased transpiration. The canopy was a modified-VSP, including the north side vertical and south side sprawl. Therefore, it is plausible that the lower crop loads may have depleted soil available moisture earlier than the UN which had a lower LAI, smaller shoots but was irrigated at the same volume. Moreover, vines with lower crop load had a greater ability to ripen their

crop and in turn reached peak berry weight prior to the UN in 2006 and 2007. Degrees Brix accumulation also followed this pattern and is discussed in Chapter 4.

The berry weight curves in the 20 cl and 40 cl display a steeper decline as DAV increased past peak berry weight relative to the 60 cl and UN. On average the 20 cl and 40 cl had the greatest % loss in berry weight across all years; although required the least DAV to reach 28.5 °Brix. In contrast, the UN had the lowest % berry weight loss but required the most DAV to peak weight and 28.5 °Brix. Berry dehydration caused by sun exposure on the clusters cannot be considered as a main driver of berry weight loss since the greatest berry weight loss was in treatments with the most canopy growth and the shortest exposure to the ambient environment. These results demonstrate that crop load affected the rate of sugar ripening, timing of peak berry weight and severity of berry weight loss. Furthermore, these data imply that the increased shoot growth (main shoots or laterals) and leaf area, as crop load reduced, potentially increased transpiration and thereby facilitated an earlier depletion of the soil moisture. Moreover, the increased shoot growth in lower crop loads must have occurred after the crop thinning given the minimal differences in the early season shoot measurements which concluded prior to the crop thinning.

Assuming crop load affected the rate of ripening, then berry hydraulic connections may have been different between crop loads. The water flow gradient can influence the amount of water reaching the berry and the amount of back flow which would account for some berry weight loss (Tilbrook and Tyerman 2008). Additionally, Keller *et al.* (2006) proposed that phloem unloading combined with solute accumulation in the berry may be responsible for the decline in xylem water influx to ripening berries and instead the xylem may recycle excess phloem water back to the shoots. During the late stages of ripening, the hydraulic conductance of the xylem is greatly reduced (Tilbrook and Tyerman 2008) and can hydraulically isolate the berry from the

parent vine (Greenspan *et al.* 1994, Keller *et al.* 2006). For the variety Shiraz, berry weight loss has been proposed to be due to a combination of reduced phloem inflow and continued transpiration (McCarthy and Coombe 1999, Rogiers *et al.* 2004, Tyerman *et al.* 2004, Keller *et al.* 2006), although backflow to the vine via the xylem may also contribute. Additionally, Rogiers *et al.* (2006) has postulated that there is a fourth phase of berry development in which the berry shrinks if left on the vine. Though detailed physiological measurements were not taken in my experiment, previous research on water relations in the berry during the late stages of ripening provide evidence to support speculation that a change in hydraulic connections may have occurred (Bondada *et al.* 2005, Keller *et al.* 2006, Tillbrook and Tyerman 2008, Mendez-Costabel 2007).

Berry weight at harvest: The xylem pathway continues to function in Stage III and Stage IV of berry development (Rogiers *et al.* 2001), although the phloem is dominant (Hrazdina *et al.* 1984). The development of hydraulic isolation of the fruit, due to xylem discontinuity, has been regarded as a prerequisite to prevent loss of solutes, via the xylem (Keller *et al*. 2006) in fruits that have apoplastic phloem unloading such as grapes (Patrick 1997). However, Tyerman *et al.* (2004) showed that the pathway remained functional, but the magnitude of reduction was variety—dependent. Additionally, Bondada *et al.* (2005) has demonstrated that xylem hydraulic conductance could still occur, provided the appropriate driving force of water flow to the berry was sustained. Findings from the above studies suggest that water movement in and out of the berry during the late stages of ripening is quite variable but certainly possible depending on the hydraulic connectivity and nature of the driving force of water into the berry. Both berry transpiration and backflow can affect berry volume (Rogiers *et al.* 2006).

Though winter rainfall was considerably different in each year of the experiment, the inverse relationship between peak berry weight and crop load was consistent. The results from this experiment substantiate previous research by Bravdo *et al.* (1984) and demonstrate that crop load reduction can increase berry weight. Berry size increased due to yield component compensation due to the crop load reduction. This is supported by the growth measurements, which indicated that vines with reduced crop load were able to grow larger shoots (i.e. longer main and lateral shoots) and subsequently had a higher LAI. Moreover, this suggests that a greater diversion of resources went to vegetative growth as crop load decreased. Presumably, berry size increased primarily due to pericarp cell enlargement rather than increased cell division based on the phenological stage when the crop was thinned i.e. E-L 31 (Mullins *et al.* 1992).

In this experiment, crop load affected the rate of fruit ripening and thus had an influence on the driving force of water into the berry. Therefore hydraulic connections may have differed between crop loads—however this remains a theory since direct measurements were not taken. The increased canopy growth and leaf area—indicated by the pruning weight and ceptometer data—confirm an increase in canopy due to the crop load reduction. Increased leaf area would change the water requirement for the plant and may have influenced the changes in water flow to the berry. All treatments were irrigated at the same volume per vine. Therefore, increased leaf area on the lower crop loads could have depleted soil moisture faster, thereby inducing a higher level of water stress on the vine and affecting the rate of berry weight loss. Water flow from the berry back to the vine was shown to occur under water stress conditions in some varieties (Lang and Thorpe 1989). Although the yield loss from extended ripening was significant, the loss from crop thinning would be far more detrimental economically to a grower paid on weight. This is discussed further in Chapter 7 (economic analysis).

Large differences in the average % berry weight loss occurred between the three seasons (2005-07). The 2005 season had the lowest average % loss in berry weight i.e. 11 % relative to 2006 and 2007 which had average losses of 13 % and 18 %, respectively. The 2005 season was distinguished by higher than average yields and slower sugar accumulation presumably due to above average winter rainfall. In contrast with 2007 which had below average yield, faster sugar accumulation and well below average rainfall. The differences in % berry weight loss may have been a direct effect of overall yield, or more likely reflect seasonal differences which influenced available soil moisture and/or the rate of sugar accumulation. Moreover, winter rainfall was the most prevalent factor distinguishing the three seasons: 2005 had much higher rainfall (559 mm) than 2007 which was well below the regional average at 113 mm. These results suggest that initial soil moisture may have influenced the rate of berry weight loss and solute accumulation, and thereby influenced the overall yield reduction.

Previous work reported that berry weight loss due to extended ripening was primarily due to dehydration (La Rosa and Nielson 1956, Coombe 1975, Hamilton and Coombe 1992, McCarthy 1997a, Bisson 2001, Battany 2005, Grant 2005). Results in my experiment suggest that the interaction of lower crop load and extended ripening accelerated berry weight loss. Similar to berry weight and °Brix results for berry development, these results indicate that lower crop loads required fewer DAV to reach the highest °Brix target of 28.5 in addition to a higher initial berry weight relative to the higher crop loads. Provided that the 20 cl and 40 cl averaged less DAV 'less time on the vine' (i.e. 77 and 87 DAV) relative to 92 and 94 DAV for 60 cl and UN, 'time on the vine' can be eliminated as the primary cause of differences in berry weight loss between crop loads. Therefore, berry weight loss may have been more related to the hydraulic connection between berry and shoot followed by berry dehydration. Certainly, results from this study and

previous research reaffirms that berry weight is reduced as DAV increase past peak berry weight and extended ripening occurs; however, the magnitude of loss may vary depending on the interactive effects of crop load, soil moisture, leaf area and hydraulic connections to the ripening berry.

3.4.2 Effect of Crop Load and Extended Ripening on other Yield Components

The significant differences in clusters/vine confirmed that the different crop loads were successfully implemented each year of the experiment. Additionally, the strong positive correlation between yield/vine *panel* and yield/vine *plot* confirmed that both methods of yield evaluation were similar; therefore yield components taken only on panel vines, i.e. mean cluster weight and clusters/vine, can be assumed to be representative of the entire treatment plot. For simplicity, only yield/vine *panel* will be discussed.

Grapevines are known to have a self regulation mechanism (Coombe and Dry 1992). This phenomenon particularly due to crop load has been demonstrated in previous research (Freeman *et al.* 1979, Smart *et al.* 1982, Bravdo *et al.* 1984, Edson *et al.* 1995, Petrie *et al.* 2000) and was apparent in this experiment. The inverse relationship between clusters/vine and mean cluster weight, berry weight and berries/cluster confirmed that the grapevines underwent increasing levels of yield compensation as crop load was adjusted. Although mean cluster weight was not always significantly different between *all* crop loads, generally the 20 cl and 40 cl were different from the 60 cl and UN. Furthermore, yield/vine was different between all crop loads each year—presumably due to the difference in clusters/vine. Moreover, the yield/vine data, as a factor of crop load only, paralleled the differences in clusters/vine. These results support the work of Edson *et al.* (1995) who reported that berries per cluster and mean cluster weight were

inversely correlated with clusters per vine in Seyval grapevines adjusted to five crop levels; additionally, yield/vine correlated with clusters/vine. Furthermore, Reynolds *et al.* (1994) and Bravdo *et al.* (1984) reported that berries/cluster, berry weight, and consequently mean cluster weight increased in vines which underwent crop reduction.

Berry weight and berries/cluster presumably contributed to the differences in yield/vine and mean cluster weight. Berries/cluster increased as crop load decreased in 2006 and 2007; an indicator of grapevine self regulation due to yield compensation. Cluster thinning was done at E-L 31; therefore differences in fruit set cannot account for the changes in berries/cluster, but could be a result of changes in the size of the inflorescence primordia (e.g. number of branches) as a result of the previous year's crop load. Although berries/cluster in 2005 were highest in the UN, the grapevine still demonstrated yield component compensation by reducing berry weight, thus causing a significant reduction in mean cluster weight relative to the 20 cl and 40 cl. In subsequent years, the UN exhibited the most self regulation in cluster size by means of reduced berries/cluster and berry size in response to crop load. In contrast, the 20 cl had the greatest berries/cluster in both 2006 and 2007. This is interesting given that average mean cluster weight was considerably lower in 2007 relative to 2006, i.e. 95 (g) and 125 (g) for 2007 and 2006, respectively. Nonetheless, berries/cluster were still similar in the 20 cl among both years. Furthermore, this suggests an 'up' regulation of yield components—specifically in berries/cluster for the 20 cl. Figure 3.4 illustrates the differences in berries/cluster for all crop loads as the seasons progressed. Values were calculated from peak mean cluster weight and peak berry weight. Individual season also influenced berries per cluster.

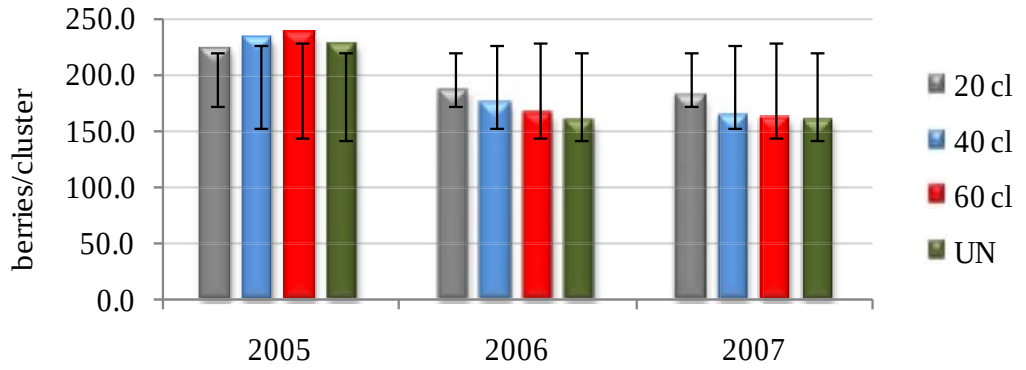


Figure 3.4: The effect of crop load on berries per cluster across three years and calculated from peak mean cluster weight and berry weight.

The increased crop load slowed down the rate of ripening and thus more DAV were required to reach target °Brix at harvest (Table 3.25). This is consistent with many other studies on crop load (Winkler 1954, Kliewer and Ough 1970, Bravdo *et al.* 1984, Bravdo *et al.* 1985, Jackson and Lombard 1993, Lakso and Eissenstat 2004) which indicated that increased crop load will alter ripening rate. The effects of extended ripening on yield/vine differed in % reduction of yield/vine among the three years; and may be explained by the variable number of DAV required to achieve °Brix targets between the three seasons. Generally, increased time on the vine (i.e. DAV) contributed to berry weight loss due to increased exposure to the ambient environment—even though effects were different between crop loads. Table 3.25 indicates that sugar accumulation in terms of °Brix was most rapid in 2007 and slowest in 2005. This trend corresponds with yield/vine—2005 had the highest yield relative to 2007 which had the lowest yields among the three years. These results support previous reports that increased yield delays maturity (Winkler 1954, Reynolds 1989, Jackson and Lombard 1993); however, in my experiment adequate sugar ripeness was still achieved in unthinned vines.

Seasonal differences are highlighted by the variation in DAV required to reach target °Brix for all crop loads. The only consistency was that higher crop loads always required more DAV to reach target °Brix. However, the differences between years suggest that season remains a large influence on ripening. Moreover, winter rainfall was substantially different among the three years of this experiment; representing respectively 157 %, 108 % and 32% of average annual rainfall in 2005, 2006 and 2007. Yield component trends were similar among years; yet, the seasonal differences in winter rainfall amounts likely influenced the annual growing and ripening cycle and must be considered when comparing data. Additionally, the acceleration between inconsistent bloom dates to consistent veraison dates each year is noteworthy.

Table 3.25: The number of days after veraison (DAV) required to reach target °Brix at harvest in each crop load for three seasons.

Crop Load	**DAV to reach target °Brix**																			
	2005					**2006**					**2007**					***Grand Mean***				
	22.5	***24***	***25.5***	***27***	***28.5***	***22.5***	***24***	***25.5***	***27***	***28.5***	***22.5***	***24***	***25.5***	***27***	***28.5***	***22.5***	***24***	***25.5***	***27***	***28.5***
20	23	40	51	60	81	29	33	42	64	78	16	24	29	50	73	***23***	***32***	***41***	***58***	***77***
40	35	45	56	64	95	30	40	52	72	83	22	29	41	62	83	***29***	***38***	***50***	***66***	***87***
60	50	57	64	82	97	35	44	61	79	89	24	34	54	76	90	***36***	***45***	***60***	***79***	***92***
UN	53	63	71	96	98	37	51	68	84	91	27	37	58	79	93	***39***	***50***	***66***	***86***	***94***
Avg	***40***	***51***	***61***	***76***	***93***	***33***	***42***	***56***	***75***	***85***	***22***	***31***	***46***	***67***	***85***					

When expressed as tonnes/ha, the losses due to both crop load and extended ripening render significant financial losses for wine grape growers paid by dollars per tonne. Both crop load and extended ripening reduced yield in all treatments relative to the UN at peak weight (Figure 3.5); however, the reduction due to cluster thinning was far more substantial. On average, extended ripening reduced t/ha by 14 % as compared with cluster thinning which reduced potential yield by 56 %. The combination of crop thinning to 20 cl/vine, an average yield of 7 t/ha, combined with extended ripening to 28.5 °Brix reduced t/ha by 63 %. These percentages are similar to

Guidoni *et al.* (2002) who reported that yield was reduced by 43 % in Nebbiolo vines with half the clusters removed at pea size. Interestingly, the average difference in DAV between the 22.5 °Brix and 28.5 °Brix targets among all crop loads was not substantially different. For example, the 20 cl, 40 cl, 60 cl and UN had on average 54, 58, 56 and 55 DAV, respectively, between the range of target °Brix (22.5-28.5). Therefore, crop load had no effect on this even though it significantly affected DAV to reach each target °Brix.

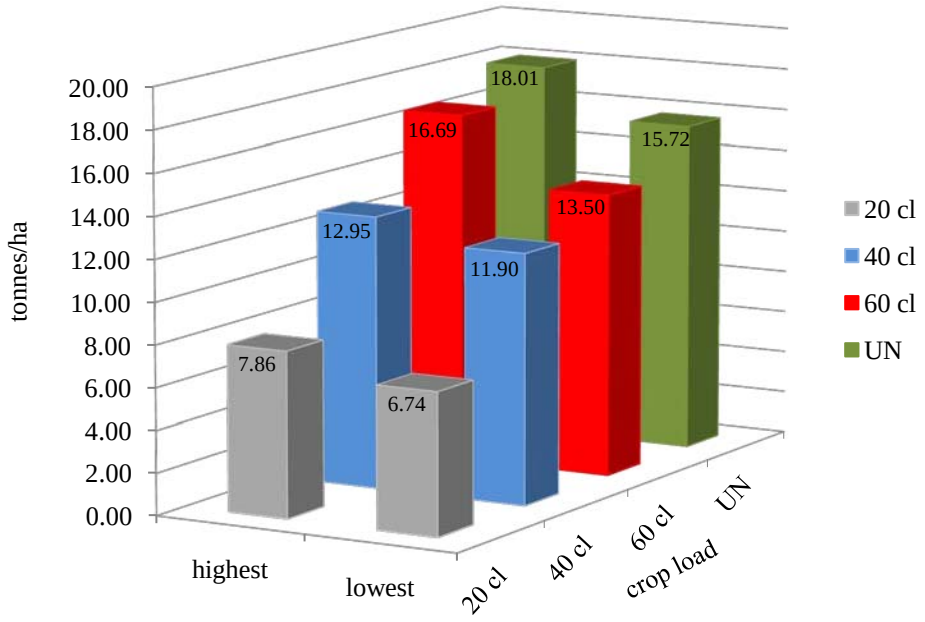


Figure 3.5: The effect of crop load and extended ripening on yield in tonnes per hectare (t/ha). The highest and lowest yield (t/ha) within each crop load are presented as an average of the three years 2005-2007. The lowest yields were always at the 28.5 °Brix target, regardless of crop load.

The effects on 'second crop' highlight the significant changes in vine growth due to crop load. Weaver (1976) described 'second crop' as the small clusters produced by lateral shoots. Presumably, increased second crop in the lowest crop load was due to a greater incidence of lateral shoots within the canopy—also indicating under cropped vines. This notion is supported by the pruning weights which increased as crop load decreased. Furthermore, LAI was greater in

the lower crop loads and the lowest PAR was in the 20 cl. Second crop is a nuisance and a costly problem for growers using machine harvesters. Second crop ripens later than primary clusters making it undesirable for winemaking. Consequently, growers are asked to keep second crop at a minimum and if necessary, second crop is hand thinned prior to harvest. Finally, the incidence and size of second crop can be used as a quick indicator of increased lateral growth and poor vine balance in Cabernet Sauvignon.

3.4.3 Effect of Crop Load and Extended Ripening on Vine Growth

Many changes occurred for vine growth, primarily due to crop load. The greatest mean cane weight and pruning weight was consistently at the lowest crop load. This contrasted with the UN which consistently had the lowest mean cane weight. Additionally, LAI increased as crop load decreased. These results correspond with Bravdo *et al.* (1984), Edson *et al.* (1993), and Edson *et al.* (1995) who reported that high crop loads were inversely related to shoot growth, leaf size and leaf area.

Although significant differences were not always consistent between the 40 cl, 60 cl, and UN, generally as crop load increased mean shoot weight decreased highlighting the vine's yield compensation response. Shoots/vine counts confirmed that winter pruning was consistent with regard to the standard node number per vine and that spring shoot thinning had consistent uniformity. Therefore, uneven cultural practices (i.e. pruning and/or shoot thinning) can be eliminated as the cause for changes in pruning weight and shoot weight. This suggests that increased mean shoot weight must have been due to increased main shoot length, shoot diameter and/or increased lateral growth. Consequently, pruning weight/vine increased due to increased mean shoot weight. All vines which underwent crop reduction increased in pruning weights

relative to the UN with one exception—60 cl in 2006 (Fig. 3.6), presumably due to the small differences in yield between the 60 cl and UN. The greatest % increase for pruning weight was in the 20 cl followed by the 40 cl, indicating that these crop loads were partitioning more of their resources towards vegetative growth rather than reproductive growth. Additionally, for 20 cl and 40 cl, the % difference relative to the UN increased as the years progressed (Figure 3.6) suggesting that the lower crop loads may have stored excess reserves which thereby contributed to the increasing difference (%) in subsequent years. These results conflict with those of Keller *et al.* (2005) who reported that cluster thinning at two phenological stages failed to influence vegetative growth for the varieties Cabernet Sauvignon, Riesling and Chenin blanc. However there was no preliminary shoot thinning in his experiment and the cluster thinning technique differed from the present experiment in that preferential thinning was done on non count shoots. Additionally, site characteristics such as soil type and climate may have caused these different outcomes.

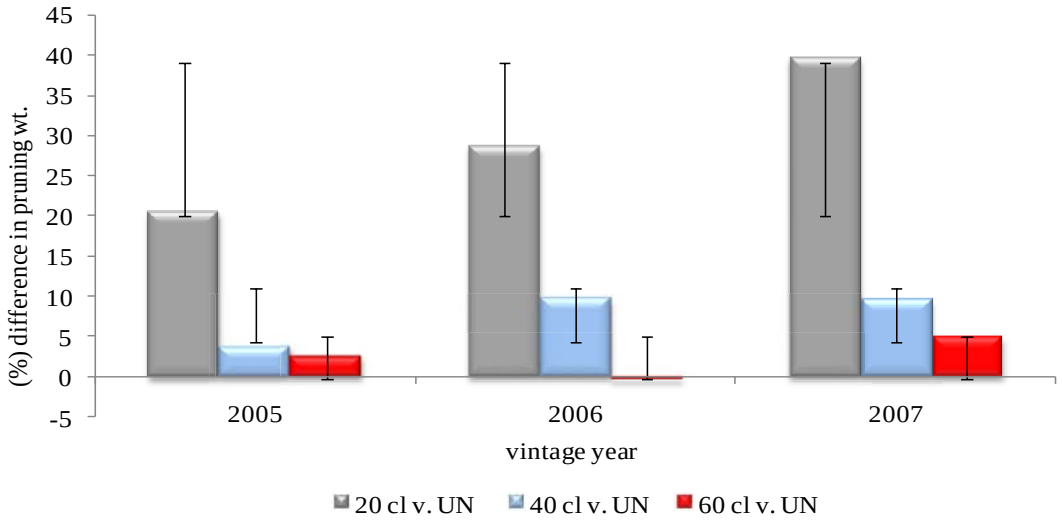


Figure 3.6: The effect of crop load on the percent (%) difference in pruning weight/vine (kg) relative to the unthinned (UN) for three vintage years.

Certainly crop thinning significantly altered the Y/P ratios although overall ratios varied between years. Generally, vines with Y/P values between 5-10 are considered to be within the optimal range (Bravdo *et al.* 1984, Bravdo *et al.* 1985); and are classified as well-balanced and capable of fully ripening their crop while producing high quality wines (Kliewer and Dokoozlian 2005). Vines with Y/P ratios below 4.0 or 5.0 may be categorized as out of balance due to being overly vigorous and under cropped. The 20 cl fell into this category with average Y/P ratios of 3.1, 1.4 and 2.3 in 2005, 2006 and 2007, respectively.

Overall, the growth data clearly demonstrates that crop thinning affected vine balance within each year. Lower crop loads were negatively affected and indicated under cropped and overly vigorous vines. The UN fell into an optimal range in 2005 and 2007, suggesting that on this site, crop reduction was unnecessary and disturbed the natural balance of the vine.

Surprisingly, the mean shoot weight and pruning weights were considerably higher in 2006 than in 2005, even though 2005 had substantially more winter rainfall. In addition, the Y/P ratios were noticeably lower than 2005 and 2007 and below optimum. This may be explained by an atypical rain event on May 15, 2006 in which 28 mm of rainfall occurred. The in-season rainfall would have supplemented the soil profile with increased moisture during the primary phase of shoot growth and most likely attributed to the substantial increase in mean shoot weight, pruning weights and consequently lower Y/P ratios in 2006. Extended ripening had minimal effects on mean shoot weight and pruning weight per vine.

LAI and PAR: Previous research by Edson *et al.* (1995) and Petrie *et al.* (2000) demonstrated that leaf area/vine was inversely related to crop load and concurs with results from this

experiment in which LAI increased as crop load decreased. Consequently there was an inverse relationship between LAI and PAR.

Interestingly, in all treatments LAI decreased and PAR increased between veraison and harvest. In fact, LAI reduced by 18 % on average between veraison and harvest, accordingly PAR increased by 15 % between veraison and extended ripening to 28.5 °Brix. There was a negative correlation between DAV and LAI (harvest) $p \leq 0.001$. Figure 3.7 illustrates that LAI (harvest) reduced as DAV increased. This relationship is most likely explained by leaf senescence of older or damaged leaves within the fruiting zone as DAV increased and suggests that extended ripening caused increased PAR interception in the fruiting zone due to reduced LAI.

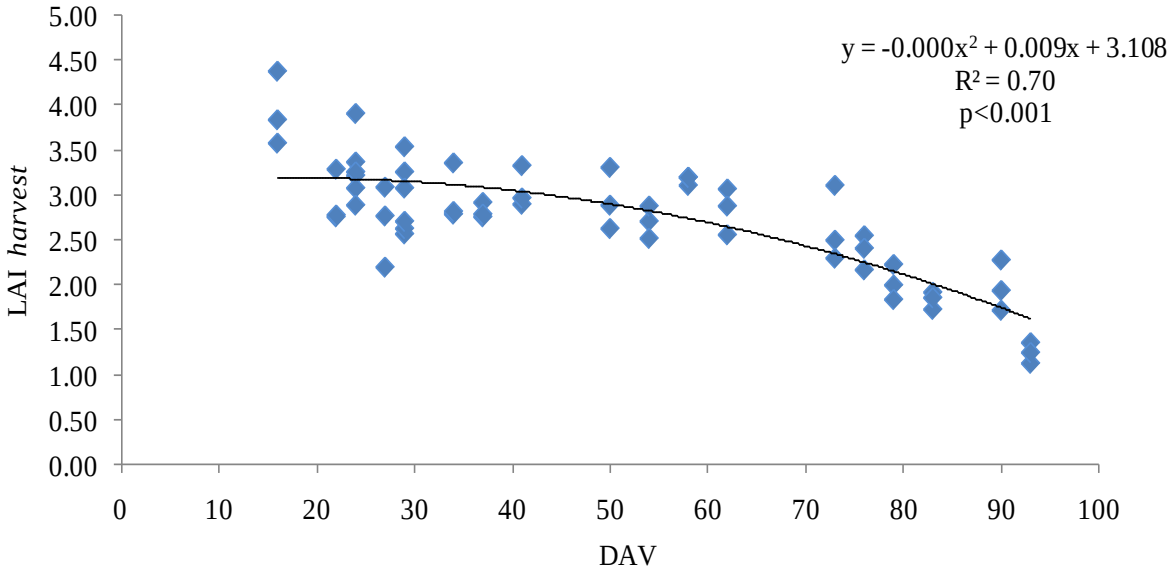


Figure 3.7: Regression analysis of leaf area index at harvest LAI (harvest) and days after veraison (DAV) required to reach target °Brix for all treatments in 2007.

Crop load effects on LAI (Figure 3.8) were likely a result of actual changes in shoot growth—supported by the mean cane weight, pruning weight and second crop data. Although differences were more significant in the harvest measurements, it should be noted that the 20 cl consistently

had the highest LAI and lowest PAR relative to all other crop loads at both veraison and harvest. Reports have established that overly-shaded fruit can be detrimental to wine quality (Smart *et al.* 1985, Smart *et al.* 1988, Ristic *et al.* 2007). Additionally, canopies with well exposed leaves and fruit have scored highest in wine quality taste panels (Smart 1982, Smart *et al.* 1991). Relative to this experiment, the 20 cl undoubtedly had the most shading on the fruiting zone throughout veraison and harvest, and most likely contributed to negative effects in wine quality—discussed further in Chapter 4.

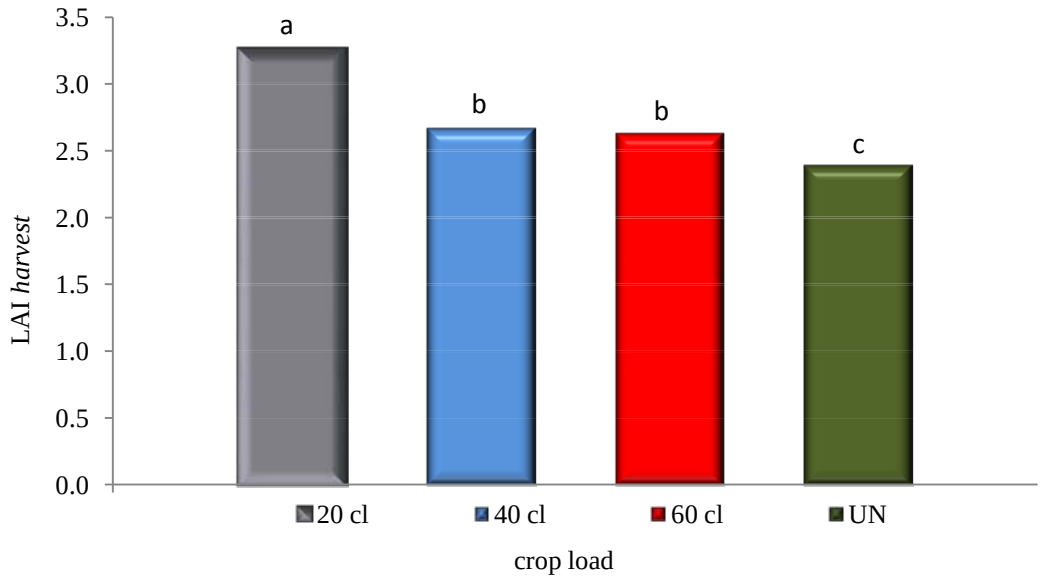


Figure 3.8: The effect of crop load on leaf area index at harvest LAI (harvest) in 2007. Each column represents the average LAI (harvest) within each crop load. Columns with different letters differ significantly at $p<0.001$.

The interaction between crop load and target ˚Brix at harvest for LAI (harvest) demonstrated that a more substantial reduction in LAI (harvest) occurred at the higher crop loads, even though LAI (harvest) decreased in *all* crop loads as °Brix at harvest increased (Figure 3.9). Furthermore, the 20 cl had the highest LAI (harvest) at each target °Brix. The UN lost 61 % of its peak LAI (harvest) by the final harvest at the 28.5 °Brix target. In contrast, the 20 cl only reduced by 33 %

at the 28.5 °Brix target. This interaction is further explained by the number of DAV required to reach the 28.5 °Brix target for each crop load. The UN required 93 DAV to reach the 28.5 °Brix target compared with the 20 cl which only required 73 DAV. Therefore the UN had more time for leaves to senescence due to water stress and/or environmental factors as late autumn conditions became more prevalent. Nonetheless, it should be noted that UN/22.5 had the lowest overall LAI *harvest* relative to all other crop loads at the first °Brix target (22.5 °Brix), in strong contrast with 20/22.5 which had the highest.

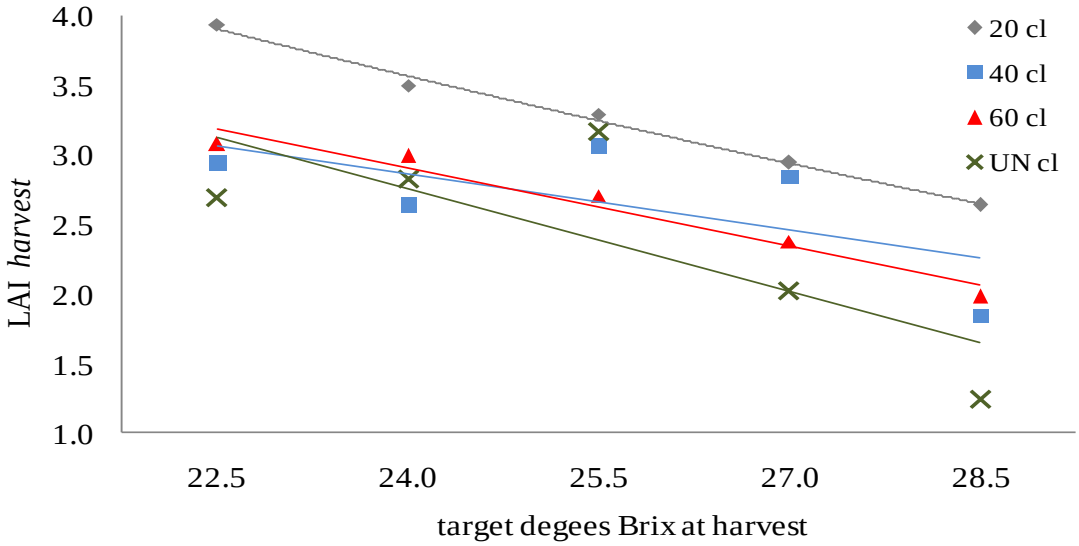


Figure 3.9: The interaction of crop load and °Brix at harvest on leaf area index LAI (harvest) measured at harvest in 2007.

Certainly, LAI and PAR reflect the effects of crop load on growth, indicating that crop reduction increased vine growth which thereby increased LAI and decreased PAR in the fruiting zone. In addition, extended ripening significantly increased PAR in the fruiting zone presumably due to increased DAV to harvest and leaf senescence. The interactive effect of high crop load and high °Brix at harvest resulted in the lowest LAI and highest PAR, dissimilar to the lowest crop load

which consistently had the highest LAI at all °Brix targets. Again, these results support that the most optimal vine balance was in higher crop loads.

3.4.4 Grapevine Nutrition and Soluble and Insoluble Carbohydrates

As a means of testing treatment effects on the reserve storage and spring shoot growth, shoot measurements began at bud break and continued until E-L 31 in 2007 and 2008. Percent bud break was measured in 2008, only, after three years of treatments and was not significant—indicating that bud break was consistent among treatments. The shoot growth measurements confirm that shoot growth, prior to crop thinning, was unaffected by the previous year's treatments. These results are additionally supported by the carbohydrate analysis which showed no differences in either starch or soluble sugar in spurs, after three years of treatments. The lack of differences between spring shoot growth prior to crop thinning supports the notion that increased shoot growth was a result of crop reduction. Furthermore, extended ripening had no effect on spring shoot growth.

The carbohydrate status of the vine was unaffected by crop load and/or extended ripening after three years of treatments. This is consistent with research by Wample and Bary (1992), Petrie *et al.* (2000), and Howell (2001) who reported that post harvest periods are not essential for sustained vine health and productivity. In contrast, Holzapfel *et al.* (2006) demonstrated that extending the length of the post harvest period by early crop removal over two consecutive seasons, increased yield by 48 % in the third season; and suggested that adequate post harvest recovery is crucial for maintaining high yield productivity. In the current experiment, the lack of differences in carbohydrate analysis confirmed that the vines ability to store carbohydrates was not hindered by the treatments implemented upon them even though there were significant

differences in canopy growth. Moreover, neither extended ripening nor crop load affected carbohydrate storage in subsequent seasons.

Finally, in this experiment there were minimal detrimental effects on vine nutrition or reserve storage after three years of treatments. However, it is unknown if significant differences may occur after longer periods (e.g. > 4 years) of crop load or extended ripening practices. The research of Wample and Bary (1992) and Gu *et al.* (2006) suggests that carbohydrate or other nutritional differences due solely to extended ripening are unlikely. However, to more thoroughly investigate the effects of crop load, future research is suggested to explore the carbohydrate status of other plant parts, i.e. roots and shoots at different phenological stages and pre-bloom sap 'bleeding. Starch measurements at the beginning and end of a multi- season experiment could provide additional information for treatment effects on vine carbohydrates. The research of Zapata *et al.* (2004) stated that root carbohydrates play the major role in supplying the growing tissues with nutrients, as compared to the starch located in trunks of canes. Bennett *et al.* (2005) reported that carbohydrate reserve storage must be complete with ripening in cool climate regions such as Canterbury, New Zealand since there is no post harvest accumulation. This contrasts with Williams (1996) who showed that starch and sugar continued to accumulate in the trunks of Thompson Seedless vines until leaf fall—however this was in a warm climate. Furthermore, it is likely that the large variations in carbohydrate reserves are influenced by grape variety and viticultural region; including climate and vineyard management (Bennett *et al.* 2005).

The bloom tissue nutrition indicated that only nitrate nitrogen was in a deficient range; however, this was consistent among all treatments and was contrasted by excessive and high levels of Total Nitrogen. These results suggest that nitrate in leaves and petioles were rapidly reduced by

Nitrate reductase and hence explain the contrasting values between Nitrate and Total Nitrogen. Unfortunately, there was no obvious explanation, based on treatment effects, as to why Potassium was greater in the 60/22.5 and UN/28.5 relative to all other treatments. By and large, there were minimal treatment effects on vine nutrition as investigated by bloom tissue analysis.

3.5 Conclusions

a) The interaction of crop load and extended ripening indicated that lower crop loads had a greater initial peak berry weight and lost a higher % of their peak berry weight while requiring less DAV to reach target °Brix. It is hypothesized that a change in the rate of ripening and hydraulic connection between berry and shoot affected this outcome.

Further investigation is necessary to thoroughly understand this interaction.

b) The grapevines progressively exhibited self regulation and yield component compensation as a result of crop load; berry weight, berries/cluster and mean cluster weight decreased as crop load increased.

c) Crop load affected the 'rate of ripening' each year. Generally, as crop load increased the days after veraison required to achieve target °Brix increased; however, all crop loads surpassed traditional levels of ripeness and achieved 'extended ripening'. Seasonal characteristics, particularly winter and spring rainfall, largely influenced the rate of ripening each year. Seasons with above average winter rainfall required more days after veraison to ripen fruit and had greater growth relative to those with below average rainfall even though overall trends remained similar amongst all three years.

d) Extended ripening to the highest °Brix target caused significant losses in yield (14 % on average); however, yield reduction due to crop thinning was far more significant. The combination of extended ripening and crop thinning caused the most significant loss, i.e. 63 % yield reduction. This renders a financially-devastating loss for a grower paid only by dollars/tonne.

e) Canopy growth was regulated by crop load: mean cane weight and pruning weight/vine reduced as crop load increased. The percent change in pruning weight for lower crop loads relative to the UN became increasingly greater as the years progressed.

f) Crop reduction had detrimental effects on vine balance. The most optimal vine balance, in terms of Y/P ratio, was consistently for the UN which overall averaged 7 Y/P. Y/P ratios became increasingly less optimal as crop load was reduced—indicating overly vigorous and under cropped vines.

g) The light interception at the fruiting zone was reduced by crop load reduction due to increased LAI and decreased PAR. Extended ripening decreased LAI and increased PAR at all crop loads—presumably due to leaf senescence. Treatment interactions indicated that the lowest crop loads had the highest LAI at all °Brix targets. LAI decreased and PAR increased considerably between the veraison and harvest measurements. LAI and PAR concur with pruning weight and vine balance results.

h) Vigor management is essential for quality production of Cabernet Sauvignon. Increased availability of soil moisture and nutrients can override the effects of crop load and cause significant changes in canopy growth and microclimate. High levels of second crop may be

an indicator of increased lateral growth and inadequate vine balance. A recommendation to growers is to observe density and size of second crop as an indicator of increased vine vigor.

i) Extended ripening and crop load had no effect on grapevine nutrition; however, previous research suggests that more detailed carbohydrate analysis i.e. roots and bleeding sap pre bud break, and/or increased duration of crop load treatments could cause differences due to crop load. Further research is recommended.

Chapter 4: EFFECTS OF CROP LOAD AND EXTENDED RIPENING ON FRUIT AND WINE COMPOSITION

4.1 Introduction and Experimental Aims

Crop load reduction is commonly required of wine grape growers as a vineyard management tool to improve fruit and wine composition; however, conflicting results have been reported. Winkler (1954) and Kliewer and Weaver (1971) reported that increased crop load delayed ripening and decreased wine quality. In contrast, under-cropping can increase acid, nitrogenous compounds and salt accumulation in berries resulting in unbalanced wines and poor flavor (Sinton *et al.* 1978). Studies by Weaver *et al.*(1961), Sinton *et al.* (1978), Freeman *et al.* (1980), Ough and Nagaoka (1984), Bravdo *et al.* (1985), Ewart *et al.* (1985), Reynolds *et al.* (1986), Zamboni *et al.* (1996), and Chapman *et al.* (2004) reported either no effects due to yield or increased wine quality in higher yielding vines. Additionally, factors such as variety, rootstock and certainly terroir, have a major impact on optimal crop load and subsequent wine quality. Ough and Nagaoka (1984) demonstrated that cluster thinning had minimal effects on ripening time, fruit and wine composition, or wine aroma—however, effects of location were far more considerable. In addition, Kasimatis (1977) reported that low cropped Zinfandel produced the highest grape and wine quality. Furthermore, canopy microclimate may lead to the explanation behind common observations that high yields cause reduced wine quality (Smart *et al.* 1985 a, b). Therefore, yield *per se* is not a good indicator of wine quality—rather, it is the environment and management that affects yield and therefore influences wine quality (P. R. Dry 2008 personal communication). Furthermore, optimum viticultural practices, specific to the site characteristics, are more likely to improve fruit and wine composition.

Grape ripeness is an important specification for wine grape purchasing contracts. Traditionally, commercial harvest was near 24.0 °Brix and many grape purchasing contracts were written to this specification (Grant 2005). However, with the new criteria of 'flavor ripeness' rather than just sugar ripeness, the current commercial harvest has shifted towards 26.0 °Brix or greater—particularly for red wines such as Cabernet Sauvignon. Additionally, many wineries have pursued nontraditional, high alcohol wine styles to satisfy apparent market demand and wine critics (Grant 2005). Deciding when a vineyard reaches 'flavor ripeness' without objective indices presents a conflicting challenge for winemakers and wine grape growers. Furthermore, limited published research exists indicating the effects of extended ripening on fruit and wine composition (Grant 2005, Gu *et al.* 2006) and the consequences of extended ripening are only understood in a broad sense (Bondada *et al.* 2006). Substantial winemaking problems are associated with extended ripening, such as arrested or sluggish fermentations—typically due to high alcohol (ethanol) in subsequent wines (Bisson 1999, 2001, Bisson and Butzke 2000).

Wine quality can be difficult to define; however, quality defects are identifiable. Generally, Australian dry red wines which are high in pH and low in color and phenolic content are considered to have quality defects (Somers 1975). Berry development predicates composition which in turn predicates quality (Coombe and Iland 1987). The bell pepper aroma in Cabernet Sauvignon (Noble *et al.* 1995, Chapman *et al.* 2004) has been correlated with concentrations of isobutylmethoxypyrazine (IBMP), which is considered the most significant methoxypyrazine from a sensory and wine quality perspective (Wilkinson *et al.* 2006). High concentrations of methoxypyrazines can dominate wine aroma and be detrimental to wine quality (Allen *et al.* 1998) or suggest an olfactory defect (Roujou de Boubée *et al.* 2002, Preston *et al.* 2008). Winemakers commonly use terms for 'vegetal' and 'nonvegetal/fruity' aromas as a main

criterion in the characterization of Cabernet Sauvignon wines, and to distinguish specific qualitative differences (Preston *et al.* 2008). Methoxypyrazines are known to break down in the presence of light (Morrison and Noble 1990, Noble *et al.* 1995, Hashizume and Samuta 1999). Consequently, extended ripening may be aiding the breakdown of methoxypyrazines and subsequently improving wine quality in Cabernet Sauvignon. These speculations and the anecdotal evidence suggesting that extended ripening improves wine quality are limited and insufficient. Therefore, it is imperative that extended ripening be investigated scientifically and parallel with commercial practices.

The aims of this study were to:

1. Monitor the development of berry composition through to the late stages of ripening.
2. Study the effects of crop load and extended ripening on fruit and wine composition.
3. Test whether decreased crop load and/or extended ripening improves wine quality based on wine phenolic and color analysis.

4.2 Materials and Methods

4.2.1 Berry Development

Sugar accumulation: Sugar accumulation measured as °Brix was monitored at weekly intervals from 100 % veraison to harvest. Berries were collected, following the berry collection protocol described in Chapter 3 and Appendix 1, and crushed by hand for 2 minutes within a plastic bag. Juice was then filtered into a glass beaker to settle. Settled juice was poured off into a 50 mL glass beaker and used for analysis. Degrees Brix was measured in triplicate using an Anton Paar DMA 35N density meter, then sent on for pH and TA measurements.

pH: Juice pH was measured on the settled juice sample—also used for sugar accumulation—using an Orion 720A pH meter and probe.

Titratable acidity: Juice titratable acidity was measured by hand titration using 100 mL of deionized water, 5 mL of juice, and a few drops of phenolphthalein indicator dye into a flask. A standard solution of 1N sodium hydroxide (NaOH) solution was used for the titration. The NaOH solution was normalized before beginning titration measurements each week to ensure accuracy in the titration calculation for titratable acidity. The detailed procedure is described in Appendix 5.

Sugar (g) per berry: The weekly °Brix and berry weight data were used to calculate sugar per berry using the following formula: Sugar (g)/berry = (°Brix/100) x (mean berry weight)

4.2.2 Harvest Juice Chemistry

Juice chemistry: Samples for juice chemistry at harvest were taken just after crushing. Degrees Brix was measured using the Anton Paar DMA 35N density meter. The juice pH and titratable acidity (TA) were measured on a TIM 865 titration manager by Radiometer Analytical.

Yeast assimilable nitrogen: Yeast assimilable nitrogen (YAN) was measured using a Randox enzymatic kit (sample preparation is described in Appendix 8). The YAN number was determined by adding alpha amino nitrogen (NOPA) and ammonium together. The YAN level dictated the nutrient additions into fermentations (see nutrient schedule in Appendix 4).

4.2.3 Fermentation and Winemaking Details

Microvinification: Macro Bins with each treatment replication (three/treatment) were balanced to the same total fruit weight/bin. In 2005 and 2006, all fermentations were balanced to 454 kg

of grapes. Due to a lower overall yield, yield compensation in higher crop loads, and extended ripening, bins in 2007 were balanced accordingly. The 2007 bins had between 386 – 363 kg of fruit. Certainly, all replications within a treatment were balanced to the same weight. Fruit from each individual bin were crushed, de-stemmed and a specific winemaking protocol was followed (Appendix 6).

Fermentation monitoring: Primary fermentation was monitored daily by checking must temperature and °Brix at 13:00 hours each day of primary fermentation. The industry standard for red winemaking in California is to achieve 32.2 °C during primary fermentation. Therefore, the goal for each fermentation was to reach a peak temperature of 32.2°C (90 °F). Heating plates were inserted if necessary to bring must temperature to the desired level.

Pressing: All treatments were pressed on the seventh day of fermentation as explained in the winemaking protocol. Wine was pumped from the Macro Bin into the corresponding barrel. The remaining skins were pressed in a one ton pneumatic basket press to a maximum pressure of 2 bars. Samples for press wine chemistry and phenolics were collected at that time—from free run wine, only. Press samples were limited to free-run wine to maintain consistency in sampling for press analysis and avoid varying levels of press fractions. The free run press samples were collected after the basket press was filled with skins, and before the bladder began filling with air. All pressings were added with previously pumped wine to the corresponding barrels. Each barrel was then inoculated with the malolactic (ML) bacteria culture CHR Hansen Viniflora® Oenos.

Press wine chemistry: TA and pH were analyzed using the TIM 865 titration manager by Radiometer Analytical. Ethyl Alcohol (ETOH) was measured using an ebulliometer by Dujardin-Salleron and FOSS WineScan —FOSS protocol is described in Appendix 9.

Malolactic fermentation: Malolactic fermentation (ML) was monitored weekly by paper chromatography followed by enzymatic analysis on the spectrophotometer. Malolactic fermentation was deemed sufficient when levels were < 100 ppm.

Racking: The wine was racked at the completion of ML fermentation and stored in neutral barrels of American oak, World Cooperage medium toast. Post ML phenolic samples were set aside at that time. The SO_2 level was monitored every 4-6 weeks and kept between 30-45 ppm free SO_2.

4.2.4 Wine Color and Phenolics

Wine color and phenolics were analyzed using a Shimadzu UV-1700 PharmaSpec spectrometer. Wine samples from both press and post ML fermentation were analyzed at Absorbance (A) 280 nanometers (nm), Absorbance (420 nm) and Absorbance (520 nm) (protocol in Appendix 7).

Total phenols: were obtained by Absorbance (280 nm) on the spectrophotometer.

Color density: The color density (CD) was obtained by combining values of Absorbance (420 nm) and Absorbance (520 nm). Absorbance (420 nm) + Absorbance (520 nm) = CD

Hue: Hue is expressed as the ratio between the Absorbance (420 nm) and Absorbance (520 nm) i.e. Absorbance (420 nm)/Absorbance (520 nm).

Methoxypyrazines: The compound iso-butyl-methoxypyrazine was measured by Gas Chromatography-Mass Spectroscopy (GC-MS) at E. & J. Gallo Winery in Modesto, CA, USA, on wine from the 2006 and 2007 vintage.

4.3 Results

4.3.1 Berry Development

Sugar accumulation: Degrees Brix increased with days after veraison (DAV) for all crop load treatments in each year (Fig 4.1). Sugar accumulation was largely affected by season. The slowest °Brix accumulation overall occurred in 2005 and the fastest in 2007 regardless of crop load. The 20 cl was consistently higher overall in °Brix development and had the highest initial °Brix each season relative to the other crop loads. In contrast, the UN had the slowest increase in °Brix compared with all other crop loads.

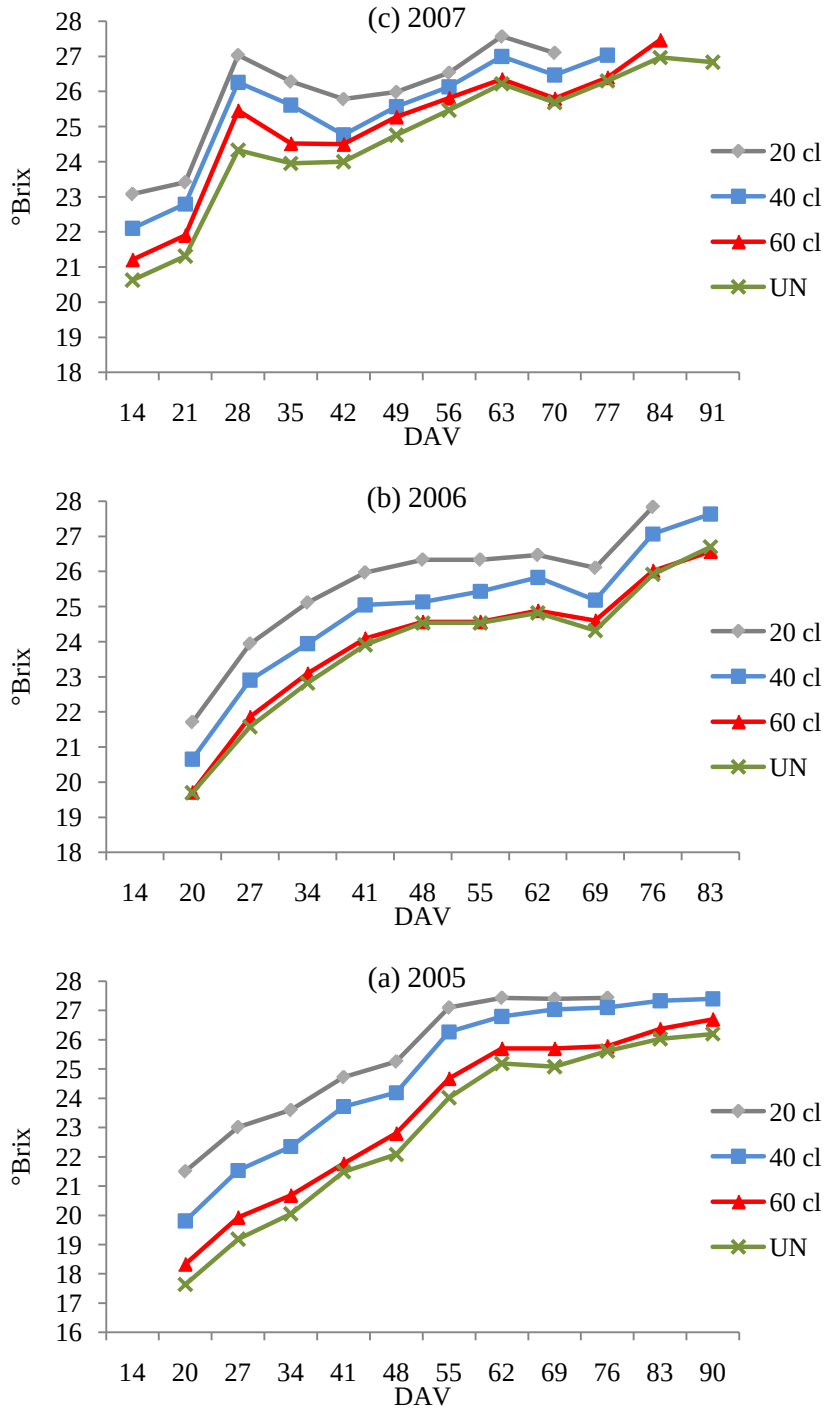


Figure 4.1: Crop load effects on °Brix development during the ripening period in 2005 (a), 2006 (b), and 2007 (c).

Sugar per berry: The sugar in grams (g) per berry for each crop load over time is presented in Figure 4.2. Crop load affected sugar (g)/berry each year in that the initial measurement of sugar (g)/berry was greater as crop load decreased. In 2007, reduction of sugar (g)/berry began earlier, at fewer DAV, in the lower crop loads (20 cl and 40 cl) relative to the higher crop loads. Figure 4.2 (c) illustrates that the 20 cl and 40 cl peaked in sugar (g)/berry near 28 DAV whereas the 60 cl and UN did not begin reducing until 56 DAV. The 20 cl reduced slightly in 2005 at approximately 24 °Brix—however it was much less noticeable than in 2007.

The magnitude of sugar (g)/berry was largely affected by the individual season. The highest levels of sugar (g)/berry were achieved in 2005 when all crop loads exceeded 0.18 (g) sugar/berry and the 20 cl attained over 0.24 (g) sugar/berry. In contrast, none of the crop loads achieved 0.18 (g) sugar/berry in 2007, although 2007 had the highest average °Brix measurements. The contrast in sugar (g)/berry and °Brix between the 20 cl and UN for all years is illustrated in Figures 4.3 a, b and 4.4 a, b. The seasonal differences are also reflected in Table 4.1 which lists the number of DAV associated with peak sugar (g)/berry. Although °Brix in the 20 cl and UN followed a similar pattern between the three years, sugar (g)/berry differed more for the 20 cl between the three years.

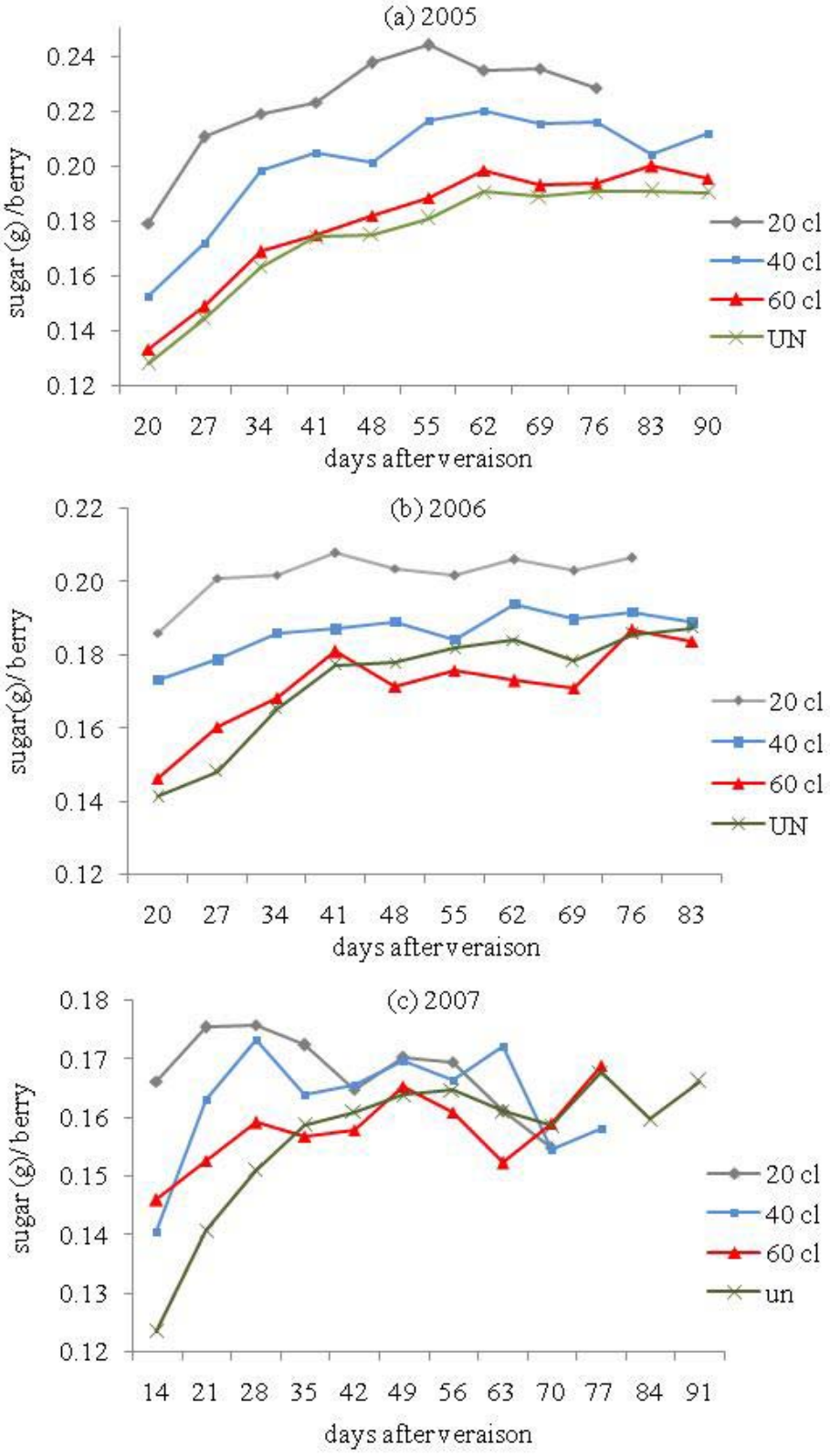


Figure 4.2: Crop load effects on sugar (g) per berry during the ripening period in 2005 (a), 2006 (b), and 2007 (c).

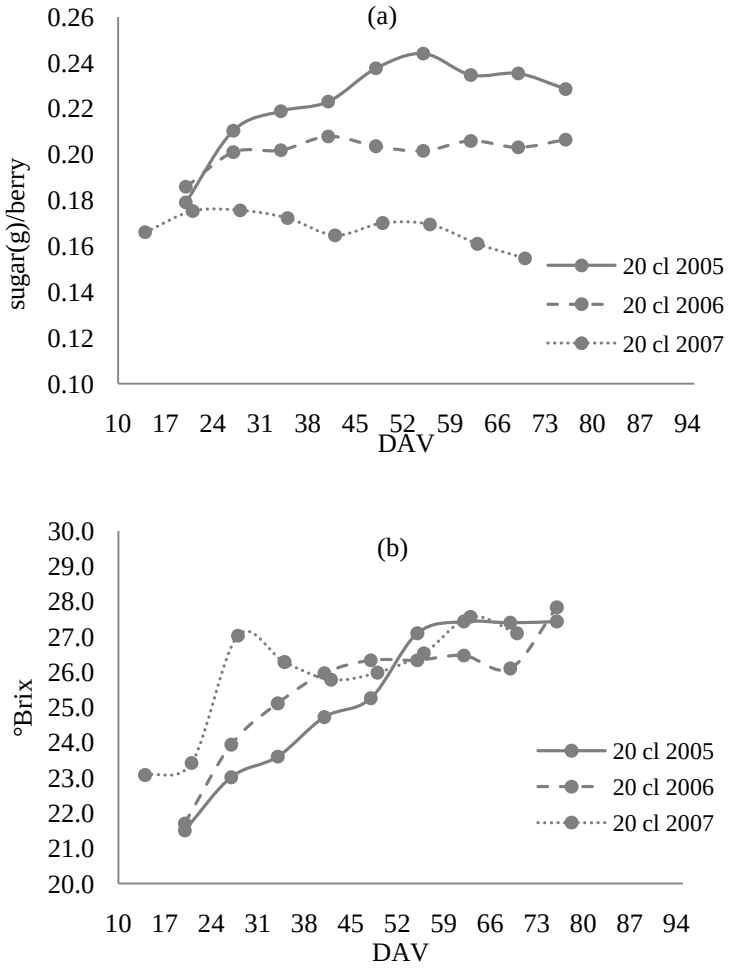


Figure 4.3: Seasonal affects on sugar accumulation in the 20 cl for sugar (g)/berry (Figure 4.3 a) and °Brix accumulation (Figure 4.3 b) in 2005, 2006 and 2007.

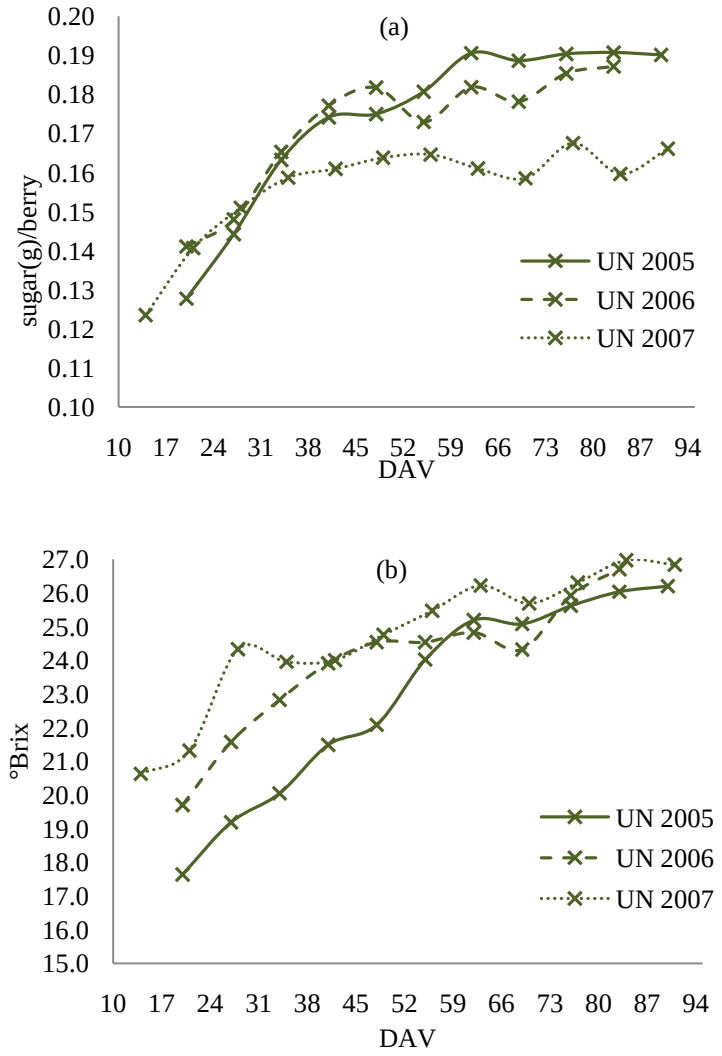


Figure 4.4: Seasonal affects on sugar accumulation in the UN for sugar (g)/berry (Figure 4.4 a) and °Brix accumulation (Figure 4.4 b) in 2005, 2006 and 2007.

Table 4.1: The effect of crop load and extended ripening on days after veraison (DAV) required to reach peak sugar (g)/ berry in three seasons.

	DAV to peak sugar(g)/ berry			
Crop Load	**2005**	**2006**	**2007**	**Grand Mean**
20	55	41	28	**41**
40	62	62	28	**51**
60	83	76	77	**79**
UN	83	83	77	**81**
Grand Mean	**71**	**66**	**53**	

pH development: The pH development for each crop load over time and each year is presented in Figure 4.5. Overall, pH increased as DAV increased for all crop loads and in each year. In general, pH was affected by crop load in that higher crop loads had lower pH relative to the lower crop loads. The pH was consistently highest in the 20 cl as compared with the UN which had the lowest pH at most sampling dates. The trend in pH was consistent across all years and concurs with the trends in titratable acidity. Seasonal differences affected the extent of change in pH. The 2006 season had higher pH measurements overall and all treatments reached >3.90 pH by the final sampling dates. In contrast, none of the treatments reached 3.90 pH or greater at any point during the 2005 or 2007 seasons. These data suggest that pH was affected by both crop load and season. Note that although TA was adjusted on harvested fruit and its subsequent must during primary fermentation, these pH development data were from berry sampling before harvest.

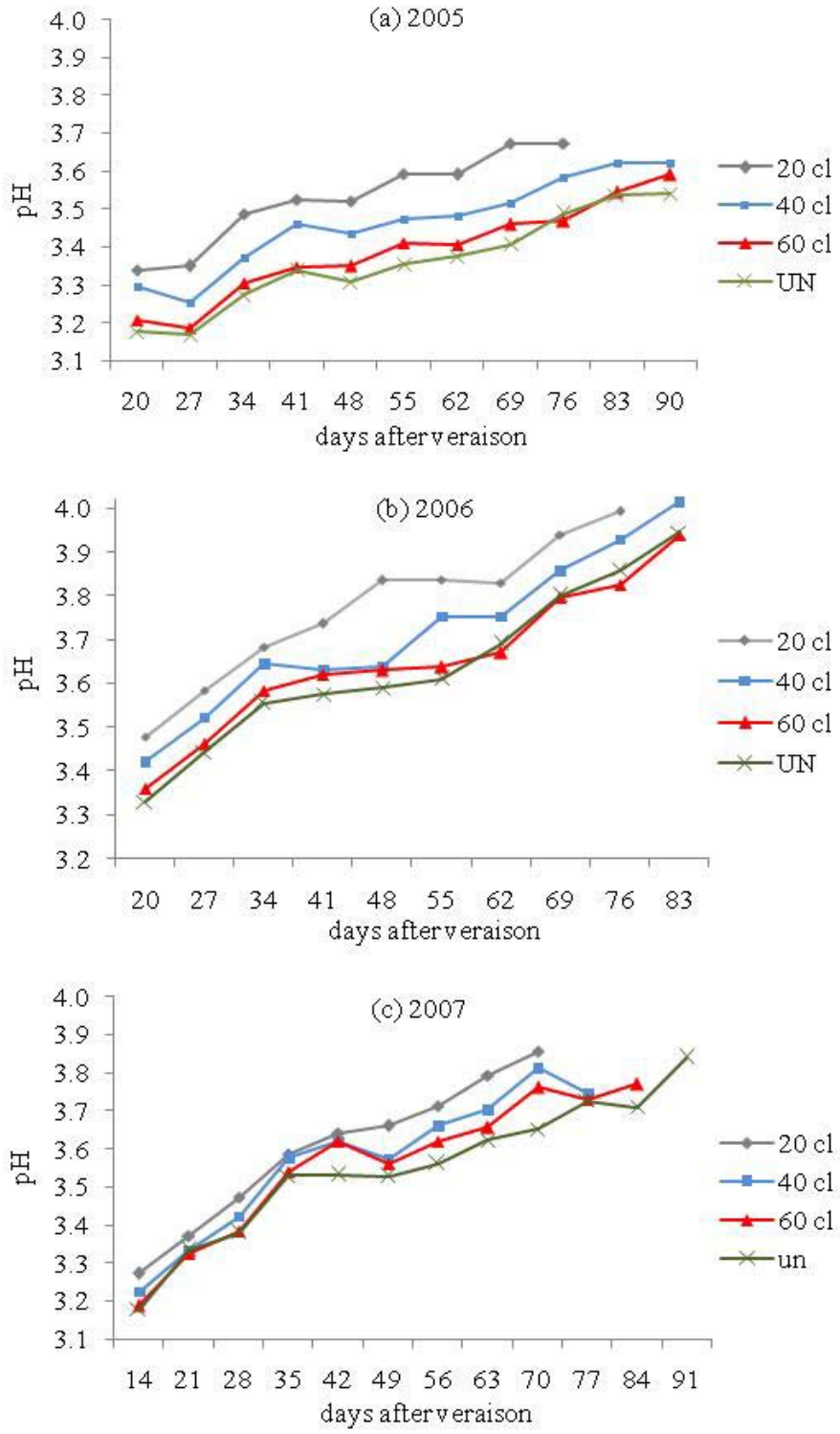


Figure 4.5: Crop load effects on pH development during the ripening period in 2005 (a), 2006 (b), and 2007 (c).

Titratable acidity development: All crop loads decreased in juice titratable acidity (TA) as DAV increased (Figure 4.6). The 20 cl had the highest initial TA measurement each year as opposed to the UN which had the lowest initial TA each year and were statistically different at $p<0.01$, $p<0.001$ and $p<0.001$, for 2005, 2006 and 2007, respectively. The overall trends were consistent across years, although 2005 had the greatest range of initial TA among all crop loads (i.e. between 7.5-10 g/L). The difference in TA between crop loads was most evident in 2005 and was maintained throughout most of the ripening period.

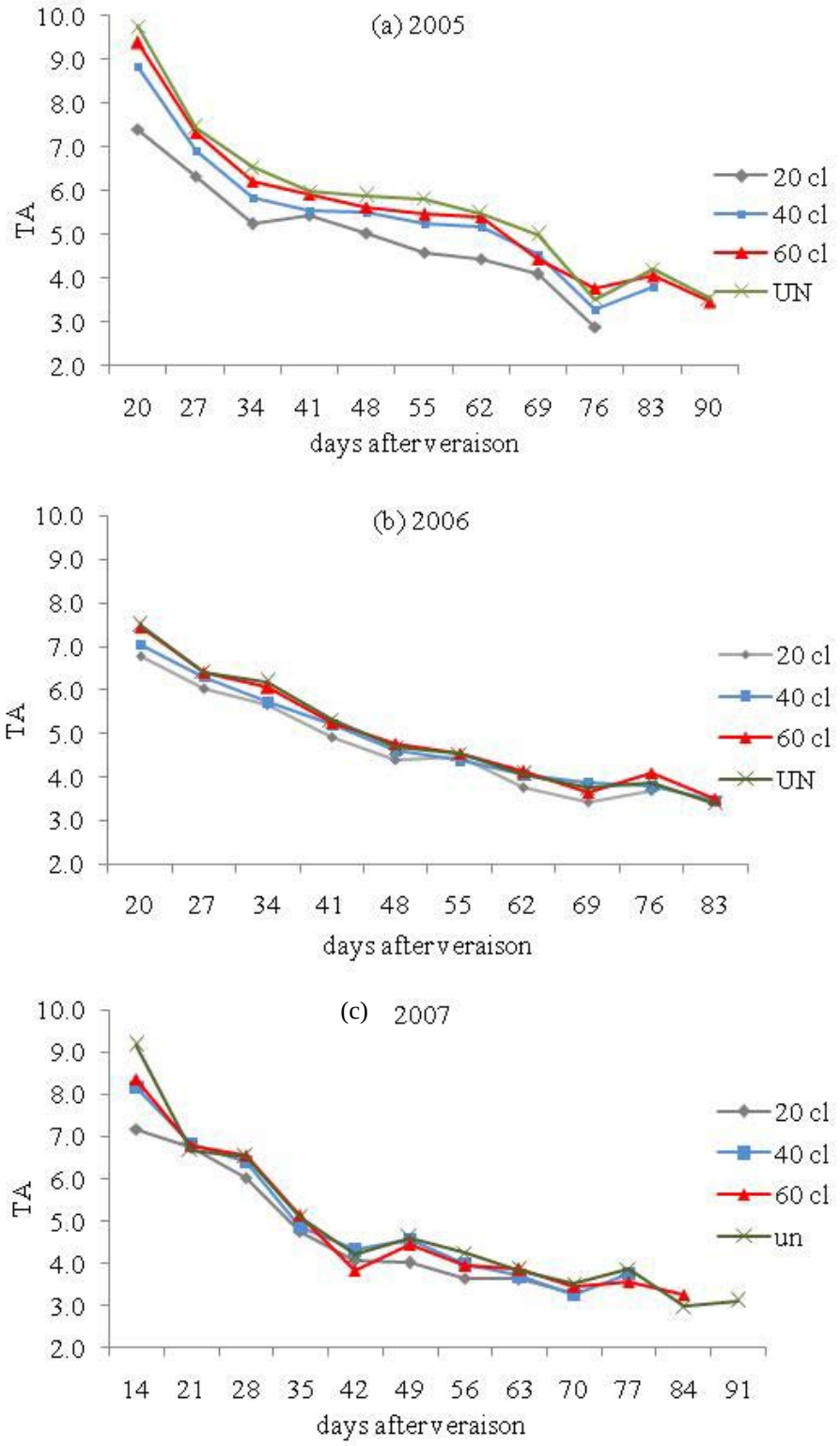


Figure 4.6: Crop load effects on titratable acidity (TA) development during ripening in 2005 (a), 2006 (b), and 2007 (c).

4.3.2 Harvest Juice Chemistry

Degrees Brix: The actual degrees Brix (°Brix) at harvest were significantly different due to both crop load and target °Brix in each year (Table 4.2). Crop load affected °Brix at harvest; however, there was no clear trend among the three years. In 2005, the 20 cl and 60 cl were not significantly different from each other, and the UN had the lowest average °Brix at harvest and was not significantly different from the 40 cl. In 2006 the 20 cl and UN were not significantly different from each other but were significantly different from the 60 cl and 40 cl. The 20 cl was significantly lower than the 60 cl and UN in 2007. The *actual* °Brix at harvest was significantly affected by the *target* °Brix at harvest in each year ($p<0.001$). Certainly, actual °Brix at harvest increased as Target °Brix increased. This is the expected outcome provided that it was part of the experimental design; however, the statistical significance between °Brix at harvest and actual °Brix confirm the validity of the treatment. Another way of perceiving this data is the comparison of DAV required to reach target °Brix in each crop load—this is presented in Table 4.3. A significant treatment interaction occurred in all years ($p<0.001$). Ripening from 22.5 °Brix to 28.5 °Brix required, on average, 56 more DAV. In addition, the average difference between ripening to each target °Brix in the 20 cl and UN was 21 DAV. The average DAV between target °Brix was greater between the 25.5 and 27.0 °Brix target. Most likely this was weather related; such as a cooling trend in early-mid October and shortened day length, thereby requiring more DAV to achieve target °Brix.

Table 4.2: Effect of crop load and target °Brix at harvest on juice soluble solids (°Brix) for three growing seasons.

	°Brix *at harvest*		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	25.1 b	25.6 b	25.1 a
40	24.7 ab	25.2 a	25.3 ab
60	25.1 b	25.2 a	25.5 c
UN	24.6 a	25.5 b	25.4 bc
	*	***	**
Target °Brix			
22.5	22.6 a	23.0 a	22.7 a
24.0	23.8 b	23.9 b	23.8 b
25.5	25.2 c	24.0 c	25.8 c
27.0	25.0 d	26.8 d	26.8 d
28.5	26.9 e	28.1 e	27.6 e
	***	***	***
Interaction	***	***	***

Means with columns separated by different letters differ significantly at p<0.001, 0.01, or 0.05 by LSD . *, **, ***, ns indicate significance at p< 0.05, 0.01, 0.001, or not significant respectively.

Table 4.3: Days after veraison (DAV) required to reach target °Brix at harvest for each crop load during three seasons.

	DAV to target °Brix																			
Crop Load	**2005**					**2006**					**2007**					**Grand Mean**				
	22.5	***24***	***25.5***	***27***	***28.5***	***22.5***	***24***	***25.5***	***27***	***28.5***	***22.5***	***24***	***25.5***	***27***	***28.5***	***22.5***	***24***	***25.5***	***27***	***28.5***
20 cl	23	40	51	60	81	29	33	42	64	78	16	24	29	50	73	**22.7**	**32**	**40.7**	**58**	**77**
40 cl	35	45	56	64	95	30	40	52	72	83	22	29	41	62	83	**29**	**38**	**49.7**	**66**	**87**
60 cl	50	57	64	82	97	35	44	61	79	89	24	34	54	76	90	**36.3**	**45**	**59.7**	**79**	**92**
UN	53	63	71	96	98	37	51	68	84	91	27	37	58	79	93	**39**	**50**	**65.7**	**86**	**94**
Grand Mean	**40**	**51**	**61**	**76**	**92.8**	**33**	**42**	**56**	**75**	**85**	**22**	**31**	**46**	**67**	**85**	**31.8**	**41**	**53.9**	**72**	**88**

pH juice: Juice pH at harvest is presented in Table 4.4. Crop load had a significant effect on pH in 2005 and 2006, but did not affect pH in 2007. The main differences occurred between the low crop loads (20 cl and 40 cl) and high crop loads (60 cl and UN) but did not follow similar trends for the two years. In 2005, the UN had a significantly lower pH than the 20 cl and 40 cl; however, in 2006 pH for the 60 cl was significantly lower than all other crop loads. Degrees

Brix at harvest had a significant effect on pH in all three years ($p<0.001$). As expected, juice pH increased as °Brix at harvest increased. In 2005, there were no significant differences in pH between the 25.5-28.5 °Brix targets.

There was a significant interaction between crop load and °Brix at harvest on juice pH in all years ($p<0.001$). On average, there was a greater change in pH for the lower crop loads during ripening in the three seasons. The 20 cl and 40 cl increased by 0.37 and 0.39 respectively between the lowest and highest pH measurements throughout ripening i.e. 22.5 to 28.5 °Brix targets. In comparison, the 60 cl and UN only increased by 0.24 and 0.26 respectively between the lowest and highest pH during ripening. Generally, these data suggest that pH was more strongly related to target °Brix than to crop load.

Table 4.4: Effect of crop load and target °Brix at harvest on harvest juice pH for three growing seasons.

	pH *juice*		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	3.65 b	3.73 b	3.51
40	3.64 b	3.73 b	3.53
60	3.62 ab	3.67 a	3.52
UN	3.58 a	3.74 b	3.54
	**	***	ns
Target °Brix			
22.5	3.49 a	3.53 a	3.35 a
24.0	3.58 b	3.62 b	3.42 b
25.5	3.66 c	3.73 c	3.49 c
27.0	3.68 c	3.84 d	3.64 d
28.5	3.70 c	3.89 e	3.74 e
	***	***	***
Interaction	***	***	***

Means within columns with different letters differ significantly at $p<0.001$ or 0.01 by LSD. *, **, ***, ns indicate significance at $p< 0.05$, 0.01, 0.001, or not significant respectively.

Titratable acidity in juice: The juice titratable acidity (TA juice) at harvest was significantly affected by crop load, °Brix at harvest and the interaction of the two (Table 4.5). Crop load affected TA juice differently in each of the three seasons. In 2005, there were no significant differences between the 20 cl and 60 cl, although they were significantly different in the subsequent years. Additionally, the 40 cl and UN were similar in 2005 only. The UN had the lowest average TA juice and was significantly lower than all other crop loads in 2006 and 2007. The highest average TA juice was in the 20 cl, averaging 5.25 g/L i.e. 0.56 g/L higher than average TA juice for the UN. In general, TA juice decreased as °Brix at harvest increased and the highest TA was consistently at the lowest target °Brix. Titratable acidity varied among the three seasons. Overall, 2006 and 2007 showed similar patterns in TA juice although in 2007 TA juice decreased as crop load and °Brix at harvest increased. The inconsistency in 2005 may have been a factor of being the first season of treatments or due to the weather conditions within that season.

Table 4.5: Effect of crop load and target °Brix at harvest on juice titratable acidity for three growing seasons.

	TA*juice* (g/L)		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	4.80 a	4.74 b	6.20 a
40	4.13 b	4.61 c	5.55 b
60	4.78 a	5.06 a	5.52 b
UN	4.36 b	4.49 d	5.14 c
	***	***	***
Target °Brix			
22.5	5.32 c	5.55 d	7.26 e
24.0	4.44 b	4.99 c	6.04 d
25.5	4.29 ab	4.45 b	5.77 c
27.0	4.08 a	4.30 a	4.94 b
28.5	4.46 b	4.33 ab	3.99 a
	***	***	***
Interaction	***	***	***

Means with columns separated by different letters differ significantly at p< 0.001 by LSD. *, **, ***, ns indicate significance at p< 0.05, 0.01, 0.001, or not significant respectively.

Alpha amino nitrogen and ammonium: Alpha amino nitrogen (NOPA) and ammonium (NH_4^+) measured on juice at harvest are presented in Table 4.6. In general, the lower crop loads had greater amounts of NOPA. Alpha amino nitrogen decreased as °Brix at harvest increased and was significant in 2005 and 2007. There was a significant interaction between crop load and °Brix at harvest on NOPA in each year. Ammonium was significantly greater as crop load decreased in 2006 and 2007. Ammonium was highest at the 22.5 °Brix target and decreased significantly as °Brix at harvest increased. There was a significant interaction between crop load and °Brix at harvest for ammonium in each year.

Table 4.6: Effect of crop load and target °Brix at harvest on alpha amino acids and ammonium for three growing seasons.

	Alpha amino nitrogen (ppm)			Ammonium (ppm)		
Treatment	**2005**	**2006**	**2007**	**2005**	**2006**	**2007**
Crop load						
20	96.1 c	139.2 b	101.6 b	50.7	81.5 b	81.6 c
40	104.1 c	126.1 ab	108.1 b	51.1	70.5 a	71.5 ab
60	74.5 a	114.6 a	82.6 a	49.8	69.2 a	72.7 b
UN	85.9 b	140.7 b	79.0 a	48.9	70.1 a	65.9 a
	***	*	***	ns	***	***
Target °Brix						
22.5	117.5 d	140.5	135.6 c	64.1 d	88.8 c	96.7 e
24.0	97.8 c	126.5	87.8 b	57.1 c	86.8 c	83.5 d
25.5	95.6 c	125.9	84.2 ab	47.5 b	74.7 b	52.4 a
27.0	75.0 b	122.1	78.3 a	43.7 b	56.3 a	70.3 c
28.5	64.9 a	135.8	78.2 a	38.1 a	57.6 a	61.8 b
	***	ns	***	***	***	***
Interaction	***	***	***	***	***	***

Means within columns with different letters differ significantly at p<0.001 or 0.05 by LSD.
*, **, ***, ns indicate significance at p< 0.05, 0.01, 0.001, or not significant respectively.

Yeast assimilable nitrogen: The yeast assimilable nitrogen (YAN) in juice at harvest was significantly affected by crop load and °Brix at harvest (Table 4.7). The average YAN was greatest in the lower crop loads. In 2005, the 40 cl had a significantly greater YAN than all other crop loads. In 2006 and 2007, YAN was greatest in the 20 cl relative to the higher crop loads. Overall, YAN decreased as °Brix at harvest increased. The lowest YAN was typically at the 28.5 °Brix target. A significant treatment interaction occurred in each year (p<0.001).

Table 4.7: Effect of crop load and target °Brix at harvest on yeast assimilable nitrogen (YAN) for three growing seasons.

	YAN (ppm)		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	146.9 b	221.3 c	181.7 c
40	155.3 b	203.9 b	179.5 c
60	124.3 a	190.6 a	155.2 b
UN	134.9 a	211.2 b	144.9 a
	***	***	***
Target °Brix			
22.5	181.5 d	229.4 d	230.6 d
24.0	154.9 c	214.1 c	171.3 c
25.5	143.2 c	200.7 b	136.4 a
27.0	118.8 b	188.0 a	148.4 b
28.5	103.2 a	201.7 b	139.9 ab
	***	***	***
Interaction	***	***	***

Means within columns with different letters differ significantly at $p < 0.001$ by LSD. *, **, ***, ns indicate significance at $p < 0.05$, 0.01, 0.001, or not significant respectively.

4.3.3 Wine Composition

pH in Wine: Wine pH was affected by crop load and °Brix at harvest in all years except for 2007, in which there were no significant differences due to target °Brix (Table 4.8). Wine pH significantly decreased as crop load increased each year; however, actual differences for the UN relative to 20 cl were only 0.22, 0.03 and 0.08 pH, respectively, for 2005, 2006 and 2007. The highest pH (wine) was consistently in wines made from the 20 cl treatments. The average pH (wine) across all treatments was greatly affected by season. The 2005 season had the lowest average pH (wine) of 3.41 relative to 2006 and 2007 which averaged 3.60 and 3.71, respectively. Wine pH was generally highest at the 28.5 °Brix target, although this was only significant in 2005 and 2006. A significant interaction between crop load and °Brix at harvest occurred in each year.

Table 4.8: Effect of crop load and target °Brix at harvest on wine pH (pH *wine*) measured at press for three growing seasons.

	pH *wine*		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	3.54 c	3.67 c	3.76 b
40	3.38 b	3.60 b	3.71 ab
60	3.40 b	3.60 b	3.69 a
UN	3.32 a	3.54 a	3.68 a
	***	***	**
Target °Brix			
22.5	3.40 ab	3.60 ab	3.70
24.0	3.37 a	3.56 a	3.68
25.5	3.36 a	3.56 a	3.71
27.0	3.43 b	3.63 b	3.71
28.5	3.48 c	3.64 b	3.75
	***	***	ns
Interaction	**	**	***

Means within columns with different letters differ significantly at p< 0.05 by LSD and Duncan's multiple range test. *, **, ***, ns indicate significance at p< 0.05, 0.01, 0.001, or not significant respectively.

Titratable acidity in wine: The titratable acidity of wine (TA wine) measured at pressing is presented in Table 4.9. The TA wine was significantly different between both crop load and target °Brix at harvest. In accordance with the winemaking protocol, must was adjusted to 7.0 g/L for each of the three fermentations per treatment—generally, wines had a TA near 7 g/L. There was a significant interaction each year.

Table 4.9: Effect of crop load and target °Brix at harvest on wine titratable acidity (TA *wine*) at press for three growing seasons.

	TA *wine*		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	6.75 a	6.55 ab	6.85 a
40	7.72 b	6.71 bc	7.07 b
60	6.84 a	6.45 a	7.08 b
UN	7.86 b	6.91 c	7.04 b
	***	***	**
Target °Brix			
22.5	6.89 a	6.38 a	6.94 b
24.0	7.27 ab	6.46 a	6.99 bc
25.5	7.70 b	6.72 b	6.82 a
27.0	7.13 a	6.83 b	7.30 d
28.5	7.46 b	6.87 b	7.00 c
	*	***	***
Interaction	*	***	***

Means within columns with different letters differ significantly at p<0.001, 0.01 or 0.05 by LSD and Duncan's multiple range test at p<0.05. *, **, ***, ns indicate significance at p< 0.05, 0.01, 0.001, or not significant respectively.

Ethanol: The ethanol (ETOH) in wine measured at pressing was significantly influenced by °Brix at harvest in all years and by crop load during the 2005 and 2007 seasons (Table 4.10). These results were expected based on the treatments—particularly target °Brix at harvest—and substantiate the soundness of the experimental design. Ethanol was not significant between crop loads in 2006. Ethanol was greatest in the 20 cl in 2005 and decreased as crop load increased. In 2007, the 20 cl and UN cl were significantly lower than the 60 cl and 40 cl. As expected, °Brix at harvest significantly changed ETOH in all years (p< 0.001) in that ETOH increased as °Brix at harvest increased.

Significant interactions occurred in all years but had differing patterns. In 2005, the 20 cl had the highest initial ETOH at the 22.5 °Brix target and the highest overall ETOH at 28.5 °Brix—

generally, the UN had the lowest ETOH at all °Brix targets. In 2006, ETOH at the 22.5 °Brix target was highest in the 20 cl and lowest in the UN, however UN/28.5 had the highest ETOH at 28.5 °Brix. Finally, in 2007 UN/22.5 had the highest initial ETOH relative to all other crop loads at the 22.5 °Brix target; in contrast with 2005, 20/28.5 had the lowest ETOH in 2007 relative to all crop loads. These differing interactions in ETOH highlight the seasonal impact on ripening and ETOH in subsequent wines.

Although significant differences occurred in all years, the range within each target °Brix was generally small. The greatest increase in ETOH was in 2006, between the 25.5 and 27.0 °Brix which increased by 1.2 % ETOH.

Table 4.10: Effect of crop load and target °Brix at harvest on wine ethanol (ETOH) at press for three growing seasons.

	ETOH		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	14.2 c	13.4	13.4 a
40	13.6 b	13.2	13.5 b
60	13.6 b	13.3	13.5 b
UN	13.3 a	13.4	13.4 a
	***	ns	***
Target °Brix			
22.5	12.7 a	12.0 a	11.8 a
24.0	13.1 b	12.5 b	12.6 b
25.5	13.7 c	13.0 c	13.7 c
27.0	14.2 d	14.2 d	14.4 d
28.5	14.6 e	14.7 e	14.8 e
	***	***	***
Interaction	**	**	***

Means within columns with different letters differ significantly at $p < 0.001$ by LSD. *, **, ***, ns indicate significance at $p < 0.05$, 0.01, 0.001, or not significant respectively.

4.3.4 Wine Color and Phenolics

Total phenols: The total phenols in treatment wines at press (*press*) had significant differences due to both crop load and extended ripening (Table 4.11). The total phenols at press were greatest in the 20 cl in 2005 and 2006. In 2007, total phenols at press were similar even though statistically significant differences did occur mainly between the 20 cl and UN. Degrees Brix at harvest significantly affected total phenols at press in each year ($p<0.001$). Total phenols increased as °Brix at harvest increased, with the greatest total phenols at press consistently in the 28.5 °Brix target. A significant interaction occurred in 2006 and 2007 which demonstrated that although the lower crop loads had higher initial total phenols at the 22.5 °Brix target, the 60 cl and UN had a greater increase due to extended ripening to 28.5 °Brix.

Table 4.11: Effect of crop load and target °Brix at harvest on total phenols A (280 nm) at pressing for three growing seasons.

	Total phenols at *press* A (280 nm)		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	40.56 b	27.02 c	32.51 a
40	36.87 a	23.90 b	33.92 b
60	36.74 a	22.80 a	33.13 ab
UN	35.40 a	23.64 ab	33.96 b
	***	***	***
Target °Brix			
22.5	36.96 b	20.68 a	30.20 b
24.0	33.08 a	22.31 b	29.00 a
25.5	36.64 b	24.34 c	31.25 b
27.0	39.22 bc	26.76 d	35.17 c
28.5	41.06 c	27.75 e	41.28 d
	***	***	***
Interaction	ns	***	***

Means within columns with different letters differ significantly at $p< 0.001$ by LSD. *, **, ***, ns indicate significance at $p< 0.05$, 0.01, 0.001, or not significant respectively.

Total phenols were also measured after the completion of malolactic fermentation to confirm trends shown in the press measurements and to detect any changes that may have occurred during the secondary fermentation (Table 4.12). Overall trends in total phenols post malolactic fermentation (*pml*) were similar to the press measurements with the exception of crop load in 2006. Crop load had a significant effect on total phenols pml in 2005 and 2007. Total phenols pml were lowest in the unthinned in 2005 and 2007. In contrast, the highest total phenols pml were in the 20 cl in 2005, and in the 40 cl in 2007. Worth noting is that total phenols pml were still highest in the 20 cl for 2006, however it was not significant. Degrees Brix at harvest had a significant effect on total phenols pml in all years ($p<0.001$). Total phenols pml significantly increased as °Brix at harvest increased with the greatest amount of total phenols pml at the 28.5 °Brix target. There was a significant interaction in 2006 and 2007 although patterns differed for each. In 2006, the higher crop loads had substantially greater total phenols pml relative to the 20 cl and 40 cl at all target °Brix, with the exception of 20/28.5. In 2007, although the 20 cl had the highest initial total phenols pml, the increase was much less between 22.5 and 28.5 °Brix (i.e. 6.97 nm). In contrast, the 40 cl, 60 cl and UN increased by 14.74 nm, 10.41 nm and 12.60 nm, respectively, with extended ripening to the 28.5 °Brix target.

Table 4.12: Effect of crop load and target °Brix at harvest on total phenols A(280 nm) post-malolactic fermentation (*pml*) for three growing seasons.

	Total phenols *pml* A(280 nm)		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	43.64 c	30.75	39.76 b
40	39.84 b	29.88	40.12 b
60	37.81 ab	30.37	39.47 b
UN	36.99 a	30.50	37.43 a
	***	ns	**
Target °Brix			
22.5	36.30 a	26.87 a	36.56 ab
24.0	37.17 ab	27.17 a	35.32 a
25.5	39.32 bc	29.18 b	38.28 bc
27.0	41.16 c	33.18 c	39.79 c
28.5	43.89 d	35.49 d	46.03 d
	***	***	***
Interaction	ns	***	***

Means within columns with different letters differ significantly at p<0.001 or 0.01 by LSD. *, **, ***, ns indicate significance at p< 0.05, 0.01, 0.001, or not significant respectively.

Color density: Treatment wines were measured at Absorbance (420 nm) and Absorbance (520 nm) to calculate wine color density at both press and post-malolactic fermentation (Tables 4.13, 4.14). The Absorbance (420 nm), Absorbance (520 nm) and color density data showed a clear increase as °Brix at harvest increased. In general, color density increased between the press to pml measurements for both factors, i.e. crop load and °Brix. The Absorbance (520 nm) was consistently higher than its corresponding Absorbance (420 nm) measurement. On average, the highest CD press was in 2007 (7.04 nm), followed by 2005 (6.28 nm). The lowest average color density was in 2006 (5.07 nm). Color density pml followed the same pattern in that CD decreased as crop load decreased.

Table 4.13: Effect of crop load and target °Brix at harvest on A(420 nm), A(520 nm), and color density (CD *press*) at press for three growing seasons.

	A(420) nm *press*			A(520)nm *press*			CD *press* A(420nm)+A(520nm)		
Treatment	**2005**	**2006**	**2007**	**2005**	**2006**	**2007**	**2005**	**2006**	**2007**
Crop load									
20	1.99 a	1.82 a	2.08 a	3.36 a	2.89 a	3.42 a	5.35 a	4.71 a	5.49 a
40	2.15 b	1.93 ab	2.47 b	4.24 b	3.24 bc	4.47 b	6.38 b	5.16 bc	6.94 b
60	2.29 b	1.91 a	2.59 b	4.27 b	3.09 ab	4.71 c	6.55 b	5.00 ab	7.30 b
UN	2.32 b	2.03 b	2.91 c	4.52 b	3.37 c	5.52 d	6.84 c	5.40 c	8.43 c
	***	**	***	***	***	***	***	***	***
Target °Brix									
22.5	1.74 a	1.22 a	1.73 a	3.39 a	1.96 a	2.89 a	5.13 a	3.18 a	4.62 a
24.0	1.89 b	1.65 b	2.09 b	3.81 b	2.78 b	3.78 b	5.70 b	4.42 b	5.88 b
25.5	2.22 c	1.89 c	2.37 c	4.45 c	3.20 c	4.20 c	6.68 c	5.09 c	6.55 c
27.0	2.47 d	2.16 d	2.86 d	4.52 c	3.54 d	5.22 d	6.99 c	5.70 d	8.08 d
28.5	2.60 d	2.69 e	3.52 e	4.31 c	4.25 e	6.56 e	6.91 c	6.94 e	10.08 e
	***	***	***	***	***	***	***	***	***
Interaction	**	***	***	**	***	***	**	***	***

Means within columns with different letters differ significantly at p< 0.001 or 0.01 by LSD. *, **, ***, ns indicate significance at p< 0.05, 0.01, 0.001, or not significant respectively.

Table 4.14: Effect of crop load and target °Brix at harvest on A(420 nm), A(520 nm), and color density (CD *pml*) post-malolactic fermentation for three growing seasons.

	A(420nm) *pml*			A(520nm) *pml*			CD *pml* A(420 nm) + A(520 nm)		
Treatment	**2005**	**2006**	**2007**	**2005**	**2006**	**2007**	**2005**	**2006**	**2007**
Crop load									
20	2.33 b	2.47 a	2.17 a	3.53 a	3.40 a	3.33 a	5.85 a	5.88 a	5.55 a
40	2.01 a	2.47 a	2.79 b	4.06 b	3.57 ab	4.67 b	6.07 a	6.04 ab	7.46 b
60	2.73 c	2.55 a	3.24 d	4.86 c	3.71 b	5.38 c	7.58 b	6.26 b	8.62 c
UN	2.49 b	2.81 b	3.06 c	4.83 c	4.25 c	5.16 c	7.32 b	7.06 c	8.13 c
	***	***	***	***	***	***	***	***	***
Target °Brix									
22.5	1.82 a	2.14 a	1.90 a	3.09 a	2.74 a	2.85 a	4.91 a	4.88 a	4.75 a
24.0	2.12 b	2.25 ab	2.18 b	3.81 b	3.12 b	3.50 b	5.93 b	5.37 b	5.68 b
25.5	2.45 c	2.37 b	2.65 c	4.55 c	3.49 c	4.42 c	6.70 c	5.88 c	7.13 c
27.0	2.66 d	2.84 c	3.18 d	4.91 cd	4.30 d	5.34 d	7.58 cd	7.14 d	8.52 d
28.5	2.88 e	3.29 d	4.17 e	5.25 d	4.99 e	7.06 e	8.13 d	8.28 e	11.23 e
	***	***	***	***	***	***	***	***	***
Interaction	*	***	***	*	***	***	*	***	***

Means within columns separated with different letters differ significantly at p<0.001 by LSD. *, **, ***, ns indicate significance at p< 0.05, 0.01, 0.001, or not significant respectively.

Hue: Wine hue was significantly affected by crop load and °Brix in each year (p<0.001) and at each stage measured. Generally, hue at press (Hue press) and post malolactic fermentation (Hue

pml) decreased as crop load increased—this is shown in Tables 4.15 and 4.16, respectively. Moreover, hue was consistently highest in the lowest crop load. The trends in hue due to °Brix were different between the press and pml measurements. Hue press was lowest at the 24.0 and 25.5 °Brix targets in 2005 and 2006, but lowest at 28.5 °Brix in 2007. Hue pml showed no significant differences among wines harvested between the 24.0 and 28.5 °Brix target in 2005, however in 2006 and 2007 wines at the 27.0 and 28.5 °Brix target were significantly lower compared with the other °Brix targets. There was a significant interaction between crop load and °Brix on hue at press and pml in each year ($p<0.001$).

Table 4.15: Effect of crop load and target °Brix at harvest on wine hue at press (Hue *press*) for three growing seasons.

	Hue *press*		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	0.60 c	0.64 b	0.62 c
40	0.54 ab	0.60 a	0.57 b
60	0.55 b	0.63 b	0.56 ab
UN	0.52 a	0.60 a	0.54 a
	***	***	***
Target °Brix			
22.5	0.55 a	0.63 bc	0.60 c
24.0	0.53 a	0.60 a	0.57 b
25.5	0.53 a	0.60 a	0.58 b
27.0	0.55 a	0.62 b	0.55 a
28.5	0.61 b	0.64 c	0.55 a
	***	***	***
Interaction	***	***	***

Means within columns with different letters differ significantly at $p<0.001$ by LSD. *, **, ***, ns indicate significance at $p< 0.05, 0.01, 0.001$, or not significant respectively.

Table 4.16: Effect of crop load and target °Brix at harvest on wine hue post malolactic fermentation (Hue *pml*) for three growing seasons.

	Hue *pml*		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	0.67 c	0.75 c	0.67 c
40	0.50 a	0.70 b	0.62 b
60	0.57 b	0.71 b	0.61 ab
UN	0.52 a	0.67 a	0.60 a
	***	***	***
Target °Brix			
22.5	0.60 b	0.78 d	0.67 d
24.0	0.57 a	0.72 c	0.64 c
25.5	0.55 a	0.69 b	0.62 b
27.0	0.55 a	0.67 a	0.60 a
28.5	0.56 a	0.66 a	0.60 a
	***	***	***
Interaction	***	***	***

Means within columns with different letters differ significantly at $p < 0.001$ by LSD. *, **, ***, ns indicate significance at $p < 0.05$, 0.01, 0.001, or not significant respectively.

Total anthocyanins: The total anthocyanins, measured post malolactic fermentation, increased significantly as °Brix at harvest increased in all years ($p<0.001$) (Table 4.17). In general, anthocyanins were highest for the UN and 60 cl—although it was not significant. Total anthocyanins on average, were lowest in 2005 which averaged 347 mg/L relative to 2006 and 2007 which were similar at 381 and 376 mg/L, respectively.

Table 4.17: Effect of crop load and target °Brix at harvest on total anthocyanins (mg/L) for three growing seasons.

	Total anthocyanins (mg/L)		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	328	379	340
40	340	374	388
60	360	383	390
UN	361	389	387
	ns	ns	ns
Target °Brix			
22.5	270 a	288 a	298 a
24.0	343 bc	332 b	339 b
25.5	328 b	378 c	394 c
27.0	379 cd	442 d	414 cd
28.5	417 d	467 d	437 d
	***	***	***

Means with columns with different letters differ significantly at $p < 0.001$ by LSD. *, **, ***, ns indicate significance at $p < 0.05$, 0.01, 0.001, or not significant respectively.

Tannins: Crop load affected tannins, measured after malolactic fermentation, in all years (Table 4.18). Tannins were highest in the 20 cl relative to the other crop loads each year. In 2007, both the 20 cl and 40 cl were significantly higher than the 60 cl and UN. There were no significant differences in tannins due to target °Brix at harvest. The relationship between tannins and clusters per vine (cl/vine) is presented in Figures 4.7 a, b, c. Tannins significantly decreased as cl/vine increased in each year however there was a large seasonal affect. Tannins averaged 780 mg/L, 361 mg/L and 664 mg/L for 2005, 2006 and 2007, respectively.

Table 4.18: Effect of crop load and target °Brix at harvest on tannins (mg/L) for three growing seasons.

	Tannins (mg/L)		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	985 b	455 b	798 b
40	753 a	341 a	712 b
60	767 a	336 a	607 a
UN	615a	311 a	539 a
	**	***	***
Target °Brix			
22.5	846	380	714
24.0	724	346	639
25.5	732	324	665
27.0	815	363	629
28.5	783	391	675
	ns	ns	ns

Means within columns with different letters differ significantly at p<0.001 or 0.01 by LSD. *, **, ***, ns indicate significance at p< 0.05, 0.01, 0.001, or not significant respectively.

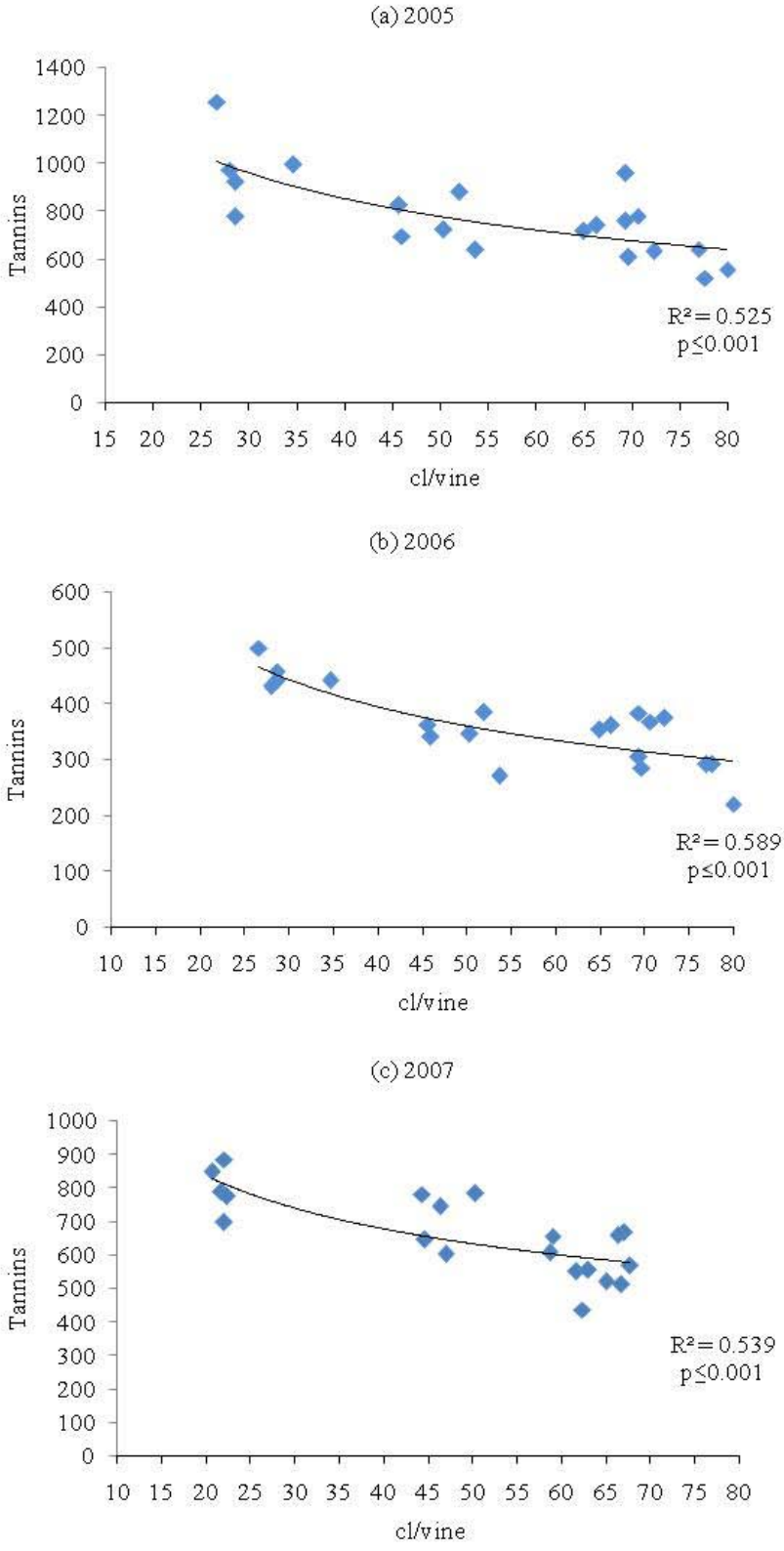


Figure 4.7: The relationship between tannins (mg/L) and clusters per vine (cl/vine) in three seasons.

Methoxypyrazines: The isobutylmethoxypyrazines (IBMP), measured in wine after six months of aging in neutral barrels, had significant differences due to both crop load and °Brix at harvest (Table 4.19). The 20 cl had a significantly greater concentration of IBMP in 2006 compared with the other crop loads. IBMP concentration decreased as °Brix at harvest increased. There were no significant interactions. In 2006, the initial levels of IBMP were highest in the 20 cl, followed by the 40 cl and lowest in the 60 cl and UN (Figure 4.8). Additionally, IBMP for the 20 cl was higher at all target °Brix relative to all other crop loads. In addition, the 40 cl was intermediate at all °Brix targets except 28.5 °Brix. The relationship between IBMP and DAV for all treatments in 2006 is presented in Figure 4.9 and certainly illustrates that IBMP reduced with DAV. However, the lower crop loads had more IBMP to break down. Provided that ≤3.0 ppt of IBMP is the target upper threshold for particular Cabernet Sauvignon styles (as indicated by the dotted grey line in Figures 4.8 and 4.9), the 20 cl and 40 cl would have required ripening to at least 28.5 °Brix in order to reduce to or below the threshold. In contrast, the 60 cl and UN reduced to 3.0 ppt IBMP by ripening to only 25.5 °Brix.

Table 4.19: Effect of crop load and target °Brix at harvest on isobutylmethoxypyrazine (IBMP) (ppt) concentrations in wine for growing season 2006.

	IBMP (ppt)
Treatment	**2006**
Crop load	
20	5.52 b
40	3.86 a
60	2.94 a
UN	3.02 a
	***
Target °Brix	
22.5	5.26 c
24.0	4.30 bc
25.5	3.94 ab
27.0	2.81 a
28.5	2.86 a
	**
Interaction	ns

Means within columns with different letters differ significantly at p<0.001 or 0.01 by LSD. *, **, ***, ns indicate significance at p< 0.05, 0.01, 0.001, or not significant respectively.

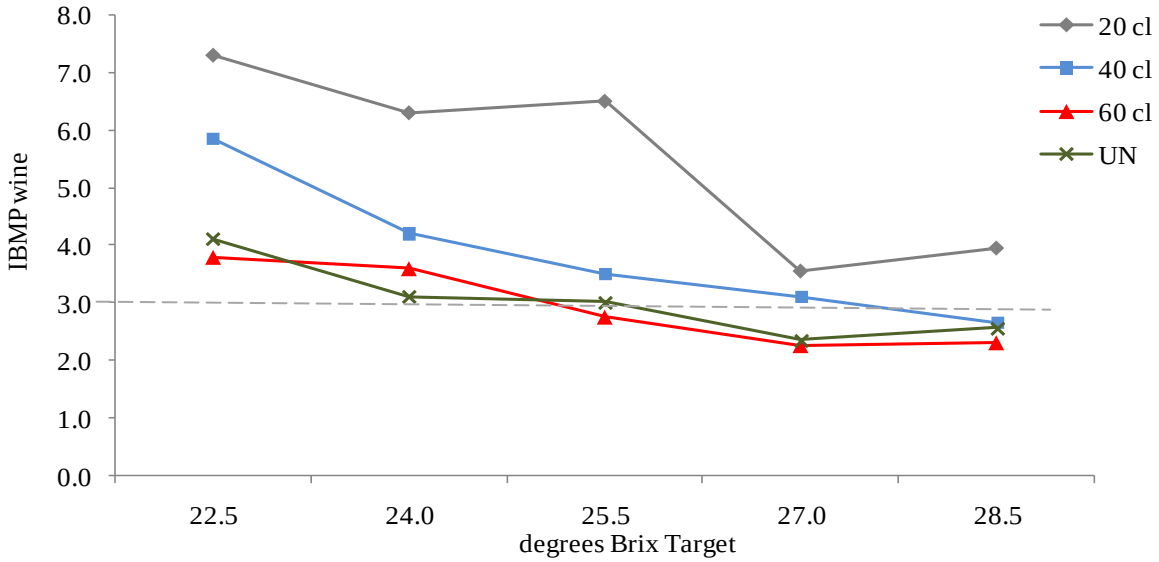


Figure 4.8: The comparison of crop load and target °Brix at harvest on isobutylmethoxypyrazines (IBMP) in wine for the 2006 season. The grey dotted line implies an optimal threshold for IBMP.

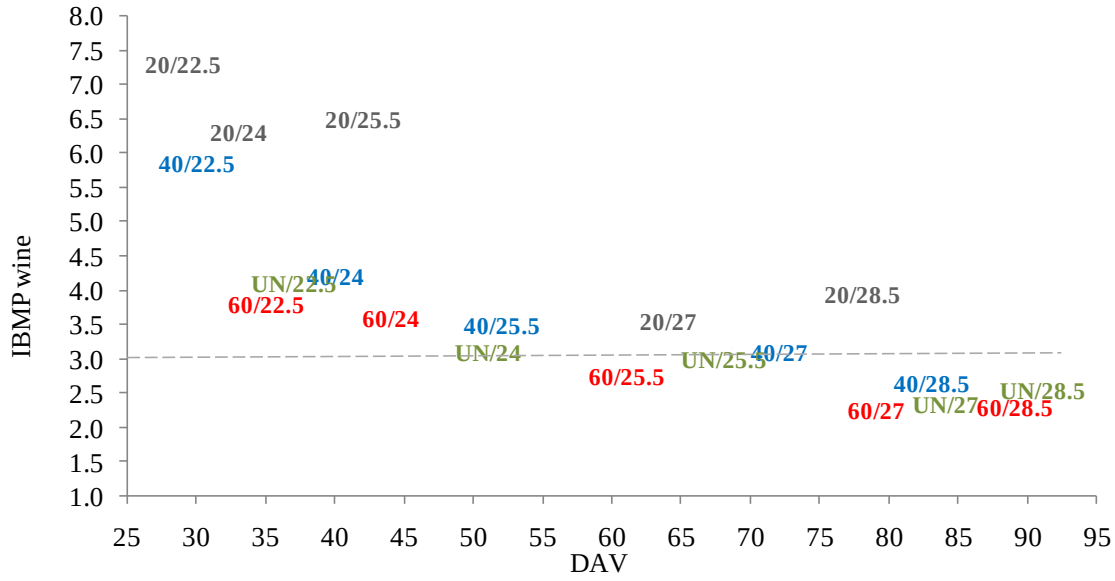


Figure 4.9: The comparison of isobutylmethoxypyrazine concentrations (IBMP) ppt in each treatment relative to days after veraison to reach the target °Brix at harvest for the 2006 season.

4.4 Discussion

4.4.1 Treatment Effects on Berry Development

Sugar accumulation: Crop load had a major effect on °Brix accumulation in each year. As crop load increased, the DAV required to reach each °Brix target increased—signifying a change in the rate of sugar ripening due to crop load. This pattern concurs with the work of Guidoni *et al.* (2002) who reported that a 50 % cluster removal at pea size increased berry soluble solids by 7 % at harvest. In addition, Gu and Wample (2006) reported that crop thinning increased the rate of sugar accumulation and therefore advanced harvest by 24 and 35 days for hand and machine pruned vines, respectively. Additional studies on crop load demonstrated that soluble solids were increased by reduced crop level (Ough and Nagaoka 1984, Bravdo *et al.* 1985, Reynolds *et al.* 1994, Ford 2007).

Petrie *et al.* (2000) reported that reduced leaf area delayed ripening. In the current experiment, it is plausible that increased shoot growth and leaf area (discussed in Chapter 3) aided in faster ripening due to a greater photosynthetic capacity and may explain in part the faster rate of sugar accumulation in the lower crop loads. However, Smart *et al.* (1985 and 1988) demonstrated that increased shading in the canopy microclimate markedly reduced sugar ripening. These results suggest that canopy microclimate can significantly alter resulting fruit and wine composition.

In contrast to °Brix accumulation, in 2007 the lower crop loads (particularly the 20 cl), began to decrease in sugar (g)/berry earlier than the UN (Figure 4.10). The average DAV required to reach peak sugar (g)/ berry in the 20 cl, 40 cl, 60 cl and UN were 41, 51, 79 and 81 DAV, respectively. In 2007, the 20 cl reduced in sugar (g)/berry near 28 DAV while the UN sustained sugar (g)/berry past 56 DAV. These results suggest that after 28 DAV, the 20 cl increased in °Brix largely due to concentration of sugars by way of berry dehydration rather than continued sugar transport from the vine into the berry. A study by Coombe (1987) on distribution of solutes within the developing berry stated that loss of the solvent (water) can increase solute concentration within the berry. The present results suggest that higher crop loads most likely continued to transport sugar into the berry past 56 DAV and early °Brix differences between crop loads were likely caused by actual differences in photosynthetic capacity. Conversely, differences in the later stages of ripening may have been partially or exclusively due to a concentration effect in the lower crop loads—as indicated by the sugar (g)/berry curves in 2007. Moreover, the work of Sanchez *et al.* (2006) demonstrated that the rise in soluble solids over time was also accompanied by a 6 % decrease in berry moisture for Merlot grapes irrigated at two different levels during the later stages of ripening. Coombe and McCarthy (2000) showed

that increased sugar/berry in Shiraz was due to decreased berry volume, caused by impeded phloem transport, and consequently increased the solute concentration within the berry.

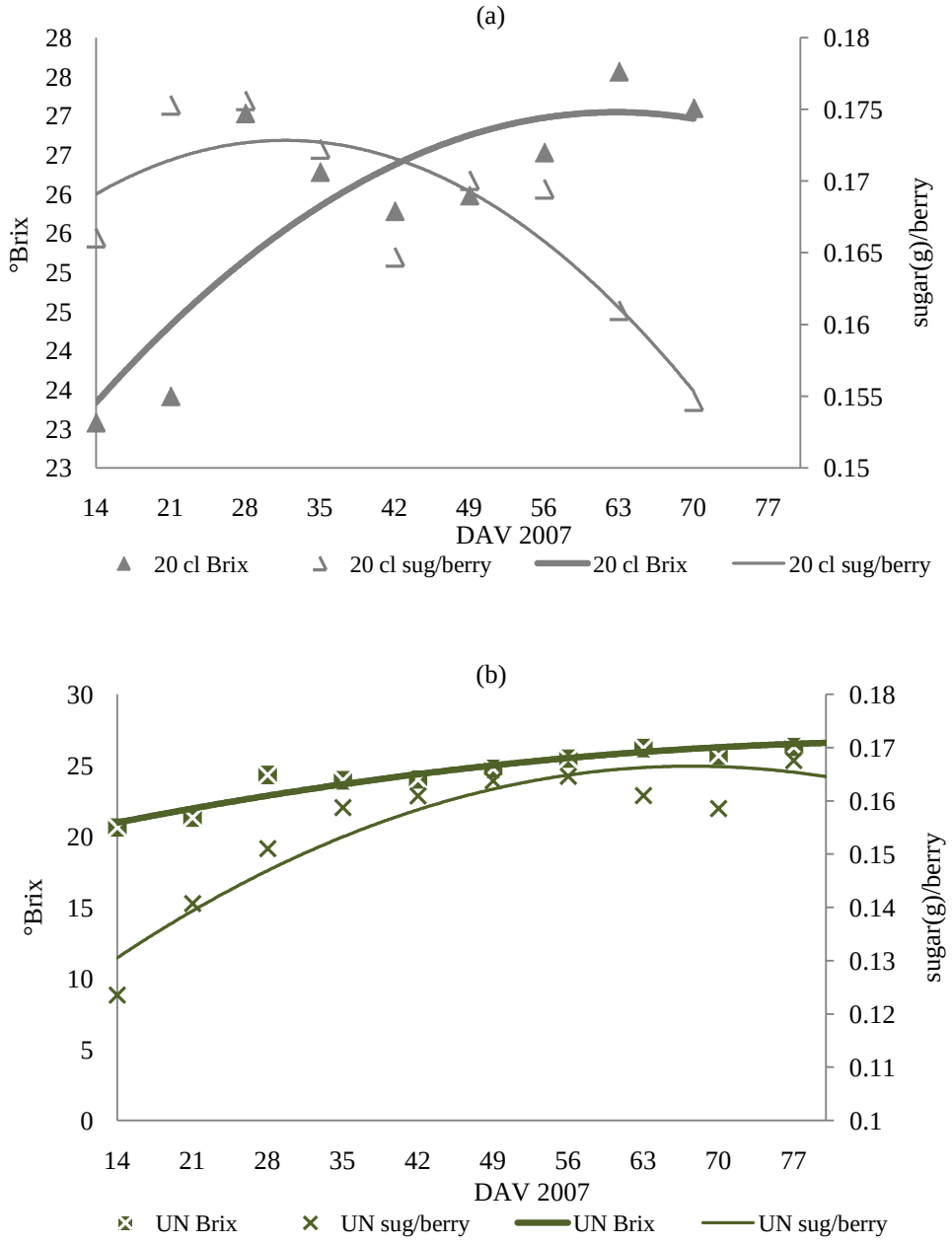


Figure 4.10: The comparison of °Brix accumulation and sugar (g)/berry in the 20 cl and UN in 2007.

Figure 4.11 illustrates the seasonal differences in rate of °Brix accumulation between 2005 and 2007 for the 20 cl and UN. Both the 20 cl and UN had a higher initial °Brix in 2007 at 14 DAV relative to 2005. The UN in 2005 exhibited a parallel pattern to that of 2007, but still had lower °Brix relative to 2007 throughout the entire season. This is further demonstrated in that both the 20 cl and UN reached 'traditional ripeness' of 24.0 °Brix substantially later in 2005 than in 2007. These differences in °Brix accumulation may be explained by yield differences due to changes in soil water supply from varying winter rainfall. The 2005 season had above average rainfall and consequently above average yield for the region. In contrast, 2007 had below average rainfall and yields. These results indicate that overall yield and ripening can be largely affected by seasonal characteristics—particularly winter rainfall.

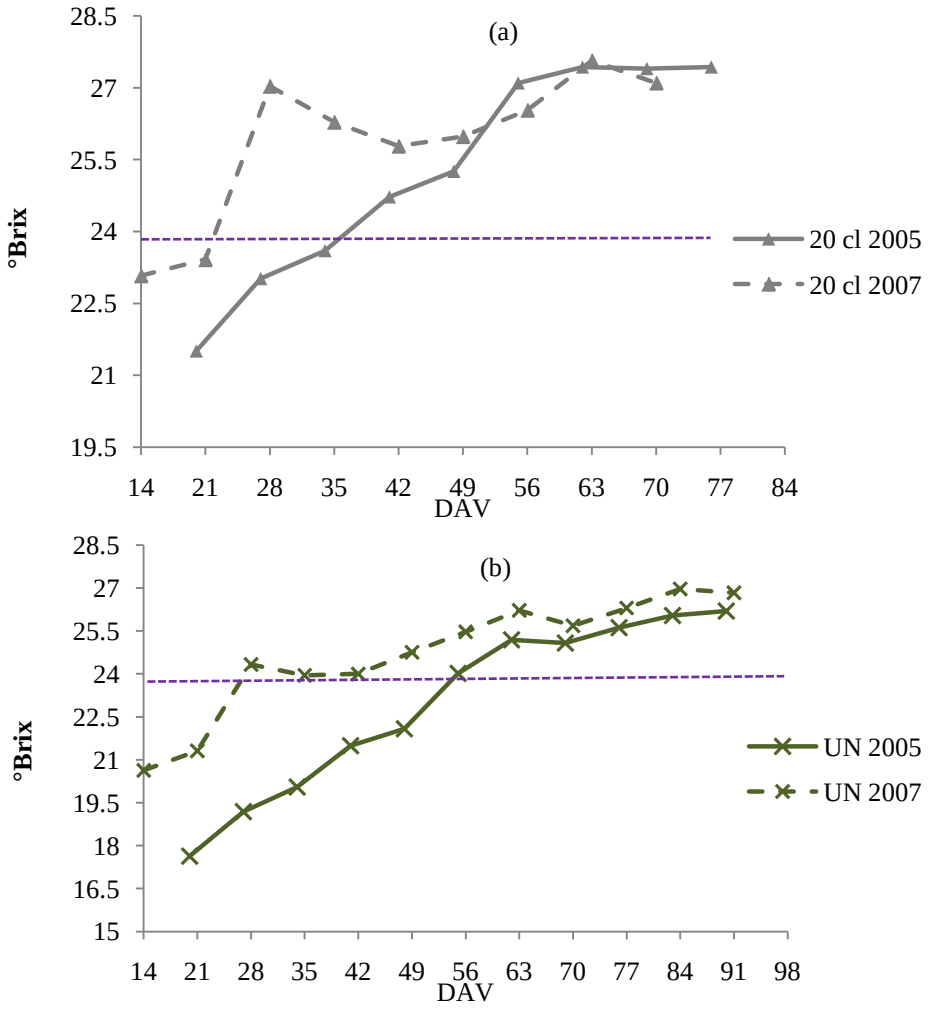


Figure 4.11: The seasonal comparison of °Brix accumulation relative to days after veraison for 2005 and 2007, in the 20 cl (Figure 4.11 a) and UN (Figure 4.11 b).

pH development: The pH in must and wine is the true concentration of hydroxonium ions also known as true acidity (Ribéreau-Gayon *et al.* 2006). The pH development indicated that higher pH corresponded with lower crop loads and increased DAV. The initial pH values were consistently inversely related to crop load; this pattern generally continued throughout ripening even though pH increased overall for all crop loads. The pH development is consistent with studies by Ough and Nagaoka (1984) which showed that pH increased with crop reduction, and

Hernandez-Orte (1999), Gu and Wample (2006) who reported that pH increased with ripening in Tempranillo.

Presumably the differences in pH due to crop load were largely due to changes in TA since pH and TA corresponded in their patterns between crop load, DAV and °Brix. The pH was highest overall in 2006 compared with 2005 and 2007, indicating the lowest acid year was in 2006. Perhaps the weather conditions caused an earlier reduction in acid or, less acid was initially present in the berries. Malic acid decline is very temperature dependent (Coombe and Iland 1987). The value of pH quite accurately corresponds with impressions in wine due to acidity described as 'freshness, greenness or thinness' (Ribéreau-Gayon *et al.* 2006). The differences in pH due to crop load and ripening may have influenced certain sensory properties and this is discussed in Chapter 5.

Titratable acidity development: Overall, titratable acidity (TA) decreased at a similar rate in all crop loads as DAV increased. The initial measurement of TA showed an effect from crop load. As crop load increased the initial TA measurement increased. The initial TA measurement was consistently highest in the UN and lowest in the 20 cl—TA generally had an inverse relationship with crop load, although differences were small in 2006. Evaluation of TA began at 14 and 20 DAV. Perhaps starting measurements earlier (i.e. at veraison) would have provided a more complete indication of differences in acidity due to crop load.

Presumably, the 20 cl which had a faster rate of sugar accumulation started its malic acid reduction earlier (i.e. fewer DAV) relative to the higher crop loads. Grape maturation is marked by an increase in the respiratory quotient (Lamikanra *et al.* 1995), which suggests the use of malic acid for energy production in the grape after veraison (Kliewer 1969, Steffan and Rapp

1979, Mullins *et al.* 1992) and particularly between 10-17 °Brix (Coombe 1987). Additionally, developing berries decline in malate by proportions comparable to the degree of increase of glucose and fructose concentrations (Coombe 1987, Bindon *et al.* 2008a). These reports support the notion that changes in crop load significantly affected berry development and specifically the rate of ripening and acid development. These results are further supported by the TA data in harvest juice chemistry.

The 2005 season had the largest differences in TA among all crop loads. This may have been influenced by the large yields overall, and/or a factor of the first season of the experiment. Furthermore, the 2005 yields averaged 10 kg/vine relative to 2006 and 2007 which averaged six and five kg/vine, respectively—and equates to a 40 % and 52 % yield reduction relative to 2005. In addition, initial TA was lowest on average for all crop loads (i.e. 7.2 g/L) in 2006 relative to 2005 and 2007 which averaged 8.9 g/L and 8.3 g/L, respectively.

There was a strong significant relationship between °Brix and pH, and °Brix and TA each year (Figure 4.12) signifying that pH and TA development are related to °Brix accumulation. As °Brix at harvest increased, pH increased and TA decreased for all years ($p<0.001$). These results are consistent with previous work by: La Rosa and Nielson (1956) Winkler *et al.* (1974), Coombe (1975), Coombe and Iland (1987), Hamilton and Coombe (1992), Jackson and Lombard (1993) and Kennedy (2004).

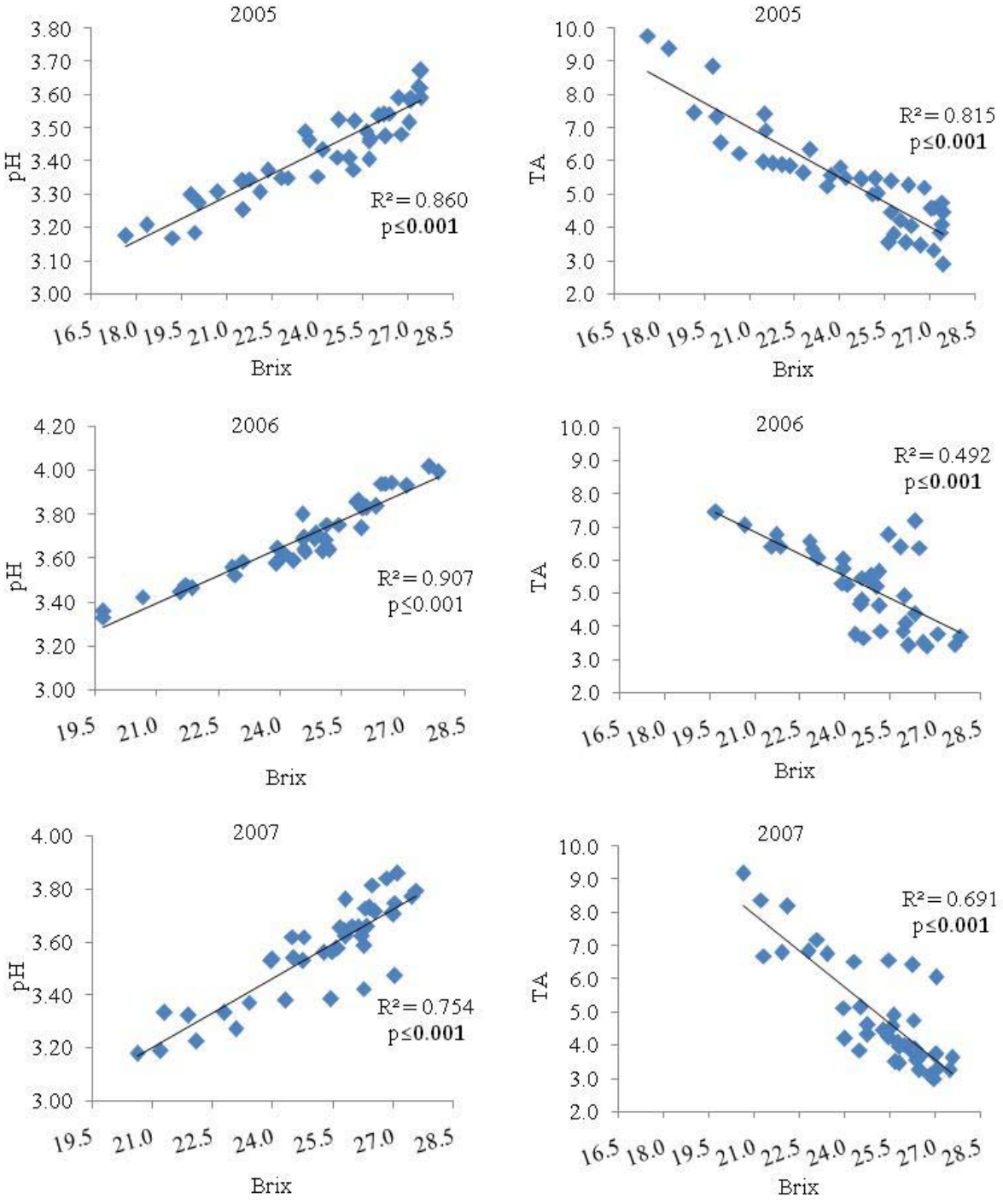


Figure 4.12: Regression analysis of pH and °Brix, and TA and °Brix for 2005-07.

4.4.2 Treatment Effects on Harvest Juice Chemistry

Degrees Brix: There were no uniform trends in °Brix at harvest due to crop load *per se*. Perhaps the statistical differences in crop load were a result of the interaction between crop load and Target °Brix. It should be noted that the field measurement of °Brix varies considerably depending on environmental factors such as ambient temperature, humidity and vapor pressure deficit within the micro and mesoclimate of the grapevine (*personal observation*). Additionally, °Brix in must often increases after 12-24 hours due to the release of sugars from raisin berries which do not immediately release all sugar and thus do not influence °Brix measurements taken just after crushing *(personal observation).* Finally, the earlier harvests (i.e. 22.5 and 24.0 °Brix targets) contained more variability among berries within individual clusters, therefore increasing the sampling error (*personal observation*). These factors may explain why the °Brix at harvest did not always match the target °Brix in all cases. There were significant differences in actual °Brix at each Target °Brix in all years. The actual °Brix increased as target °Brix increased, but the DAV to reach the target °Brix was dependent on crop load. This data concurs with sugar accumulation curves presented in berry development (4.3.1). Although trends were similar, the actual °Brix measurements were largely affected by season. For example, 2005 only reached 26.9 °Brix at the 28.5 °Brix target with 93 DAV of ripening time. In 2006 and 2007, the 28.5 °Brix target achieved 28.1 and 27.6 °Brix, respectively, both requiring 85 DAV. Furthermore, these data confirm that season had a major impact on °Brix accumulation.

There was a significant interaction in all years although trends were dissimilar between years. The interaction may have been, in part, a result of changes in the rate of ripening and the DAV to each Target °Brix. The 2005 season was the slowest ripening year in which the 20 cl had the

greatest percent increase (22 %) in °Brix from the first harvest at the 22.5 °Brix target to the final harvest. In contrast, the 20 cl had the lowest % increase relative to the other crop loads in 2006 and 2007, but required the least DAV to reach the final °Brix target. Although each crop load was harvested at the same Target °Brix, the DAV to reach each target °Brix were quite different. This should be considered when interpreting the interactions between crop load and Target °Brix on *actual* °Brix at harvest.

Generally, increased crop load or over cropping is thought to delay sugar maturity (Kliewer and Weaver 1971,Winkler *et al.* 1974, Jackson and Lombard 1993); however, sugar concentration may be unaffected if vines have the same leaf area to fruit ratio as demonstrated by Zamboni *et al.* (1996). Therefore the large differences in DAV to reach target °Brix between crop loads may be explained by differences in canopy growth and leaf area. Although studies have demonstrated that crop thinning accelerates the rate of sugar ripeness (Winkler 1954, Kliewer and Weaver 1971, Keller *et al.* 2005), 'flavor ripeness' does not always synchronize with sugar ripeness.

pH at harvest: Generally, pH at harvest had significant differences due to crop load, but no consistent trend existed between the three years. This lack of consistency suggests that changes in pH were not necessarily a result of crop load. Possibly crop load only had an indirect effect on pH. The 2005 season was a slow ripening year due to higher than average crop loads. These factors may have attributed to the minor changes in pH among crop loads. The 20 cl and 40 cl started with the lowest overall pH. Furthermore, the largest increase in pH, relative to all crop loads was in the 20 cl and 40 cl which required less time (DAV) to achieve sugar ripeness. Therefore, the interaction between crop load and °Brix indicates that pH increased more rapidly,

and with less DAV in the lower crop loads—suggesting that increases in pH were more associated with the rate of sugar ripening, as influenced by crop load.

Red wine quality has been shown to be reduced by high pH levels (Somers 1975, Boulton 1980) partially due to dull color (Somers 1975, 1977). The optimal range for pH in red wines is generally considered to be between 3.3-3.7 (Mpelasoka 2003); however, the pH optimum at harvest for Cabernet Sauvignon intended for commercial brands at J. Lohr Winery is between 3.6 and 3.7 pH (S. Peck, 2008 personal communication). Optimum pH was achieved at different target °Brix in each of the three seasons. In 2005, optimal pH was achieved close to 25.5 °Brix in the lower crop loads and at 28.5 °Brix in the higher crop loads. In 2006, all crop loads were close to optimal at the 25.5 °Brix target compared with 2007, in which all crop loads were optimal between the 27.0 to 28.5 °Brix targets.

This lack of consistency in pH between years concurs with previous research with opposing outcomes of crop load effects on pH. Reynolds *et al.* (1994) reported increased pH with crop level reduction. Other work by Sinton *et al.* (1978) has shown pH to decrease with increasing crop load. Finally, the research of Zamboni *et al.* (1996) indicated that vines with 30 and 50 buds/vine had no differences in pH or TA; however, both treatments had the same leaf to fruit ratio. The results from my experiment and others confirm that pH is affected by many factors in addition to crop load, ripening and season. Finally, although TA of must is commonly adjusted during primary fermentation—subsequently affecting pH—initial pH of fruit and juice remains an important indicator of ripeness (Ribereau-Gayon 2006).

Titratable acidity: The effects of crop load on TA juice indicated that TA juice was consistently highest for the 20 cl. The UN had the lowest average TA juice and a significantly lower TA

relative to all other crop loads in 2006 and 2007. The TA development curves indicated that crop loads followed a similar pattern with respect to TA juice, particularly for the earlier measurements and suggest that crop reduction increased juice TA.

Shoot growth and leaf area index increased as crop load decreased (illustrated in Chapter 3) thereby increasing the photosynthetic capacity of the vines and may have caused differences in organic acid accumulation. Tartaric and malic acid represent on average 90 % of the organic acids in grapes and are synthesized in the leaves and grapes and mainly produced in grapes prior to *veraison* (Ribéreau-Gayon *et al.* 2006). Malic acid is formed by leaves and young green grapes during the dark phase of photosynthesis. Ribéreau-Gayon *et al.* (2006) stated that during the herbaceous growth phase (prior to veraison) sugars accumulating from photosynthesis are transformed into malic acid. The growth data indicated increased growth in the lower crop loads and decreased PAR which thereby suggests that increased shading in the fruiting zone may have attributed to increased TA for the lower crop loads. This is supported by the work of Dokoozlian (1990) who reported that shading during stage III of berry development decreased malate degradation. Alternatively, higher TA in the lower crop loads may have resulted from less DAV to reach target °Brix and therefore less time for acid loss relative to the 60 cl and UN. The previous studies and my trends in TA suggest that crop load had an indirect effect on TA by affecting the accumulation of malic and tartaric acid through changes in both photosynthetic capacity of the vine, light environment within the fruiting zone, and amount of time required to reach target °Brix. Although TA juice was lower in the higher crop loads in 2006 and 2007, pH juice did not necessarily follow the expected pattern of increasing as TA decreased. This may have been due to different levels of potassium (K^+) in berries at harvest. Higher pH values are associated with high K^+ values (Mpelasoka *et al.* 2003). Potassium neutralizes organic acids and

has an important role in controlling acidity and pH of fruit juice (Hepner and Bravdo 1985, Mullins 1992). Cultural practices affect the partitioning of K^+ within the vine and its distribution from other organs to the fruit (Smart *et al.* 1985a,b). Furthermore, crop load reduction has been associated with increased K^+ (Hepner and Bravdo 1985). Although K^+ in must and wine was not measured, changes in K^+ due to crop load may have affected titratable acidity and pH.

Titratable acidity clearly decreased as °Brix at harvest and DAV increased. This trend is supported by the well known reduction in TA as sugar ripening occurs (Winkler *et al.* 1974, Mullins *et al.* 1992, Dry and Coombe 2004). Additionally, Keller *et al.* (2005) demonstrated a strong relationship between TA and soluble solids in which TA decreased as soluble solids increased in Cabernet Sauvignon that was crop thinned at different phenological stages.

Both the TA development curves and TA juice emphasize that average TA and its changes with ripening varied each season. The 2005 season was characterized by slower sugar accumulation, and had the lowest average TA juice (4.52 g/L) but the highest initial measurements of TA in the development curves. In contrast, 2007 had the highest average TA juice (5.6 g/L) and the fastest sugar accumulation. Ribéreau-Gayon *et al.* (2006) stated that at maturity, the sum of tartaric and malic acid is highly variable, depending on vintage conditions. Furthermore, data from my experiment suggest that changes in TA were predominantly associated with target °Brix at harvest followed by crop load, and season.

Yeast assimilable nitrogen: Overall, the trends in NOPA and ammonium in harvest juice were quite similar although NOPA was consistently greater than ammonium. Amino acids are known to be the most prevalent form of total nitrogen in grape juice and wine, representing 50-90 % of the total nitrogen (Ribéreau-Gayon *et al.* 2006).

In this experiment the combination of alpha amino nitrogen and ammonium make up the yeast assimilable nitrogen (YAN) measurement. Ammonium ions and alpha amino nitrogen compounds are preferentially used by the principal yeast, *Saccharomyces cerevisiae,* for fermentation and are present in the form of primary amino acids (Monteiro and Bisson 1991b, Henschke and Jiranek 1993, Jiranek *et al.* 1995a). Must nitrogen composition and concentration have both direct and indirect effects on wine composition—impacting the resulting wine both negatively and positively (Bell and Henschke 2005). The highest initial YAN was consistently in the lower crop loads and at the 22.5 °Brix target. Kliewer and Ough (1970) reported that concentrations of the nitrogenous compounds arginine, proline, total free amino acids and total nitrogen in berry juice—greatly increased with crop reduction: this was correlated with increased leaf area per vine. Vines which underwent crop reduction had more alpha amino nitrogen and ammonium available to the fruit. Leaf area and shoot growth increased with crop reduction, and perhaps the increased photosynthetic capacity of the vines allowed a greater translocation of amino acids and ammonium into the fruit—resulting in increased YAN for the lower crop loads. YAN decreased as °Brix increased in all crop loads—signifying that DAV and ripening significantly affected YAN. In fact, there was a strong negative correlation between YAN and DAV each year (Table 4.20).

The relationship between must amino acid concentration and its organic acids is well known: the most acidic grapes are always the richest in amino acids (Ribéreau-Gayon *et al.* 2006). This relationship is further supported by the data in this experiment. Overall, lower crop loads had higher YAN and higher TA juice. In addition, the lowest °Brix targets i. e. 22.5 had the highest YAN and TA juice. In theory, increased YAN for the lower crop loads and °Brix targets would have improved its fermentability, even though the relationship between YAN and wine quality

remains unresolved (Bell and Henschke 2005). Optimal YAN for fermentation ranges between 330 to 530 mg YAN/L depending on yeast strain and sugar content of the must (Jiranek *et al.* 1995a). However, it is generally agreed that adequate fermentation can still proceed at suboptimal rates as low as 150 mg YAN/L (Henschke and Jiranek 1993). Therefore, most treatments did not reach optimal level according to Henschke and Jiranek (1993); however, the majority were above 150 mg/L. In addition, all fermentations were supplemented with nutrient additions. These results do suggest that fermentation nutrition is lowered with extended ripening and increased nutrient supplements are needed to prevent arrested fermentation.

Keller *et al.* (1999) showed that increased nitrogen application in the vineyard resulted in increased growth and carried through to the subsequent fermentations resulting in reduced anthocyanins, total phenols, and increased malic acid and pH. The relationship between YAN and pH is presented in Figure 4.13 illustrating that as YAN increased, pH decreased ($p<0.001$). Furthermore, results from Keller *et al.* (1999) and *this* experiment suggest that vine and must nitrogen is correlated with wine color and acidity. Significant changes occurred in YAN due to crop load and extended ripening, although further research is recommended to better understand the interactions between crop load, ripening, must nitrogen and their effects on wine quality.

Table 4.20 : Correlation coefficient (r) and statistical significance level (p value) of the negative linear relationship between YAN and days after veraison. Significance level is denoted by *** and * for significance at $p<0.001$ and $p<0.05$, respectively.

Year	r	P value
2005	-0.56	***
2006	-0.40	*
2007	-0.62	***
mean	-0.53	***

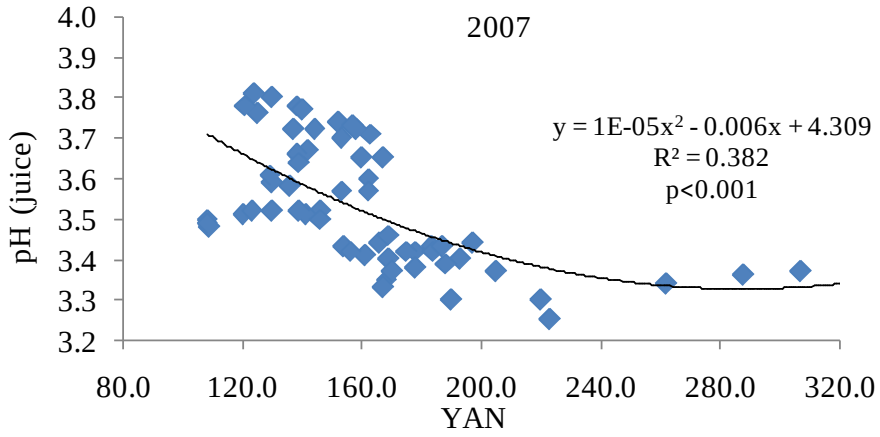


Figure 4.13: The relationship between yeast assimilable nitrogen (YAN) ppm and pH in juice (pH juice) in 2007. Data for all crop load and target °Brix treatments.

YAN differed considerably between seasons. The grand mean of all treatments was 140.3 ppm, 206.8 ppm and 165.3 ppm in 2005, 2006, and 2007, respectively. Once again, these differences highlight that season is influential on final fruit and wine composition.

4.4.3 Treatment Effects on Wine Composition

pH in treatment wines: The statistical analysis indicated that wine pH was significantly affected by both crop load and target °Brix at harvest: wines made from lower crop loads had a higher pH (wine). Titratable acidity of all treatments were adjusted to 7 g/L near the beginning of primary fermentation; therefore, significant differences between treatments and pH (wine) may indicate pH changes during fermentation. The trends of pH in *juice* and *wine* were similar only in 2005 and overall, the differences due to crop load in pH (wine) were greater than those for pH (juice). The difference between the highest and lowest pH (juice) for each crop load was 0.07, 0.06, and 0.03 in 2005, 2006, and 2007, respectively. The difference between pH (wine) was 0.22, 0.13, and 0.08 in 2005, 2006, and 2007, respectively. Most likely the fermentation process caused the greater changes in pH (wine). It is generally known that pH increases substantially during

primary fermentation (S. Peck, 2008, personal communication). Additionally, the acid adjustment in must after crushing could have attributed to the differences in pH wine.

The pH wine increased as °Brix at harvest increased and was significant in 2005 and 2006. This pattern was similar to pH juice, although the differences between highest and lowest pH juice were greater. For example, in 2005, 2006 and 2007, the greatest differences between pH juice due to ripening from 22.5 to 28.5 °Brix were 0.21, 0.36 and 0.39, respectively, and 0.08, 0.09 and 0.07 for pH wine. The dissimilar patterns in pH overall may be a factor of the interaction which occurred between crop load and °Brix. In 2005 and 2006, the lowest initial pH wine was in 20/22.5 and the highest was in UN/22.5, with the highest overall pH in 20/28.5. The interaction in 2007 followed the same pattern as 2005 and 2006 for initial pH wine, however the highest overall pH wine was in UN/28.5. Presumably, extended ripening in 2007 affected pH wine differently to the previous years. Certainly, both crop load and ripening had an impact on pH in resulting wines—however it is difficult to isolate the actual cause of these changes since pH is affected by many factors. Seasonal differences largely affected pH juice and pH wine in a similar pattern to other fruit composition results.

Titratable acidity in treatment wines: The trends in TA wine were dissimilar to those of TA juice for both factors—crop load and target °Brix at harvest. The TA juice was substantially lower than TA wine. Differences in TA *wine* were most likely associated with primary fermentation. Many changes occur as a result of primary fermentation (Boulton *et al.* 1996, Ribéreau-Gayon *et al.* 2006) and the must TA was adjusted to 7 g/L at the start of fermentation (as specified in the winemaking protocol). Presumably, TA juice is a more accurate indication of treatment effects on titratable acidity. Temperature determines the rate of respiration i.e the metabolism of tartaric and malic acid and is thereby greatly affected by temperature and climate.

Differences in leaf area can also restrict biosynthesis and respiration—and thus affect acidity in fruit.

Ethanol: Aside from water, ethanol (ethyl alcohol) is the most abundant compound in wine (Ribéreau-Gayon *et al.* 2006) making it an important part of wine composition. The effect of crop load on ethanol (ETOH) was significant in 2005 and 2007, but did not follow the same pattern each year. In 2005 the 20 cl had the greatest amount of ETOH and the UN had the lowest among all crop loads. The 2005 season had the slowest sugar ripening relative to the other seasons, most likely due to above average crop loads. Presumably, crop reduction to 20 cl enabled faster sugar ripening and consequently higher ETOH in the resulting wine. In 2007, the 40 and 60 cl had significantly greater amounts of ETOH relative to the 20 cl and UN. The 2007 season was characterized by faster than average sugar ripening and had lower crop load relative to the other years. In 2007, sugar (g)/berry had a large reduction in the 20 cl relative to the higher crop loads, indicating changes in actual sugar synthesis and accumulation. These changes in ETOH for the 2007 season may be more related to the means by which sugar increased in the berry. Undoubtedly, these data indicate a significant increase in ETOH as °Brix at harvest increased. Ethanol in wine is mainly produced by the alcoholic fermentation of sugar in must (Boulton *et al.* 1996, Ribéreau-Gayon *et al.* 2006) and substantiates why increased °Brix at harvest would increase ETOH in the resulting wine. Moreover, the ETOH results confirm the differences in °Brix at harvest among treatments in addition to the validity of the treatments. Ethanol content has been identified as a potential cause of stuck or sluggish fermentations (Casey and Ingledew 1985, Bisson and Butzke 2000) and is thereby problematic from a winemaking perspective. Additionally, wines sold with greater than 14.0 % alcohol require a higher excise tax in the United States than those below 14 %—and thus have greater financial ramifications.

Ethanol has solvent properties useful for dissolving phenols from pomace during fermentation (Ribéreau-Gayon *et al.* 2006); consequently the changes in ETOH due to increased °Brix at harvest may have affected the extraction of phenols during primary fermentation.

Although a significant treatment interaction occurred in all years, there was not a consistent pattern among years (Figure 4.13) except that all crop loads increased in ETOH as °Brix at harvest increased. In 2005, ETOH in the 20 cl increased considerably between the 25.5-28.5 °Brix targets relative to the other crop loads. In 2006, the UN increased most notably at the 27.0 and 28.5 °Brix targets compared with the other crop loads and in 2007 the 20 cl had a large decrease at the 27.0 and 28.5 °Brix targets. Seemingly, the interaction between crop load and °Brix at harvest was largely influenced by the individual season, with most differences occurring at the higher °Brix targets. The differences in ETOH may be more related to the 'rate of ripening' i.e. DAV to reach the target °Brix in each crop load than °Brix alone.

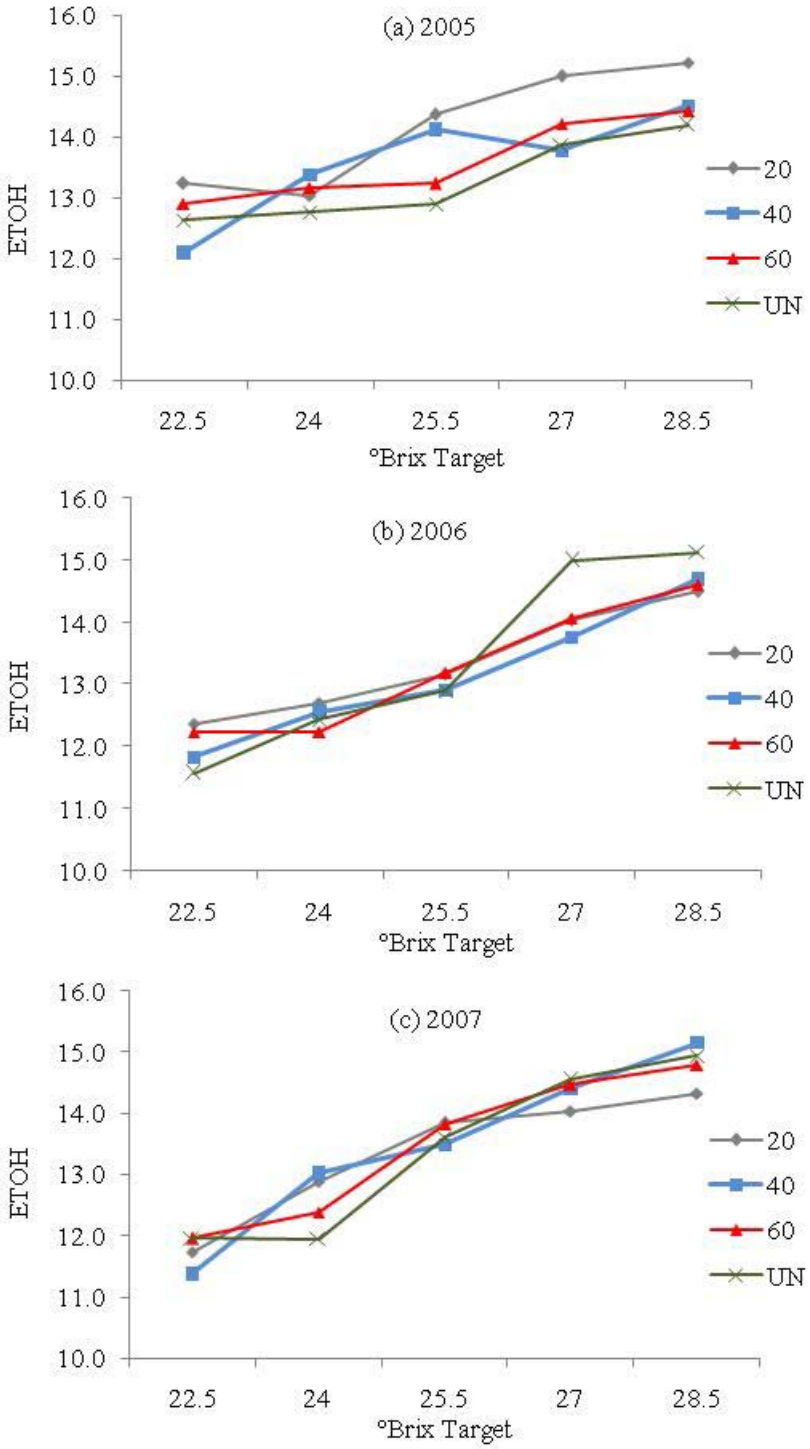


Figure 4.14: The interaction of crop load and target °Brix at harvest on ethanol (ETOH) in treatment wines for three seasons.

Methoxypyrazines: Isobutylmethoxypyrazine (IBMP) compounds are not bound to any other compounds and thus the IBMP concentration in wine is primarily dependent on the IBMP concentrations in the grape at harvest (Bell and Henschke 2005). This suggests that differences in IBMP between treatment wines were due to crop load and target °Brix rather than fermentation effects. Nitrogen application in the vineyard may have an indirect effect on IBMP concentrations in resulting wines due to changes in canopy microclimate (Bell and Henschke 2005). The leaf area index (LAI) data presented in Chapter 3 clearly demonstrates that LAI increased as crop load decreased. Allen (1993) reported that grape IBMP concentration increased as leaf layer number in the canopy increased, and consequently, IBMP was higher in less exposed fruit. In the current experiment, IBMP concentration decreased as °Brix at harvest increased for all crop loads although IBMP was consistently higher in the lower crop loads. In fact, the 20 cl and 40 cl had substantially greater IBMP concentration than the 60 cl and UN until the 28.5 °Brix target. Furthermore, the 20 cl consistently had the highest IBMP at all target °Brix. Chapman *et al.* (2004b) reported that methoxypyrazine concentrations, specifically 2-Methoxy-3-Isobutylpyrazine (MIBP), were negatively correlated with buds per vine; therefore lower yields had higher MIBP concentrations. The current results suggest that crop thinning indirectly reduced the degradation of methoxypyrazines, even with extended ripening to 28.5 °Brix. This was most likely a result of increased shading in the fruit zone for the higher crop loads. Excess shade would have interfered with the breakdown of methoxypyrazine compounds, which are light and temperature sensitive, and thus photo-degraded in ripening grapes (Hashizume and Samuta 1999). In addition, the higher crop loads required more DAV to reach each °Brix target thus allowing more time for methoxypyrazines to degrade, although the initial levels were still greatest in the lower crop loads. Ryona *et al.* (2008) concluded that light

exposure is critically important to IBMP accumulation and that initial IBMP may be a factor of both shading and stimulated vegetative growth.

Methoxypyrazines are nitrogen-containing compounds—therefore it is conceivable that increased vine nitrogen may increase the formation of methoxypyrazines in nitrogen responsive sites (Bell and Henschke 2005). IBMP is synthesized in the leaves, where it is mainly located, then transported from leaves to clusters between fruit set and 2-3 weeks before veraison (Roujou du Boubée *et al.* 2000, 2002). Although no differences were detected in vine nitrogen status based on the petiole and blade nutritional analysis data, YAN was highest in the lower crop loads signifying a change due to crop load. Treatment effects on YAN indicate differences in nitrogen within the berry which could have facilitated an increased formation of IBMP compounds in the lower crop loads. Currently, there are no published studies on nitrogen application in the vineyard or increased vine or must nitrogen and its subsequent effect on methoxypyrazines in fruit and wine composition. However, existing research does suggest that IBMP could have been affected directly, i.e. increased IBMP formation due to increased nitrogen in the vine, or indirectly due to an impact on canopy microclimate (Morrison and Noble 1990, Allen 1993, Chapman *et al.* 2004b, Wilkinson *et al.* 2006). Moreover, methoxypyrazines in grapes are directly correlated with methoxypyrazines in finished wines (Roujou de Boubée *et al.* 2002, Chapman *et al.* 2004b) The effects of methoxypyrazines on wine sensory are discussed in Chapter 5.

4.4.2 Treatment Effects on Wine Color and Phenolics

Total phenols: Wine phenolics are critically important to the quality of all wines (Peynaud 1996) and thus provide both a quantitative and qualitative measure of wine quality. Total

phenols at press and post malolactic fermentation followed similar trends with the exception of the 2006 crop load effect on total phenols pml for which there were no significant differences. In general, the lower crop loads had higher levels of total phenols relative to the UN (control). In particular, the 20 cl was greater than the UN at all measurements except for 2007 at press.

The measurement of total phenols is useful but limited in that it provides only a general measurement of all compounds present with aromatic rings, but no information about subclasses of phenolics (Harbertson and Spayd 2006). Perhaps total phenols were higher in the lower crop loads due to increased tannins and/or other non phenolic aromatic compounds i.e. alpha amino nitrogen. The clear trend in A(420 nm), A(520 nm) and color density indicated that phenolic compounds contributing to wine color density were highest for the higher crop loads and at the highest °Brix target. These findings support the idea that increased total phenols in the lower crop loads may have been due to increased tannins or other non phenolic compounds containing aromatic rings. Tannins were significantly higher in the lower crop loads. Additionally, astringency ratings on sensory analysis were higher in the lower crop loads and lower °Brix targets (discussed in Chapter 5). Tannins are widely known to be responsible for the astringent component of the mouthfeel of wine (Gawel 1998, Downey *et al.* 2003a). This may explain, in part, the higher level of total phenols in the lower crop loads.

Wines with total phenol values of less than 30 Absorbance (280 nm) are believed to have a low capacity for aging (Somers and Verette 1988). Therefore, wines produced from the 2006 season may have a low aging potential. These data emphasize that the season, in addition to cultural practices, still has a substantial impact of final wine quality.

Color density: Wine color intensity or density has traditionally been represented by the sum of absorbance at 420 nm and 520 nm (Zoecklein 1995). Color density was measured on relatively young wines (less than 6 months of aging); therefore, it is reasonable that the Absorbance (520 nm) was consistently higher than the Absorbance (420 nm). Polymeric pigments are generally regarded to absorb at A(520 nm) (Harbertson and Spayd 2006) and the absorption maxima of anthocyanins at wine pH are also at wavelength 520 nm (Somers 1971). In this experiment, both increased crop load and extended ripening positively affected wine color density. The higher crop loads had more optimal vine balance, as indicated by the growth data in Chapter 3. Undoubtedly, this contributed to the increased color in resulting wines. These results concur with Bravdo *et al.* (1985) who reported that wine color and crop load were negatively correlated and the most optimal vine balance, determined by Y/P ratio, was in unthinned vines.

Lower crop loads had less light in the fruiting zone, increased canopy growth, below optimum Y/P ratios and larger berries than the higher crop loads. Therefore, the influence of crop load on the light environment within the fruiting zone (PAR) most likely contributed to the differences in color density. This is further supported by a significant relationship ($p \leq 0.001$) between CD and LAI (Figure 4.15) in which color density decreased as LAI (harvest) increased—highlighting the influence of canopy microclimate on color development. Previous research has shown that overly shaded fruit negatively affects grape and wine color (Smart *et al.* 1985b, Price *et al.* 1995, Haselgrove *et al.* 2000, Ristic *et al.* 2007).

In contrast, wines made from grapes well exposed to sunlight had greater levels of phenolics and color (Mazza *et al.* 1999). However a high degree of berry temperature can also be detrimental to wine color and quality (Haselgrove *et al.* 2000, Tarara *et al.* 2008). The different crop loads resulted in yield compensation in berry weight, mean cluster weight and berries per cluster.

These changes may indicate different stress levels in the vine and therefore a stress response that may have affected grape phenolic composition and concentration.

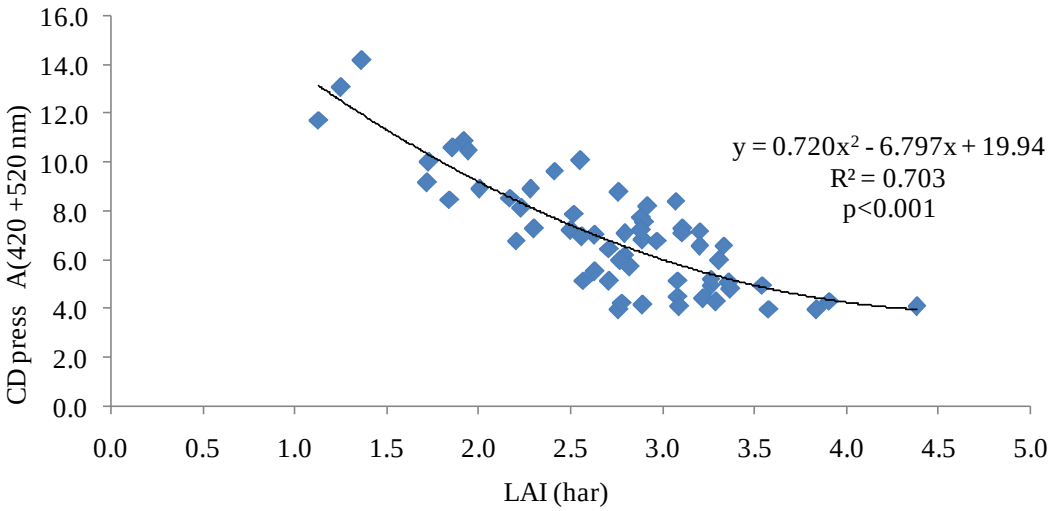


Figure 4.15: The relationship between leaf area index at harvest LAI (harvest) and color density at press (CD press) in 2007.

Extended ripening significantly increased Absorbance (420 nm), Absorbance (520 nm) and color density in wine at all stages. What remains unknown is whether extended ripening caused an actual increase in the synthesis of phenolic material attributing to wine color, increased extractability, or both. In all likelihood, extractability increased as °Brix at harvest increased due to a greater amount of ethanol produced during fermentation. Ethanol is known to increase extractability of pomace during fermentation due to its solvent properties (Ribéreau–Gayon *et al.* 2006) but has also been demonstrated to decrease 420 nm and 520 nm (Somers and Evans 1979). Canals *et al.* (2005) demonstrated that the extraction of anthocyanins from skins and proanthocyanidins in skins and seeds increased with ripening.

Results from color density data indicated an interaction between crop load and target °Brix at harvest. Due to the steady increase in CD as °Brix at harvest increased, it is conceivable that

extended ripening increased the extractability of phenolic material from berry skins. The berry shrivel that occurred with extended ripening may have aided in skin extractability due to loosening and increased break down within the skins cell wall.

In addition to °Brix at harvest, increased crop load certainly improved CD relative to the lower crop loads. This was most likely an indirect affect of crop load. Zoecklein *et al.* (1995) reported that the rate of color change in wine is affected by both phenolic concentration and composition.

There was a significant interaction between crop load and target °Brix at harvest on Absorbance (420 nm), Absorbance (520 nm) and color density at both wine stages and in all years. The main interaction between crop load and target °Brix was that CD press increased as crop load and target °Brix increased. In general, higher crop loads had a higher initial CD (both at press and pml) and remained higher overall as CD increased with extended ripening.

In both 2005 and 2007, the UN had the highest initial CD measurement at the 22.5 °Brix target, contrasting with the 20 cl and 40 cl which had the lowest CD (Figure 4.15 a, c). Additionally, Figure 4.15 highlights the overall increase in CD press as the target °Brix increased. Moreover, the 20 cl had the lowest CD at press and pml relative to the other crop loads even with extended ripening to the 28.5 °Brix target. Furthermore, the lower crop loads did not attain CD levels equivalent to that of the initial CD in the 60 cl and UN until near 25.5-27.0 °Brix. Table 4.21 lists the equations and coefficient of determination for the regression analysis in Figure 4.16.

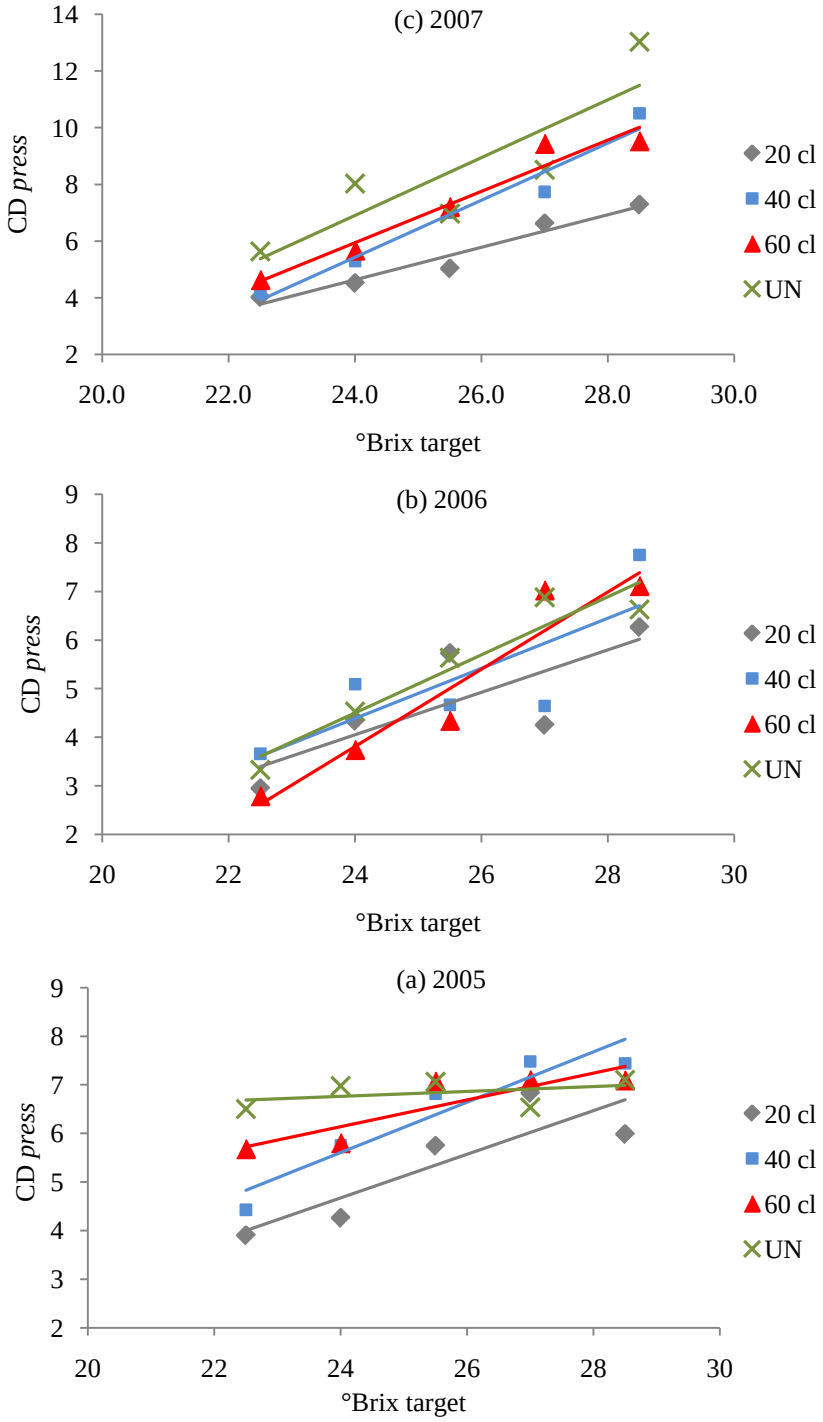


Figure 4.16: Linear regression model and interactions of color density at press (CD press) in wine made from crop load and extended ripening treatments in each year.

Table 4.21. Equations and coefficient of determination for parallel regression model of color density accumulation in 2005, 2006, and 2007.

	Color Density Accumulation								
	2005			2006			2007		
Treatment	**constant**	**slope**	**R^2**	**constant**	**slope**	**R^2**	**constant**	**slope**	**R^2**
20 cl	-6.11	0.45	0.75	-6.43	0.44	0.62	-9.18	0.58	0.95
40 cl	-6.81	0.52	0.89	-7.99	0.52	0.63	-18.8	1.10	0.96
60 cl	-0.48	0.28	0.78	-15.27	0.80	0.92	-15.8	0.90	0.95
UN	5.55	0.05	0.17	-9.82	0.60	0.91	-17.54	1.00	0.75

Undoubtedly, crop load and target °Brix affected CD; however, the overall magnitude of CD was largely affected by the individual season (Figure 4.16). The 2007 season had the highest CD relative to 2005 and 2006, and in contrast 2006 generally had the lowest. In summary, extended ripening, appropriate crop load and optimal vine balance improved wine color; although, seasonal characteristics remained largely influential on wine color and composition.

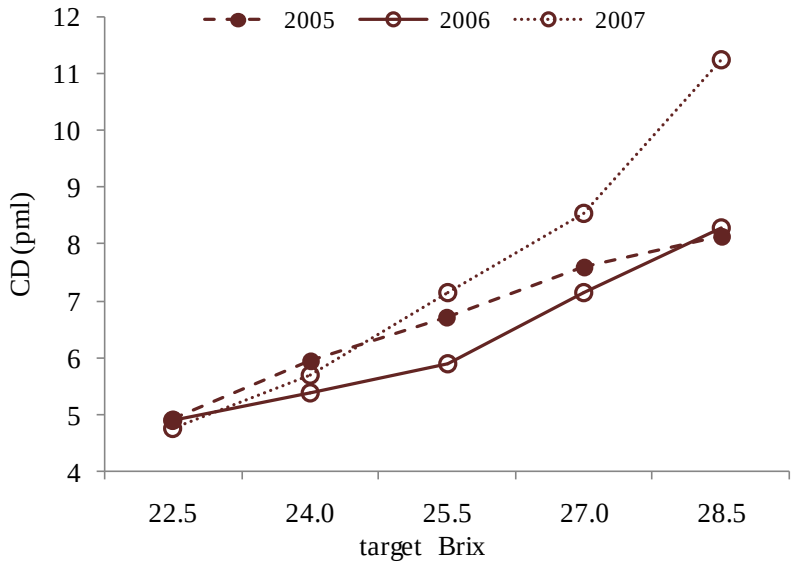


Figure 4.17: Seasonal affects on color density (CD) post malolactic fermentation (pml) for 2005, 2006, and 2007 relative to target °Brix at harvest.

***Hue*:** The tone or hue of wine is calculated as the ratio of Absorbance at 420 nm and 520 nm (Sudraud 1958, Glories 1984) and is used to monitor changes in wine as the ageing process causes wine to shift from red to brick red. The hue of color has been designed as its dominant wavelengths visible to humans e.g. red, yellow, green or blue. For red wine color analysis, the hue is specifically defined as the ratio of absorbance at the wavelengths of 420 nm and 520 nm i.e. A(420nm)/A(520nm) (Zoecklein *et al.* 1995). Hue at press and Hue pml decreased as crop load increased suggesting a change in either the Absorbance (420 nm) or Absorbance (520 nm) which affected the ratio Absorbance (420nm)/Absorbance (520 nm). As wine matures there is a shift in absorption maxima to between 400-500 nm. Hence, wine hue naturally changes as wine ages, but in young wines a lower hue is more desirable—signifying an absorption maxima near A(520 nm) (Zoecklein *et al.* 1995), a higher amount of blue pigment and better aging potential (J. Meier, 2008 personal communication). Although color density increased as crop load and target °Brix at harvest increased, the lowest or 'best' hue did not always follow this pattern. The lowest crop load (20 cl) consistently had the least desirable (highest) hue. Hue based solely on target °Brix was more ambiguous. The lowest hue shifted among the °Brix targets and there were less significant differences among means due to °Brix than crop load. Hue is greatly influenced by wine pH and may explain in part, the dissimilar patterns between hue and color density. This highlights that hue is a useful tool but should be considered a secondary quality parameter and used in conjunction with other phenolic measurements such as color density and total phenols.

Total anthocyanins: In an effort to conserve costs, treatment wines analyzed by Enologix® Laboratory were a blend of each treatment's field/fermentation replication. Hence, an interaction could not be measured statistically on this data set. The effect of target °Brix at harvest on total

anthocyanins followed trends similar to color density in all years and further supports that extended ripening improved wine color. Light is known to be an important factor in the regulation of anthocyanin synthesis (Pirie and Mullins 1980, Smart *et al.* 1985b, Mullins 1992, Spayd *et al.* 2002) which may explain the higher anthocyanin content in the higher crop loads which received more light in the fruiting zone.

Additionally, temperature influences anthocyanin synthesis. Studies have indicated that anthocyanin synthesis is inhibited and/or degraded when excessively high berry temperature occurs e.g. greater than 30-35 °C (Buttrose 1971, Spayd *et al.* 2002); although the specific critical temperature affecting anthocyanins may change with variety. In this experiment the UN had the greatest PAR in the fruiting zone, and subsequently the highest anthocyanins and color density. These results suggest that, although light increased in the UN fruiting zone relative to the other crop loads, it did not detrimentally affect berry temperature to the point of anthocyanin degradation. Moreover, this signifies adequate vine balance and a good canopy microclimate in the higher crop loads.

Tannins: Tannins in treatment wines increased as crop load decreased. Tannin accumulation is most active between flowering and fruit set and is complete in berry skins at veraison and in seeds 1-2 weeks post-veraison (Robinson 2006). Crop was adjusted post fruit set and prior to veraison, and perhaps affected the initial tannin accumulation within each crop load. Tannins were shown to increase with increased shade in the fruiting zone (Ristic *et al.* 2007). Therefore, it is likely that tannins increased as an indirect effect of crop reduction, and increased shoot growth and shading in the fruiting zone. The tannin maturation phase occurs from veraison to harvest, resulting in a significant decrease in extractable tannins (Robinson 2006) due to tannins binding to the fruit tissues (Kennedy *et al.* 2000, Downey *et al.* 2003a). Although there were no

significant differences found in tannins due to target °Brix at harvest, tannins were only measured in wine and at one stage. A more extensive investigation on seed, skins and wine tannins throughout ripening may have revealed greater differences and warrants further research. In addition, there were large seasonal affects on tannins even though extended ripening had no significant effect. These results suggest that tannin levels within the berry are established early in development, and their magnitude is greatly affected by seasonal characteristics. However, the 'maturity' of tannins is unknown based on this analysis. Fournand *et al.* (2006) reported that with increased berry ripeness, a higher amount of cell wall oligo-and polysaccharides are extracted into wine and can interfere with tannin perception. Harbertson *et al.* (2002) reported that the number of seeds/berry was the major factor in tannin differences between three varieties: Cabernet Sauvignon, Syrah and Pinot Noir. Therefore, it is conceivable that larger berries with more and/or larger seeds could have affected tannin in treatment wines—however seed number or size/berry was not measured. Moreover, the relationship between tannins and ripening merits continued research.

4.5 Conclusions

a) Crop load reduction increased the rate of sugar ripening, although the overall rate of °Brix accumulation and °Brix maximum was largely influenced by the individual season. All crop loads achieved maturity greater than 26 °Brix, which suggests that vine photosynthetic capacity was sufficient to ripen fruit irrespective of large differences in crop load.

b) In 2007, sugar (g)/berry decreased after a certain point in ripening even though °Brix sustained an increasing trend. These results suggest that, in some cases increased °Brix is more

related to a concentration effect within the berry due to dehydration, than new sugar synthesized and transported from leaves to berries.

c) Higher crop loads in general had higher titratable acidity and lower pH. All crop loads decreased in titratable acidity and increased in pH as °Brix at harvest and/or days after veraison increased. Wine pH was consistently higher than juice pH.

d) Yeast assimilable nitrogen became significantly higher as crop load decreased. Additionally, extended ripening caused YAN to decrease in all crop loads.

e) Isobutyl methoxypyrazine concentration was highest in the lowest crop load: initial concentrations of IBMP were inversely related to crop load, i.e. as crop load decreased IBMP increased. IBMP decreased as °Brix at harvest and DAV increased. These results, largely supported by previous research, suggest that crop reduction increased shading in the fruiting zone, consequently increased accumulation and the initial concentration of IBMP in berries, and hindered the breakdown of IBMP during ripening.

f) As target °Brix at harvest increased so did ethanol in subsequent wines.

g) Crop load and extended ripening significantly affected wine color and phenolics. The A(420 nm), A(520 nm), color density and anthocyanins increased as both crop load and °Brix at harvest increased. Total phenols increased as °Brix at harvest and DAV increased. Total phenols were generally lowest in the UN and highest in the 20 cl and corresponded with trends in tannins.

h) Vines with the most optimal vine balance, as determined by Y/P ratio, had the highest color density in subsequent wines.

i) Crop load, extended ripening and vine balance significantly affected wine color; however, phenolics and final fruit and wine composition were also influenced by the individual season.

Chapter 5: EFFECTS OF CROP LOAD AND EXTENDED RIPENING ON WINE SENSORY ANALYSIS

5.1 Introduction and Experimental Aims

Following vineyard experiments through to sensory analysis of wines is necessary if the ultimate goal is to influence wine sensory attributes through vineyard management (Chapman *et al.* 2004, Chapman *et al.* 2005, Heymann 2006). Most previous experiments on vineyard practices have concluded at juice analysis of only °Brix, pH and TA, rather than carrying through to wine sensory. This is limiting because very few analyses can accurately predict wine sensory properties. Additionally, wine quality on its own is subjective if sensory panelists are not trained; hence wine quality ratings can be objective with the use of a trained panel. Descriptive analysis is described to transcend quality and difference measurements by determining sensory attributes that differ among wines without reliance on preferences of the judges (Lawless and Heymann 1998). Therefore, the quantitative measurement of wine sensory properties and separately, consumer liking would greatly increase the understanding of vineyard effects on subsequent wine quality.

A general definition of quality from the International Standards Organisation (Standards Australia 2001) states that quality is "the degree to which a set of inherent characteristics fulfill requirements". For wine quality, inherent characteristics may include sensory properties such as appearance, smell and flavor; in addition to non sensory attributes such as price, brand reputation and familiarity (Lattey *et al.* 2007).

Crop load has an integral effect on ripening and resultant wine quality, however most studies on crop load have not included wine sensory analysis. Furthermore, few studies exist on the effects of extended ripening on wine sensory and therefore warrant further investigation.

The aims of this study were to:

1. Study the effects of crop load and extended ripening treatments within the vineyard on resultant wine quality as determined by an expert panel and relative to potential price point for an established commercial brand.

2. Identify and quantitatively rate the attributes which describe wines made from the crop load and extended ripening treatments through descriptive analysis testing and scaled attribute ratings.

3. Investigate the effects of crop load and extended ripening vineyard treatments on consumer preference in consequent wines.

5.2 Materials and Methods

5.2.1 Difference Testing

The Triangle test was conducted for the 2005 and 2006 vintages to determine whether any significant differences occurred in wines due to the vineyard field replications, fermentation replications, and/or barreling of wines. Procedures from Lawless and Heymann (1998) were followed. The field replications were tested against each other within a given treatment e.g. R1 vs R2, R2 vs R3 and R3 vs R1 for each of the 20 total treatments and for the corresponding vintage. Each session included a total of six panelists whom tasted four flights each. Panelists were wine industry professionals and were consistent within a given session, but not among all sessions. There was one session per day for 20 days (four business weeks) in which one treatment (including the three replications) were tested. Wines were presented in randomized order for the six panelists. Panelists had a forced break of one minute between directional tastings. Wines were poured to 44.4 mL per glass. Panelists were told that two wines in each

flight were the same; identify the wine which was different. Wines were not color masked. A sample tasting set up is presented in Appendix 12. Panelists were also asked to identify whether the difference was due to sulfides, volatile acidity (VA) or other, to deduce whether differences found were a mistake in cellar maintenance practices or were truly reflective of the field replications.

The 2007 vintage had less wine per fermentation due to uncontrollable yield differences; therefore, it was necessary to blend the barrel replications after malolactic fermentation (i.e first racking) in order to keep the barrels full. Due to harvest time logistics, the triangle testing was not possible. However, a formal difference screening was conducted by the Associate Enologist and Viticulturist prior to any blending of treatment lots. Both the Associate Enologist and Viticulturist had a screening evaluation sheet which was filled out individually, then compared. The evaluation had a 1-5 rating system for the categories of color, aroma, and flavor profile with 1 being no difference and 5 being extremely different (Appendix 13). Any scores of '3' or greater dictated that the particular barrel would be rejected out of the treatment blend.

5.3 Expert Panel

Treatment wines were scored by the expert panelists in January of each year following the previous vintage. The panelists consisted of J. Lohr Vineyards and Wines management who annually score and categorize commercial wines to determine their market placement including: price point and brand within the J. Lohr portfolio. There were on average 12 expert panelists who included: Vice President (VP) of Winemaking, Red Winemaker, Enologist, Associate Enologist, VP Sales and Marketing, President/Proprietor, Vineyard Managers (2), Viticulturists

(3), and other associated upper management (2). The expert panelists had on average 15 years of wine industry experience and between 8-20 years scoring wines specific to J. Lohr brands.

Treatment wines were taken directly from barrels two days prior to the sensory testing. Wine prepared for bottling followed the standard practice of J. Lohr Winery wherein, they were adjusted to 25 ppm of Sulfur Dioxide ($SO_{2)}$ and 1 ppm of Copper Sulfate ($CuSO_4$) as a standard practice to suppress any sulfides—sometimes present in young wines. Wines were scored based on the grading system presented in Table 5.1. Notably, wine quality and expert score are inversely related; therefore, a lower score indicates higher wine quality. Panelists had a minimum of 3 previous years experience with the scoring system and therefore were familiar with the scoring system. Treatment wines were presented blind, in four flights of five wines, and in randomized order for each panelist. All wines were evenly poured to 1.5 ounces per glass. Panelists were instructed to taste wines at a comfortable pace, break when needed, and score wines based on the letter grade (e.g. A-1) which was later converted to the equivalent number score. In addition to quality score, in 2006 and 2007 panelists were asked to rate these three attributes: vegetative intensity, body and flavor intensity on the 1-9 category (box) scale. These attributes were selected given their role in largely distinguishing wines from the 2005 vintage and are known to be pivotal in the quality perception of Cabernet Sauvignon. Additionally, these attribute ratings were used to investigate other trends and reasoning for the expert scores and to connect trends among the other sensory tests.

Table 5.1: Commercial wine scoring system used for expert panel to grade treatment wines into market value price point. Note: Wine quality and price point increase as score decreases.

Letter Grade		Score Value	Price Point
A	1	**1**	>$50.00 USD
	2	**2**	
	3	**3**	
B	1	**4**	$50.00 - 30.00 USD
	2	**5**	
	3	**6**	
C	1	**7**	$20.00-16.00 USD
	2	**8**	
	3	**9**	
D	1	**10**	$10.00-8.00 USD
	2	**11**	
	3	**12**	
E	1	**13**	$5.00-3.00 USD (no J.Lohr program)
	2	**14**	
	3	**15**	

5.3.1 Descriptive Analysis

The descriptive analysis testing was conducted by Vinquiry laboratory in July of each year following the previous year's vintage. As explained in section 5.2.1, difference testing was performed at J. Lohr Winery to determine if there were significant differences among the three field/fermentation replications within each treatment. Prior to descriptive analysis, wine treatments and associated replications that exhibited significant differences were excluded from the analysis. In most cases, the difference testing confirmed that a composite sample of all three replications was suitable for descriptive analysis. In addition, the composite sample eliminated subtle variability and was cost sensitive.

The treatment wines were blended from three replications to one lot in June of each year at J. Lohr Winery. A stainless steel hopper was used to contain and blend each treatment lot. Wine

was then bottled (750mL) and finished with synthetic corks. Bottled wines were shipped overnight approximately 2 weeks later via Federal Express to Vinquiry Laboratories, St. Helena California.

One bottle of each treatment wine was used for panel training and selection of the descriptive terms (language development). The other three bottles were used for the formal sessions.

Sensory panel: Five panelists participated in the study. One judge had experience as a professional winemaker and all five had many years of experience tasting wines professionally and with descriptive analysis.

Descriptive terms and standards: Descriptive analysis training was conducted in one, two hour session. The five judges were presented with the twenty treatment wines and asked to generate descriptors for the appearance, aroma, flavor and after-flavor of the wines. Panelists tasted the wines and rinsed with distilled water between tastings. Panelists were given the following instructions for tasting during the language development session.

1. Evaluate the 20 wines in front of you using the "Descriptive Analysis Worksheet."
2. Use the "Word List for Red Wines," if needed.
3. For "Appearance," refer to the 2 hand-outs.
4. Smell all 20 wines first and make comments. Take short breaks or smell the glass of water if you experience adaptation.
5. Return to the first sample and begin tasting. Please expectorate all wines.
6. Try to wait 90 seconds between wines. Rinse your mouth with water and eat crackers, if needed.
7. After you have completed the profiling, please arrange the 20 wines into groups based on their degree of similarity. You may create any number of groups and use any criteria to sort the wines. Please write down the groups with the wine code numbers below.

After evaluating all wines, each panel member was individually asked to state the descriptors/attributes which described the wines, and were concurrently written on a board.

Judges discussed and decided which descriptors best identified key attributes and differences among the wines. Descriptors chosen included visual, aroma and flavor attributes. Visual reference standards were used for the attributes color depth and red color intensity. The judges had been previously trained using aroma reference standards for DA testing.

The original descriptors in 2005 were: *red color intensity*, *color depth, sulfides*, *acidity*, *berry aroma, jam/dried fruit aroma, herbaceous aroma*, *vegetative aroma*, *woody aroma, acidity*, *berry flavor, jam/dried fruit flavor*, *vegetative flavor, astringency, mouthfeel/body, ethanol burn* and *duration of flavors*. Descriptors which were deemed confusing or not prominent in subsequent years were dropped accordingly in 2006 and 2007. The descriptor 'red color intensity' was used to describe wines which had a stronger red-orange (i.e. more brick or garnet color) rather than a purple-pink red. The latter is considered more reflective of higher wine quality, and therefore a higher red color intensity score suggests lower wine quality. The color depth rating increased as wine color became darker and deeper. Increases in color depth were described as related to increased wine quality. During the training period, panelists reached a consensus on the set of visual reference standards. Wine 20/24.0º was the reference standard for "brownish-red", 60/28.5º for "ruby-red", UN/25.5º for "blueish-red", 60/22.5º for "low depth of color" and 20/28.5º for "high depth of color". Reference standards used for training are listed in Appendix 17. Panelists transitioned to formal evaluation after the training period and when all panelists mutually agreed on the chosen descriptors.

Experimental design and tasting procedure: A semi-randomized complete block design was used for the formal sessions. Because there were color differences among the wines, a subset of ten of the twenty wines was evaluated at each session. Six formal sessions were held on different days during a three week period to evaluate the 20 wines in triplicate. Wines 20/22.5,

20/25.5, 20/28.5, 40/24.0, 40/27.0, 60/22.5, 60/25.5, 60/28.5, UN/24.0 and UN/27.0 were evaluated together in each of the three sessions. The remaining ten wines were evaluated together in the other three sessions.

Each judge rated the intensity of the attributes using unstructured line scales anchored at the ends with terms “blueish” and “brownish”, “light” and “dark”, “low” and “high”, “thinner” and “thicker” or “short” and “long”. All wines were served in clear, tulip-shaped wineglasses of 220 mL capacity, and coded with 3-digit random number codes. A 60 mL sample was poured into each glass and then covered with a 5.7 mm diameter plastic Petri dish cover at least 15 minutes prior to evaluation. The tests were conducted in a sensory room illuminated with fluorescent lighting. All wines were served between 16°-22° C on tables with white surfaces. Panelists were separated from each other by partitions. Sessions were held in the morning. Wines were served in two flights of five wines. Each flight was placed on a tray and delivered to the tasting booth. Appearance reference standards were present in each tasting booth. Judges evaluated one wine at a time for appearance, aroma and flavor—beginning with the first wine in the flight. Judges were not allowed to return to previous wine samples. Judges took a 10-minute break after the first flight and then proceeded to evaluate the second flight. Water and bland crackers were provided as a palate cleanser. Panelists expectorated the wines and rinsed with distilled water between evaluations.

Data analysis: Data entry and statistical analysis were conducted using FIZZ sensory software. Analysis of variance (AOV) was run on each attribute rated by the judges. Means of the 20 wine attributes were additionally analyzed by principal component analysis (PCA).

5.3.2 Scaled Attribute Rating

A scaled attribute rating was conducted on the 2005 vintage at the University of Adelaide sensory lab. Four attributes were selected and rated on the 9 point category scale. The attributes were: *fruit intensity, vegetative, body, and quality*. There was one session per week for three weeks and included twenty panelists per session. The panelists were given training prior to the first session to establish common descriptors for each of the attributes. The training presented panelists with wine samples that had intensified levels of vegetative or fruit characteristics, and three quality levels of purchased wine. The intensified vegetative characteristic was made using 50 mL of a base wine (Yalumba cask wine, Cabernet Sauvignon), and adding 20 grams of capsicum. The intensified fruit characteristics were made by adding 10 mL of Ribena syrup to 50 mL of wine. The three quality tiers were distinguished by price and style and included Charles Melton Cabernet Sauvignon (high; retails for ≈ 39.00 Australian Dollar AUD), Peter Lehman Cabernet Sauvignon (medium; retails at ≈18.00 AUD), and Yalumba Cask wine (low; retails ≤ 12.99 AUD). The body descriptor was described to panelists as oil, milk and water, corresponding to high, medium and low body, respectively.

5.3.3 Consumer Preference

A likability rating was conducted to capture consumer preference on the 2006 vintage. The testing was conducted at the University of Adelaide sensory laboratory. Three sessions were held throughout a three week period with 20-24 panelists per session—number of panelists was limited by wine quantity and logistics. Panelists were asked to rate each of the twenty treatment wines on the 9 point hedonic scale based on likability. The wines were labeled with three digit codes and were presented to each panelist in a different tasting order. Testing methods followed

Lawless and Heymann (1998, Chapter 13 p 450-456.) Additionally, a demographic survey was given to each panelist to address and connect preference trends.

5.3.4 Statistical Analysis

Analysis of variance on data was analyzed using GenStat® 10th edition and FIZZ sensory software for the descriptive analysis. Mean separation was done using LSD and Duncan's multiple range test for means at $p<0.05$.

5.4 Results

5.4.1 Difference Testing

Triangle test: Results from the triangle test (Table 5.2) are the summed number of correct judgments out of a total of 24 possible. These data indicate that differences between replications were more prominent at the lower °Brix levels relative to the higher °Brix (i.e. 27.0 and 28.5). At the 22.5 °Brix target the majority of triangle tests correctly identified significant differences at $p<0.05$ or greater in 2005 and 2006. However, overall there were more significant differences detected in the 2005 wines as compared with 2006.

Table 5.2: Triangle test results of crop load and °Brix at harvest vineyard treatments and subsequent wine replications for two years. Data presented is the number of correct judgements out of a possible 24. Trials which have 15 or more correct judgements are significant at p<0.001 (***) and indicate that a significant difference was detected.

Comparison	2005	2006	2005	2006	2005	2006	2005	2006
	20/22.5		**40/22.5**		**60/22.5**		**UN/22.5**	
R1 vs R2	13	20*	22***	23***	13	17***	10	17***
R2 vs R3	20***	15***	23***	17***	13	9	23***	7
R3 vs R1	24***	9	13	17***	9	18***	16***	11
	20/24.0		**40/24.0**		**60/24.0**		**UN/24.0**	
R1 vs R2	13	14	21***	17***	12	5	10	14
R2 vs R3	12	18***	11	12	7	6	19***	10
R3 vs R1	9	13	11	16***	10	9	16***	6
	20/25.5		**40/25.5**		**60/25.5**		**UN/25.5**	
R1 vs R2	8	14	16***	9	22***	8	16***	8
R2 vs R3	8	11	10	5	9	12	13	7
R3 vs R1	13	11	11	8	17***	12	10	7
	20/27.0		**40/27.0**		**60/27.0**		**UN/27.0**	
R1 vs R2	9	10	9	11	14	7	11	7
R2 vs R3	9	11	12	12	7	13	10	11
R3 vs R1	12	8	9	11	19***	7	12	8
	20/28.5		**40/28.5**		**60/28.5**		**UN/28.5**	
R1 vs R2	15***	8	10	8	9	8	13	13
R2 vs R3	5	14	9	14	14	10	19***	13
R3 vs R1	13	12	10	11	11	11	11	8

5.4.2 Expert Panel

Expert quality score: Treatment wines were scored by the expert panel in all years (Table 5.3). Crop load significantly affected wine score in 2006 and 2007. On average, wine score decreased as crop load increased, signifying an improvement in wine quality as crop load became greater. The UN had the lowest (best) average score among all years (8.9), and the 20 cl had the highest (worst) average score (9.4).

Degrees Brix at harvest significantly affected wine score in all years and demonstrated a clear and consistent trend ($p \leq 0.001$). Wine score decreased as °Brix at harvest increased in all years. There were no significant differences between the 27.0 and 28.5 °Brix targets in any years. Although the 22.5, 24.0 and 25.5 °Brix targets were not different from each other in 2005, they were each significantly different from each other in 2006 and 2007. A significant interaction occurred each year.

Table 5.3: Effect of crop load and target °Brix at harvest on wine score by an expert panel for three growing seasons. Wine score is based on a 1-15 scale with 1 being of highest quality and 15 of lowest quality.

	Wine score		
Treatment	**2005**	**2006**	**2007**
Crop load			
20	9.6	9.5 bc	9.0 b
40	9.2	9.7 c	8.5 a
60	9.3	9.2 ab	8.9 b
UN	9.1	9.0 a	8.7 ab
	ns	**	*
Brix			
22.5	10.1 b	11.0 d	10.3 d
24.0	9.7 b	9.9 c	9.4 c
25.5	9.7 b	9.3 b	8.5 b
27.0	8.4 a	8.5 a	7.7 a
28.5	8.6 a	8.1 a	7.8 a
	***	***	***

Expert attribute ratings: The attributes vegetative, body, flavor intensity and quality, were rated by the expert panel on treatment wines in 2006 and 2007 (Table 5.4). Crop load significantly affected the vegetative attribute which was rated highest in the 20 cl in both 2006 and 2007. Generally, the vegetative attribute rating decreased as crop load increased although in 2007 the UN was statistically similar in mean separation by LSD to the 20, 40 and 60 cl. The vegetative

rating significantly decreased as °Brix at harvest increased. The lowest ratings were consistently at the 27.0 and/or 28.5 °Brix targets. An interaction ($p<0.05$) occurred in 2006 only.

The wine attribute 'body' was significantly affected by crop load in 2007 and more notably by °Brix at harvest in both 2006 and 2007 ($p<0.001$). The highest rating of the body attribute due to crop load was in the 40 cl; this was also statistically similar to the UN. Although no significant differences were found due to crop load in 2006, it should be noted that the highest rating was in the UN. The body rating increased significantly as °Brix at harvest increased and there was a significant interaction each year.

Flavor intensity ratings due to crop load were only significant in 2006 in which the UN had the highest rating for flavor intensity relative to all other crop loads. Degrees Brix at harvest had significant effects on flavor intensity ratings ($p<0.001$). The flavor intensity rating significantly increased as each target °Brix increased with the exception of 2007 when the 27.0 and 28.5 °Brix were not statistically different from each other. A significant interaction occurred in each year.

In 2006, the expert panel rated each wine on the attribute 'quality' in addition to the quality score based on the commercial grading system. There were significant differences due to crop load, °Brix at harvest and an interaction. The highest rating for the attribute quality was in the UN which was rated significantly higher than the 20 and 40 cl. The quality rating became significantly higher as °Brix at harvest increased. The interaction showed that the UN started with the highest quality rating at the 22.5 °Brix target and remained considerably higher than the 20 and 40 cl until the final harvest at 28.5 °Brix. The highest ratings for the quality attribute were in treatments 20/28.5 and UN/28.5.

Table 5.4: Effect of crop load and target °Brix at harvest on the wine attributes *vegetative* , *body* , *flavor intensity* and *quality*, rated by an expert panel using the 1-9 category scale for two growing seasons. A rating of 1 reflects that the presence of the attribute or quality was extremely low and a rating of 9 reflects that the attribute or quality presence was extremely high.

	Vegetative		Body		Flavor intensity		Quality
Treatment	**2006**	**2007**	**2006**	**2007**	**2006**	**2007**	**2006**
Crop load							
20	4.2 d	5.0 b	4.6	4.8 a	4.5 a	5.2	4.4 a
40	3.4 c	4.2 a	4.2	5.3 b	4.3 a	5.6	4.3 a
60	3.0 a	4.3 a	4.5	4.5 a	4.7 b	5.0	4.6 ab
UN	3.0 a	4.5 ab	4.7	4.9 ab	5.0 c	5.2	4.9 b
	***	*	ns	*	**	ns	**
Brix							
22.5	3.8 c	5.7 c	3.3 a	3.3 a	3.3 a	3.7 a	3.3 a
24.0	4.2 c	5.1 c	3.9 b	4.1 b	4.0 b	4.3 b	4.0 b
25.5	3.6 b	4.4 b	4.4 c	5.1 c	4.6 c	5.6 c	4.5 c
27.0	2.7 a	4.1 b	5.1 d	6.0 d	5.3 d	6.4 d	5.2 d
28.5	2.7 a	3.4 a	5.7 e	5.8 d	5.9 e	6.1 d	5.7 e
	***	***	***	***	***	***	***
Interaction	*	ns	*	***	**	***	**

Means with columns separated by different letters differ significantly at $p<0.05$ *, **, ***, ns, indicate significance at $p<0.05$, 0.01, 0.001, or not significant respectively.

The linear regressions of attributes and wine scores from the expert panel in 2006 and 2007 treatment wines were significant each year and are presented in Figure 5.1 (a-f). A significant relationship occurred between the vegetative ratings and wine score ($p<0.01$, 0.001) in that wine score increased (quality decreased) as the vegetative rating increased (Figures 5.1 a, d). In contrast, the relationship between body and wine score (Figure 5.1 b, e), and flavor intensity and wine score (Figure 5.1 c, f), illustrated that wine score decreased (quality increased) as the body and flavor intensity attribute increased ($p<0.001$). Furthermore, these relationships provide insight to the quality perceptions of the expert panel showing that for Cabernet Sauvignon, higher quality wines are associated with low vegetative attributes, high flavor intensity and high

body. Moreover the expert panel was trained to score wines based on quality and potential price points of established commercial brands.

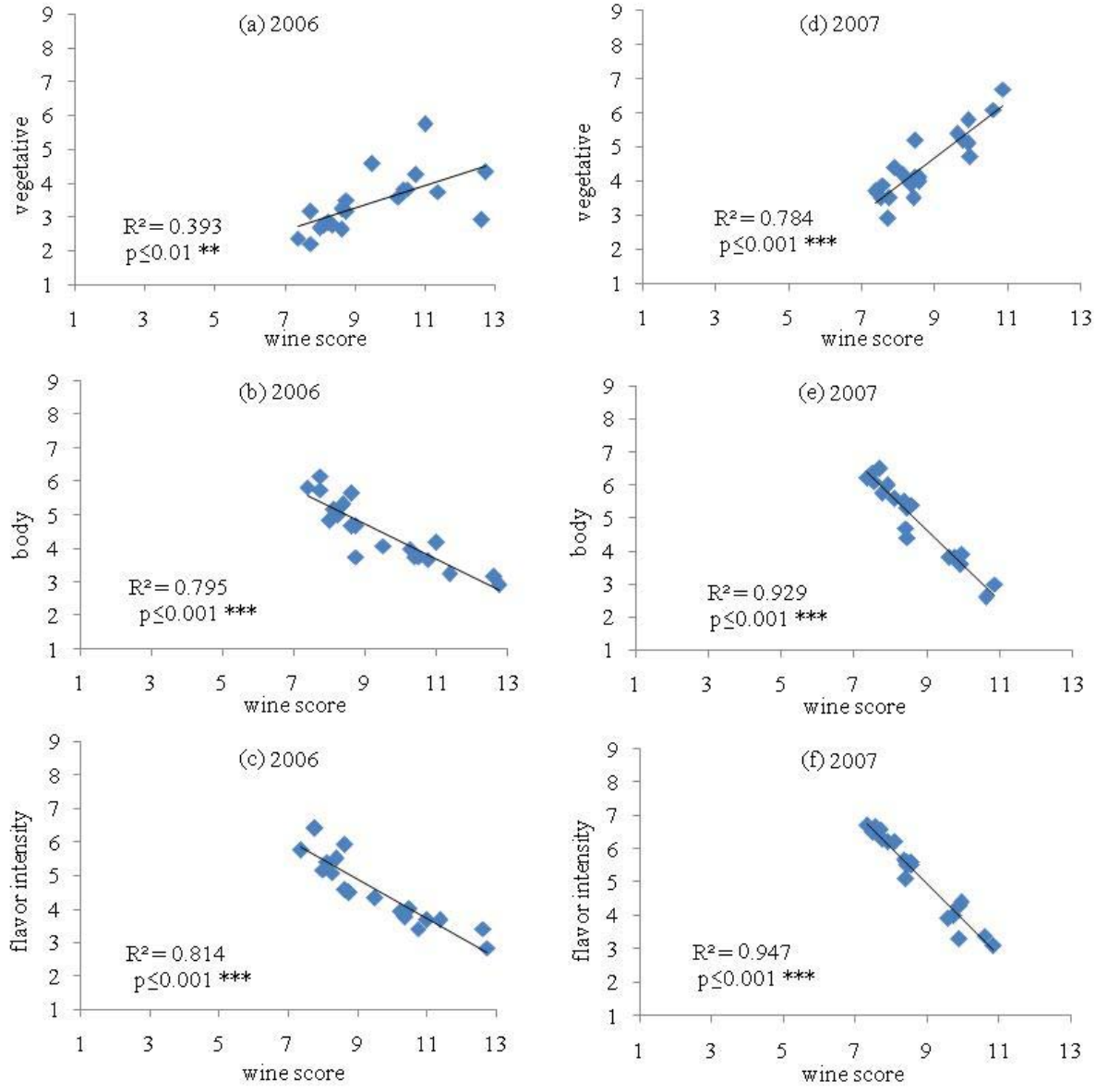


Figure 5.1: Linear regression of attributes and wine scores from the expert panel in 2006 and 2007 treatment wines.

5.4.3 Descriptive Analysis

The analysis of variance (ANOVA) of the descriptive analysis testing for 2005, 2006, and 2007 is presented in Tables 5.5, 5.6 and 5.7, respectively, and indicates many differences due to: wines, judges (panelists), session replications and their interactions. The flavor attributes are presented in capital letters and the visual or aroma attributes are in lower case.

Treatment wines: The descriptive analysis showed many significant differences among the 20 treatment wines made from the crop load and °Brix treatments in each year. Treatment wines were significantly different in the attributes: red color, color depth, sulfides, BERRY, mouth feel (MF)/BODY, ETHANOL BURN (ETOH), and DURATION in each year. Wines were significantly different in two out of three years for the attributes: berry, jam/dried fruit (df), VEGETATIVE and JAM. There were no differences in the attributes acidity, herbaceous, ASTRINGENCY or woody.

Judges: The Judges contributed to a significant source of variation in all years, and in most cases $p < 0.001$. Color depth in 2006 was the only attribute within all years measured that had no significant differences due to judges.

Replications: Replications were a significant source of variation for red color, color depth, BERRY and ASTRINGENCY in two out of the three years. Sulfides, volatile acidity, ETHANOL BURN, and VEGETATIVE were significant in one of the three years. Although significant differences occurred each year, there were no consistent trends among the three years. Sulfide differences in 2005 were most likely a winemaking issue since they did not persist among replications in the following years.

Interactions: A significant interaction between wine and judge (W x J) occurred on the attributes: color depth, ETHANOL BURN, berry and MF/BODY in two of the three years. The attributes: BERRY, sulfides, JAM and vegetative were significant in only one year. These differences indicate that the attribute terms were not used the same way by all judges, however there were no consistent interactions among all years. There were very few significant differences due to the interaction of wine and descriptive analysis replications (W x R) in any year. The only significant interactions were in jam/dried fruit in 2005 and red color and jam/dried fruit in 2006. There were no significant interactions in 2007.

A significant judge by replication (J x R) interaction occurred in 2005 and 2006 for the attributes jam/df, berry and BERRY—indicating that panelists were reproducible between sessions and years.

Table 5.5: Analysis of variance of descriptive analysis attribute ratings on wines made from crop load and extended ripening treatments for three seasons. F ratios are for wines, judges, session replications (reps), and their interactions.

	2005 F-ratios						
Attributes	**Wines (W)**	**Judges (J)**	**Reps (R)**	**W x J**	**W x R**	**J x R**	**EMS**
Visual and Aroma							
red color	12.84***	5.61***	6.49*	0.90	1.46	1.14	2.50
color depth	24.47***	24.37***	6.22*	1.49*	1.50	1.40	1.48
sulfides	1.79*	10.77***	12.13***	0.80	1.00	1.17	3.38
volatile acidity	0.89	7.77***	9.63**	1.03	1.27	1.07	1.83
berry	1.79*	26.73***	1.87	1.07	0.70	3.02*	2.95
jam/dried fruit	2.22**	23.52***	2.81	1.09	1.76*	4.98**	2.58
herbaceous	1.11	9.04***	2.38	0.97	1.20	1.04	2.87
vegetative	1.95*	11.95***	0.47	0.80	1.25	2.32	3.13
woody	0.66	4.62**	0.89	0.75	0.74	0.89	4.46
Flavor							
ACIDITY	2.05*	27.25***	0.14	1.04	0.42	0.62	3.03
BERRY	3.48***	20.90***	4.84*	0.80	1.01	4.02**	2.56
JAM	1.52	26.57***	2.82	0.84	1.16	0.75	3.32
VEGETATIVE	2.46**	7.74***	0.10	1.23	0.69	0.19	1.72
ASTRINGENCY	1.56	12.76***	1.52	1.00	0.99	0.45	2.77
MF/BODY	2.97***	6.98***	1.18	1.14	0.60	0.38	2.67
ETHANOL BURN	2.06*	35.16***	0.00	1.52*	0.87	0.99	2.86
DURATION	2.02*	21.80***	0.01	0.72	0.44	0.59	2.83

*, **, ***, ns indicate significance at $p<0.05$, 0.01, 0.001, or not significant respectively by LSD.

Table 5.6: Analysis of variance of descriptive analysis attribute ratings on wines made from crop load and extended ripening treatments for 2006. F ratios are for wines, judges, session replications (reps), and their interactions.

	2006 F-ratios						
Attributes	**Wines (W)**	**Judges (J)**	**Reps (R)**	**W x J**	**W x R**	**J x R**	**EMS**
Visual and Aroma							
Red Color	16.66***	5.57***	1.90	1.08	2.13*	0.13	1.93
Color Depth	37.95***	1.61	9.87**	1.45	1.35	3.29*	1.31
sulfides	2.64**	19.94***	2.01	1.59*	1.71	2.96*	1.83
volatile acidity	0.50	38.89***	0.18	0.78	0.91	0.08	0.42
berry	1.53	25.31***	0.72	1.80**	0.29	7.48***	0.88
jam/dried fruit	1.34	11.64***	0.55	0.65	0.88	3.50*	1.94
herbaceous	1.51	46.11***	0.79	1.29	1.43	2.87*	0.95
vegetative	1.08	41.50***	0.02	0.70	0.67	2.17	1.92
woody	0.99	36.32***	2.65	0.75	1.04	0.80	0.69
Flavor							
ACIDITY	0.69	66.01***	0.69	0.79	0.75	3.72**	1.39
BERRY	6.00***	53.35***	4.46*	1.90**	1.12	10.04***	0.71
JAM	8.18***	134.63***	2.30	2.75***	2.08*	10.25***	0.52
VEGETATIVE	1.37	108.9***	4.91*	1.18	0.52	0.98	0.76
ASTRINGENCY	1.40	150.53***	6.46*	1.41	1.22	1.44	1.02
MF/BODY	8.99***	8.36***	0.06	1.53*	1.06	2.20	1.21
ETHANOL BURN	5.32***	133.56***	15.15***	1.47*	1.00	0.36	0.89
DURATION	4.39***	22.37***	5.80*	1.05	0.95	2.94*	1.38

*, **, ***, ns indicate significance at $p<0.05$, 0.01, 0.001, or not significant respectively by LSD.

Table 5.7: Analysis of variance of descriptive analysis attribute ratings on wines made from crop load and extended ripening treatments for 2007. F ratios are for wines, judges, session replications (reps), and their interactions.

	2007 F-ratios						
Attributes	**Wines (W)**	**Judges (J)**	**Reps (R)**	**W x J**	**W x R**	**J x R**	**EMS**
Visual and Aroma							
Red Color	22.83***	9.36***	5.16*	1.36	0.98	1.23	2.35
Color Depth	44.87***	5.14***	0.08	1.94***	0.82	2.78*	1.39
sulfides	2.44**	15.89***	2.32	1.10	0.99	1.45	1.83
volatile acidity	0.91	29.24***	4.88*	0.63	0.84	2.29	0.26
berry	3.10***	49.11***	1.70	1.55*	0.81	2.28	1.60
jam/dried fruit	2.73***	33.98***	0.11	1.69**	0.78	1.07	2.04
herbaceous	1.01	8.39***	2.74	0.97	1.28	0.35	2.64
vegetative	1.87*	12.81***	0.16	1.75**	0.80	0.56	3.23
woody	0.64	19.88***	0.10	0.82	1.14	2.29	2.49
Flavor Attributes							
ACIDITY	0.91	45.68***	0.05	1.23	0.57	2.54*	1.79
BERRY	2.91***	19.47***	0.39	1.11	1.42	0.95	2.48
JAM	2.93***	58.54***	1.73	1.32	0.75	1.88	1.64
VEGETATIVE	2.04*	3.34**	0.54	0.84	0.72	0.84	2.50
ASTRINGENCY	1.59	15.46***	8.23**	0.76	0.46	2.72*	2.33
MF/BODY	4.83***	3.34**	0.38	1.51*	1.43	0.90	2.55
ETHANOL BURN	2.96***	57.05***	0.01	0.89	0.58	2.73*	2.20
DURATION	4.81***	45.66***	0.35	1.31	0.90	0.84	1.36

*, **, ***, ns indicate significance at $p<0.05$, 0.01, 0.001, or not significant respectively.

Treatment effects: There were many significant differences in all years among the attributes for the treatment wines (Tables 5.8, 5.9, 5.10).

Vegetative (veg) aroma and flavor (VEG) significantly decreased as °Brix at harvest increased in 2005 and 2007. The highest average vegetative aroma and individual score was in the 20 cl in both 2005 and 2007. In contrast, the lowest overall average for vegetative aroma was in the UN in both 2005 and 2007 and VEG flavor was lowest in the UN in 2007.

The scores for attributes: color depth, berry, BERRY, MF/BODY, DURATION and ETOH increased as °Brix at harvest increased in all years.

Table 5.8: The effect of crop load and °Brix at harvest on mean intensity values for wine attributes from descriptive analysis on 2005 treatment wines.

	2005 Attributes: Visual, Aroma and FLAVOR (below)					
Treatment	**red intensity**	**color depth**	**sulfides**	**berry**	**jam**	**vegetative**
20/22.5º	7.51 A	1.88 JK	2.01 D	2.02 EF	1.04 F	3.69 AB
20/24.0º	7.56 A	2.15 IJK	2.40 BCD	2.31 DEF	3.74 A	2.43 BCDEF
20/25.5º	5.00 CDE	3.33 GH	4.36 A	2.67 ABCDEF	1.50 EF	4.51 A
20/27.0º	5.79 BC	5.97 BCD	2.36 CD	4.11 A	1.60 EF	2.00 CDEF
20/28.5º	2.48 H	6.70 AB	2.34 CD	3.66 ABCD	1.93 CDEF	1.90 DEF
40/22.5º	7.20 AB	1.58 K	2.04 D	2.77 ABCDEF	2.18 CDEF	2.43 BCDEF
40/24.0º	2.80 GH	2.29 HIJK	3.73 ABC	2.71 ABCDEF	2.06 CDEF	2.83 BCDEF
40/25.5º	5.04 CDE	4.29 FG	2.45 BCD	3.93 AB	1.73 DEF	3.00 ABCDE
40/27.0º	2.96 FGH	5.84 BCD	2.97 ABCD	3.81 ABCD	1.60 EF	2.70 BCDEF
40/28.5º	4.71 CDE	7.31 A	1.81 D	3.84 ABCD	2.39 ABCDEF	1.35 F
60/22.5º	5.41 CD	1.74 JK	4.01 AB	1.69 F	3.64 AB	3.53 ABC
60/24.0º	3.14 FGH	2.81 HIJ	2.55 BCD	3.22 ABCDE	2.06 CDEF	3.42 ABCD
60/25.5º	2.37 H	5.48 CDE	1.80 D	2.95 ABCDEF	2.26 BCDEF	1.75 EF
60/27.0º	5.38 CDE	5.10 DEF	2.00 D	2.79 ABCDEF	2.79 ABCDE	2.05 CDEF
60/28.5º	4.25 DEF	6.35 ABC	4.00 AB	4.04 A	3.13 ABCD	2.20 BCDEF
UN/22.5º	2.71 GH	1.43 K	1.88 D	2.71 ABCDEF	1.55 EF	2.94 ABCDE
UN/24.0º	1.93 H	5.17 DEF	2.75 ABCD	2.44 BCDEF	1.68 EF	2.65 BCDEF
UN/25.5º	2.02 H	3.02 HI	2.45 BCD	3.43 ABCDE	1.48 EF	1.77 EF
UN/27.0º	3.99 EFG	4.59 EF	2.81 ABCD	3.85 ABC	3.21 ABC	2.12 BCDEF
UN/28.5º	4.30 DEF	5.32 CDEF	2.54 BCD	2.40 CDEF	2.40 ABCDEF	2.04 CDEF
Treatment	**ACIDITY**	**BERRY**	**VEG.**	**MF/BODY**	**ETOH**	**DURATION**
20/22.5º	4.59 ABCD	1.72 G	3.22 A	3.68 BCDEF	2.70 F	2.52 D
20/24.0º	2.77 E	2.71 EFG	2.19 ABCDE	2.76 EFG	4.16 BCDEF	3.29 CD
20/25.5º	5.01 ABC	3.66 CDEF	1.46 DEF	4.17 ABCDE	4.05 BCDEF	3.92 BCD
20/27.0º	3.89 CDE	4.65 ABC	0.95 F	4.29 ABCD	4.52 ABCDE	4.63 ABC
20/28.5º	4.56 ABCD	4.65 ABC	0.97 F	4.04 ABCDE	4.54 ABCDE	4.77 ABC
40/22.5º	4.02 BCDE	2.45 FG	2.41 ABCDE	2.94 DEFG	3.25 EF	3.46 CD
40/24.0º	5.97 A	3.55 CDEF	2.82 AB	2.42 FG	3.13 EF	4.05 BC
40/25.5º	3.37 DE	3.76 BCDEF	1.46 DEF	4.50 ABC	3.40 DEF	4.03 BC
40/27.0º	4.31 BCDE	5.55 A	2.33 ABCDE	5.14 A	4.89 ABCD	5.61 A
40/28.5º	4.21 BCDE	5.10 AB	1.30 EF	4.84 AB	4.30 ABCDE	4.69 ABC
60/22.5º	4.59 ABCD	2.66 EFG	2.68 ABC	3.02 DEFG	3.26 EF	3.81 BCD
60/24.0º	5.47 AB	3.40 CDEF	2.26 ABCDE	4.04 ABCDE	3.50 CDEF	3.75 BCD
60/25.5º	4.90 ABCD	4.04 BCDE	1.72 BCDEF	4.65 ABC	4.34 ABCDE	4.47 ABC
60/27.0º	4.06 BCDE	3.78 BCDEF	1.39 EF	5.01 AB	5.77 A	5.61 A
60/28.5º	4.62 ABCD	4.59 ABC	1.31 EF	4.14 ABCDE	4.94 ABC	4.36 ABC
UN/22.5º	6.00 A	3.27 CDEF	1.65 CDEF	2.08 G	3.57 BCDEF	4.38 ABC
UN/24.0º	4.92 ABCD	2.94 DEFG	2.62 ABCD	4.21 ABCDE	4.07 BCDEF	4.61 ABC
UN/25.5º	4.95 ABC	3.73 BCDEF	1.75 BCDEF	3.36 CDEFG	4.46 ABCDE	4.60 ABC
UN/27.0º	3.88 CDE	4.32 ABCD	1.76 BCDEF	4.61 ABC	4.40 ABCDE	5.25 AB
UN/28.5º	4.30 BCDE	3.94 BCDE	1.25 EF	4.78 ABC	5.06 AB	4.65 ABC

Means within columns with different letters differ significantly at $p<0.05$ by LSD.

Table 5.9: The effect of crop load and °Brix at harvest on aroma attributes from descriptive analysis on treatment wines in three years. Means with different letters differ significantly by $p<0.05$ by LSD.

	2006 Attributes: Visual, Aroma and FLAVOR (below)				
Treatment	**red intensity**	**color depth**	**sulfides**	**berry**	**vegetative**
20/22.5°	6.72 AB	2.55 GH	2.48 BCDEFGH	2.88	3.33
20/24.0°	6.25 BC	3.12 FG	3.51 AB	3.01	2.26
20/25.5°	6.09 BC	3.90 EF	3.40 ABC	2.99	2.98
20/27.0°	3.64 FGHI	5.89 BC	2.84 ABCDEFG	3.01	3.05
20/28.5°	5.31 CD	6.00 BC	1.49 H	3.25	2.53
40/22.5°	6.77 AB	5.02 CD	2.64 ABCDEFG	2.69	2.69
40/24.0°	2.74 HIJKL	2.17 GHI	3.37 ABC	3.10	2.15
40/25.5°	2.08 KL	2.65 GH	2.25 CDEFGH	3.67	2.48
40/27.0°	3.35 GHIJ	4.50 DE	3.72 A	2.62	2.47
40/28.5°	3.90 EFGH	8.09 A	2.64 ABCDEFG	3.67	2.33
60/22.5°	7.68 A	1.20 IJ	3.72 A	2.02	2.94
60/24.0°	3.32 GHIJK	2.06 HI	1.77 GH	3.09	3.48
60/25.5°	2.40 IJKL	5.04 CD	2.09 EFGH	3.09	1.92
60/27.0°	1.81 L	6.30 B	2.20 CDEFGH	3.54	2.60
60/28.5°	4.30 DEFG	7.34 A	1.94 EFGH	3.00	1.99
UN/22.5°	5.03 CDE	0.99 J	1.81 FGH	3.02	2.15
UN/24.0°	1.93 L	3.11 FG	3.32 ABCD	3.12	2.73
UN/25.5°	2.29 JKL	4.52 DE	3.09 ABCDE	2.76	2.49
UN/27.0°	4.07 EFG	7.57 A	3.01 ABCDEF	3.36	2.07
UN/28.5°	4.68 DEF	7.67 A	2.14 DEFGH	3.58	1.90

	2006 Attributes: Flavor					
Treatment	**BERRY**	**JAM**	**VEG.**	**MF/BODY**	**ETOH**	**DURATION**
20/22.5°	3.27 DEF	2.11 FGHI	2.81	3.50 FGH	2.29 EFGH	3.97 CDEF
20/24.0°	3.05 EFG	1.90 HI	1.94	2.69 HIJ	2.10 FGH	3.11 EFG
20/25.5°	3.06 EFG	2.27 FGH	3.03	3.87 CDEF	2.56 DEFGH	4.08 CDE
20/27.0°	3.90 ABCD	2.54 EFG	1.97	4.74 BC	3.30 ABCD	4.76 ABC
20/28.5°	4.50 A	3.38 ABC	2.13	4.95 B	4.07 A	5.32 AB
40/22.5°	3.06 EFG	2.14 FGHI	2.55	3.06 FGHI	2.16 EFGH	3.53 DEF
40/24.0°	2.74 FG	2.43 EFGH	2.60	3.60 EFGH	2.83 CDEF	3.51 DEF
40/25.5°	3.50 CDE	2.69 DEF	2.83	3.85 CDEF	2.23 EFGH	4.28 BCD
40/27.0°	3.31 DEF	2.43 EFGH	2.55	2.87 GHIJ	2.79 CDEF	3.63 DEF
40/28.5°	4.43 A	3.78 AB	2.40	5.96 A	4.00 A	5.40 A
60/22.5°	2.34 G	1.52 I	2.77	2.03 J	1.89 GH	2.40 G
60/24.0°	2.84 EFG	1.92 GHI	2.49	2.91 FGHIJ	2.29 EFGH	3.58 DEF
60/25.5°	3.55 BCDE	2.58 EF	2.23	3.39 FGH	2.73 CDEFG	4.18 CD
60/27.0°	4.12 ABC	3.03 CDE	2.37	4.66 BDC	3.53 ABC	4.79 ABC
60/28.5°	4.28 AB	3.25 BCD	1.98	4.49 BCDE	3.51 ABC	4.49 ABCD
UN/22.5°	2.95 EFG	2.11 FGHI	2.51	2.29 IJ	1.88 H	2.97 FG
UN/24.0°	2.97 EFG	2.15 FGHI	2.04	3.74 DEFG	2.61 DEFGH	3.92 CDEF
UN/25.5°	3.34 DEF	2.11 FGHI	2.45	3.42 FGH	2.94 CDE	3.82 CDEF
UN/27.0°	4.50 A	3.23 BCD	2.04	4.79 BC	3.14 BCD	4.69 ABC
UN/28.5°	4.13 ABC	3.90 A	2.14	5.33 AB	3.78 AB	4.80 ABC

Table 5.10: The effect of crop load and °Brix at harvest on wine attributes from descriptive analysis on 2007 treatment wines.

	2007 Attributes: Visual, Aroma and FLAVOR (below)					
Treatment	**red intensity**	**color depth**	**sulfides**	**berry**	**jam**	**veg.**
20/22.5º	7.88 A	0.84 K	2.98 AB	2.40 J	1.75 G	3.61 AB
20/24.0º	7.19 AB	3.14 I	2.41 BCDE	2.88 GHIJ	2.99 ABCDE	3.98 A
20/25.5º	6.25 BC	6.16 EFG	1.45 F	3.23 EFGHIJ	3.74 A	2.95 ABC
20/27.0º	3.14 FGH	5.84 FG	2.27 BCDEF	3.92 BCDE	2.20 EFG	2.02 CDE
20/28.5º	5.22 CD	5.73 G	1.99 CDEF	3.97 BCDE	3.48 ABC	2.46 BCD
40/22.5º	7.70 A	1.31 K	3.71 A	2.79 HIJ	1.79 FG	2.65 BCD
40/24.0º	3.32 EFG	6.15 EFG	1.83 DEF	3.59 CDEFGH	2.94 ABCDE	3.06 ABC
40/25.5º	2.00 IJK	6.64 DEF	2.18 BCDEF	3.76 BCDEF	1.96 FG	1.16 E
40/27.0º	3.09 FGH	7.02 BCD	2.00 CDEF	3.62 CDEFGH	3.28 ABCD	2.60 BCD
40/28.5º	2.42 GHIJ	7.85 A	1.79 DEF	4.60 AB	3.70 A	2.84 ABCD
60/22.5º	5.97 C	2.22 J	2.89 ABC	2.73 IJ	2.53 CDEFG	3.62 AB
60/24.0º	2.86 FGHI	4.84 H	1.55 EF	2.97 FGHIJ	2.37 DEFG	1.64 DE
60/25.5º	1.61 JK	7.48 ABC	2.84 ABC	3.88 BCDE	2.39 DEFG	2.15 CDE
60/27.0º	2.15 HIJK	7.43 ABCD	2.01 CDEF	3.72 CDEFG	2.58 CDEFG	1.94 CDE
60/28.5º	2.78 GHI	7.74 AB	1.44 F	4.28 ABC	3.65 AB	1.89 CDE
UN/22.5º	4.24 DE	2.32 J	2.61 BCD	3.17 EFGHIJ	1.91 FG	2.38 CD
UN/24.0º	1.39 JK	4.79 H	2.42 BCDE	3.55 CDEFGHI	2.68 CDEFG	2.34 CDE
UN/25.5º	1.21 K	6.93 CDE	1.51 EF	4.85 A	2.72 BCDEF	2.15 CDE
UN/27.0º	3.85 EF	6.85 CDE	1.72 DEF	3.29 DEFGHI	3.25 ABCD	1.92 CDE
UN/28.5º	3.08 FGH	7.79 AB	1.62 EF	4.11 ABCD	3.77 A	2.09 CDE
Treatment	**BERRY**	**JAM**	**VEG.**	**MF/BODY**	**ETOH**	**DURATION**
20/22.5º	2.28 H	1.63 FG	2.98 A	3.13 GHI	1.88 DE	3.18 H
20/24.0º	3.24 DEFGH	3.04 ABC	2.59 AB	3.13 GHI	2.84 BCD	4.45 DEF
20/25.5º	3.14 EFGH	3.48 A	1.73 BCDEF	4.43 CDEF	3.44 AB	4.49 CDEF
20/27.0º	4.07 BCDE	2.58 BCD	1.56 BCDEF	4.70 BCDE	3.16 BC	4.64 BCDE
20/28.5º	4.18 BCDE	3.32 ABC	1.17 DEF	4.61 CDE	3.45 AB	4.70 BCDE
40/22.5º	2.81 GH	1.68 EFG	2.41 ABC	2.58 HI	2.08 DE	4.09 EFG
40/24.0º	3.60 CDEFG	2.59 BCD	1.95 ABCDE	4.02 DEFG	2.79 BCD	4.65 BCDE
40/25.5º	4.31 BCD	1.95 DEFG	0.89 EF	5.30 ABC	3.53 AB	4.69 BCDE
40/27.0º	4.46 ABC	2.96 ABC	1.50 CDEF	4.61 CDE	3.59 AB	5.31 AB
40/28.5º	4.18 BCDE	3.10 ABC	0.93 EF	5.97 A	4.22 A	5.64 A
60/22.5º	2.86 FGH	2.58 BCD	2.54 ABC	3.75 EFG	2.10 DE	3.45 GH
60/24.0º	3.63 CDEFG	2.51 CDE	2.20 ABCD	2.35 I	2.01 DE	2.94 H
60/25.5º	4.35 ABC	2.87 ABC	0.95 EF	4.65 BCDE	3.42 AB	4.55 BCDE
60/27.0º	3.92 CDEF	3.23 ABC	1.02 EF	4.84 BCDE	3.26 AB	4.69 BCDE
60/28.5º	4.51 ABC	2.47 CDEF	0.88 F	4.92 ABCD	3.57 AB	4.31 DEF
UN/22.5º	3.18 EFGH	1.78 DEFG	1.74 BCDEF	3.45 FGH	2.21 CDE	3.23 H
UN/24.0º	3.65 CDEFG	1.33 G	1.74 BCDEF	3.38 FGHI	1.78 E	3.72 FGH
UN/25.5º	5.38 A	2.65 ABCD	0.96 EF	4.69 BCDE	3.27 AB	4.24 EFG
UN/27.0º	4.13 BCDE	3.40 AB	1.24 DEF	5.72 AB	3.71 AB	5.25 ABC
UN/28.5º	5.07 AB	2.96 ABC	1.22 DEF	5.27 ABC	3.74 AB	5.09 ABCD

Means within columns with different letters differ significantly at $p<0.05$ by LSD.

Correlations among treatment wines and sensory attributes: The correlation coefficients among the descriptive analysis attributes are presented in Tables 5.11, 5.12 and 5.13, for 2005, 2006 and 2007, respectively.

The following attributes had highly significant positive correlations in each year: color depth, berry, BERRY, BODY, DURATION, ETOH, JAM and jam. The positive correlations in color depth indicate that increased color depth was connected to increased berry, BERRY, BODY, ETOH, and DURATION in all years ($p<0.001$) and also to jam and JAM in 2006 and 2007. The flavor and aroma attributes for berry had significant positive correlations in all years for the attributes: BODY, DURATION and ETOH. Likewise, the flavor attributes BODY, DURATION and ETOH had highly significant positive correlations in all years—$p<0.01$ in 2005 and $p<0.001$ in 2006 and 2007.

Both veg aroma and VEG flavor were negatively correlated with color depth, berry, BERRY, ETOH, BODY and DURATION. These correlations demonstrate that as the veg aroma and VEG flavor ratings increased, ratings for color depth, berry, BERRY, ETOH, and DURATION decreased.

Table 5.11: The correlation matrix of 2005 treatment wines from the descriptive analysis testing. The correlation coefficients highlighted indicate a significance at $p<0.05$

2005	red color	color depth	sulfides	berry	jam	veg	ACID	BERRY	VEG	AST	BODY	ETOH	DUR.
red color	1												
color depth	-0.31	1											
sulfides	-0.07	-0.10	1										
berry	-0.28	0.65	-0.10	1									
jam	0.27	0.01	0.23	-0.13	1								
veg	0.22	-0.60	0.52	-0.48	-0.23	1							
ACID	-0.63	-0.24	0.22	-0.19	-0.41	0.28	1						
BERRY	-0.45	0.82	0.05	0.80	-0.01	-0.51	-0.07	1					
VEG	0.16	-0.65	0.14	-0.61	-0.05	0.48	0.23	-0.66	1				
AST	0.38	-0.10	-0.20	-0.24	-0.30	0.29	0.02	-0.39	0.26	1			
BODY	-0.12	0.80	-0.11	0.47	-0.05	-0.29	-0.35	0.61	-0.43	0.00	1		
ETOH	-0.23	0.71	-0.06	0.40	0.24	-0.58	-0.27	0.66	-0.66	-0.17	0.63	1	
DURATION	-0.55	0.67	-0.05	0.52	0.07	-0.54	-0.01	0.78	-0.53	-0.47	0.57	0.79	1

Table 5.12: The correlation matrix of 2006 treatment wines from the descriptive analysis testing. The correlation coefficients highlighted in green are significant at $p< 0.05$.

2006	red color	color depth	sulfides	berry	BERRY	JAM	BODY	ETOH	DUR
red color	1								
color depth	-0.18	1							
sulfides	0.14	-0.23	1						
berry	-0.45	0.60	-0.47	1					
BERRY	-0.24	0.90	-0.46	0.70	1				
JAM	-0.26	0.85	-0.44	0.77	0.90	1			
BODY	-0.29	0.85	-0.31	0.79	0.87	0.91	1		
ETOH	-0.31	0.86	-0.31	0.66	0.87	0.88	0.90	1	
DURATION	-0.35	0.83	-0.44	0.77	0.91	0.87	0.95	0.89	1

Table 5.13: The correlation matrix for 2007 treatment wines from the descriptive analysis testing. The correlation coefficients highlighted indicate significance at $p<0.05$.

2007	red color	color depth	sulfides	berry	jam	veg	BERRY	JAM	VEG	AST	BODY	ETOH	DUR
red color	1												
color depth	-0.76	1											
sulfides	0.50	-0.74	1										
berry	-0.74	0.82	-0.58	1									
jam	-0.16	0.63	-0.71	0.48	1								
veg	0.71	-0.57	0.37	-0.48	0.06	1							
BERRY	-0.78	0.85	-0.60	0.88	0.45	-0.61	1						
JAM	-0.07	0.59	-0.57	0.32	0.71	0.03	0.39	1					
VEG	0.77	-0.88	0.54	-0.87	-0.42	0.71	-0.84	-0.38	1				
AST	0.42	-0.11	-0.02	-0.32	0.11	0.22	-0.23	0.23	0.24	1			
BODY	-0.50	0.80	-0.52	0.72	0.56	-0.38	0.68	0.55	-0.82	-0.02	1		
ETOH	-0.39	0.84	-0.59	0.72	0.67	-0.32	0.71	0.72	-0.79	0.15	0.91	1	
DURATION	-0.30	0.73	-0.38	0.59	0.62	-0.16	0.59	0.63	-0.63	0.29	0.81	0.89	1

Principal component analysis: A principal component analysis (PCA) was done for the 2005, 2006 and 2007 vintages to analyze treatment and attribute means from the descriptive analysis testing (Figures 5.2, 5.3 and 5.4). Overall, the PCA had similar trends in all years, although some differences between vintages did occur. These trends are demonstrated by the position of the attributes and wine treatments within the PCA diagrams. Emanating from the central origin are vectors representing each attribute tested. The length of the vector may be interpreted as an indication of its influence on that PC. Short vectors indicate attributes of relatively low importance; conversely long vectors indicate high importance. Close alignment of a vector with the PC axis indicates a high correlation between the attribute represented by the axis and the variability between wines explained by the PCA. The principal component plots show the position of each wine as a single point. The closer the treatment wines are to each other or to the nearby attributes, the more strongly correlated they are. In contrast, treatment wines and attributes which are far apart are dissimilar and not strongly correlated to each other or the opposing attributes. The principal component analyses were generated from the correlation matrix in each corresponding year and with no rotation.

Crop load and °Brix at harvest had a strong influence on results from the DA and subsequent PCA diagrams in all years. There were substantial differences between crop loads harvested at the lower °Brix targets i.e. 22.5 and 24.0 and those harvested at the higher targets i.e 27.0 and 28.5 °Brix. Wines at the lower °Brix targets were strongly correlated with vegetative aroma and flavor, sulfides and astringency. In contrast, wines from the higher °Brix targets were strongly correlated with the attributes: berry, BERRY, BODY, DURATION, jam, JAM, ETOH and color depth.

2005: The first two principal components accounted for 50.8 % and 23.7 % of the variance respectively, for a total of 74.5 %. In Figure 5.2, wines are separated along the first PC according to the intensity of their BERRY flavor, DURATION and color depth. To a lesser extent red color also contributed to the separation of wines—demonstrated by the large angle between its vector and the first PC.

Generally, in 2005 most wines with low °Brix levels i.e. 22.5 and 24.0 ºBrix displayed vegetative aromas and flavors. This was the case for all crop loads except for unthinned at 24.0 ºBrix. Three wines with low crop load and low °Brix levels i.e. 20/24.0, 40/22.5 and 20/22.5 appeared brownish-red. Other low °Brix level wines such as: 60/22.5, 60/24, 40/24.0 and UN/22.5 were more red and light in color depth. Wine UN/24.0 was an exception due to more blueish-red tones and deeper color. Berry aroma and flavor, body, ethanol burn and duration characterize most wines with high °Brix levels i.e. 25.5, 27.0 and 28.5—irrespective of crop load.

2006: The first two principal components accounted for 66 % and 25 % of the variance, respectively for a total of 91 % (Figure 5.3). Only these PCs had eigenvalues above one—indicating that only the first two PCs are significant. In Figure 5.3, treatment wines were separated along the first PC according to the intensity of their color depth and BODY. The position of wines on the second PC was determined by red color.

Treatment wines 20/28.5, UN/28.5, 60/28.5, UN/27.0 and 40/28.5 grouped in the first two quadrants signifying strong correlations with the attributes color depth, BERRY, JAM, BODY, DURATION, ETOH, and berry. Treatment wines low in color depth grouped in the opposite direction and included treatments UN/22.5, 60/24.0, 40/24.0 and UN/24.0. Additionally, treatments 40/22.5, 20/22.5, 20/24.0 and 60/22.5 were correlated to high red color—signifying a brownish-red appearance as opposed to bluish-red red color rating which was correlated with UN/25.5, 60/25.5 and 60/27.0.

Overall, the 2006 vintage had differences among the wines in “green” characteristics such as vegetative aroma, flavor and acidity. However, the differences were less noticeable given that these attributes were not significantly different among the twenty wines. Seven of the eight wines made from grapes with °Brix targets of 27.0 and 28.5 were characterized by deep color, high berry aroma, high berry and jam flavors, long finish, thick body and high ethanol burn. Treatment 40/27.0 had lower color and was more correlated to sulfide aroma relative to the other crop loads at 27.0 or 28.5 °Brix targets. All wines made from grapes harvested at 22.5, 24.0 and 25.5 °Brix levels were low in color depth (thin or light). This included treatments: 60/22.5, 20/22.5, 20/24.0 and 40/22.5 while the others displayed more red and blue tints.

2007: In 2007 (Figure 5.4), treatment wines 20/28.5, 40/27.0, 40/28.5, UN/27.0 and UN/28.5 grouped in the first quadrant and correlated with the attributes: JAM, jam, DURATION, ETOH and BODY. Treatment 20/25.5 was also in the first quadrant, but correlated with astringency and to a lesser degree with JAM. There was a strong correlation between the attributes for berry aroma and flavor and treatments: 20/27.0, 60/27.0, 60/25.5, 60/28.5, 40/25.5 and UN/25.5.

On the opposing side were vegetative aroma and flavor in which treatments 40/24.0, 20/24.0, 60/22.5, 40/22.5, and 20/22.5 grouped. These treatments also correlated with sulfide aroma as did the treatments 60/24.0, UN/22.5 and UN/24.0 which were less correlated to the vegetative attributes, but still generally related.

The 2007 season had similar trends relative to 2005 and 2006 despite different seasonal characteristics and average yield between the three years.

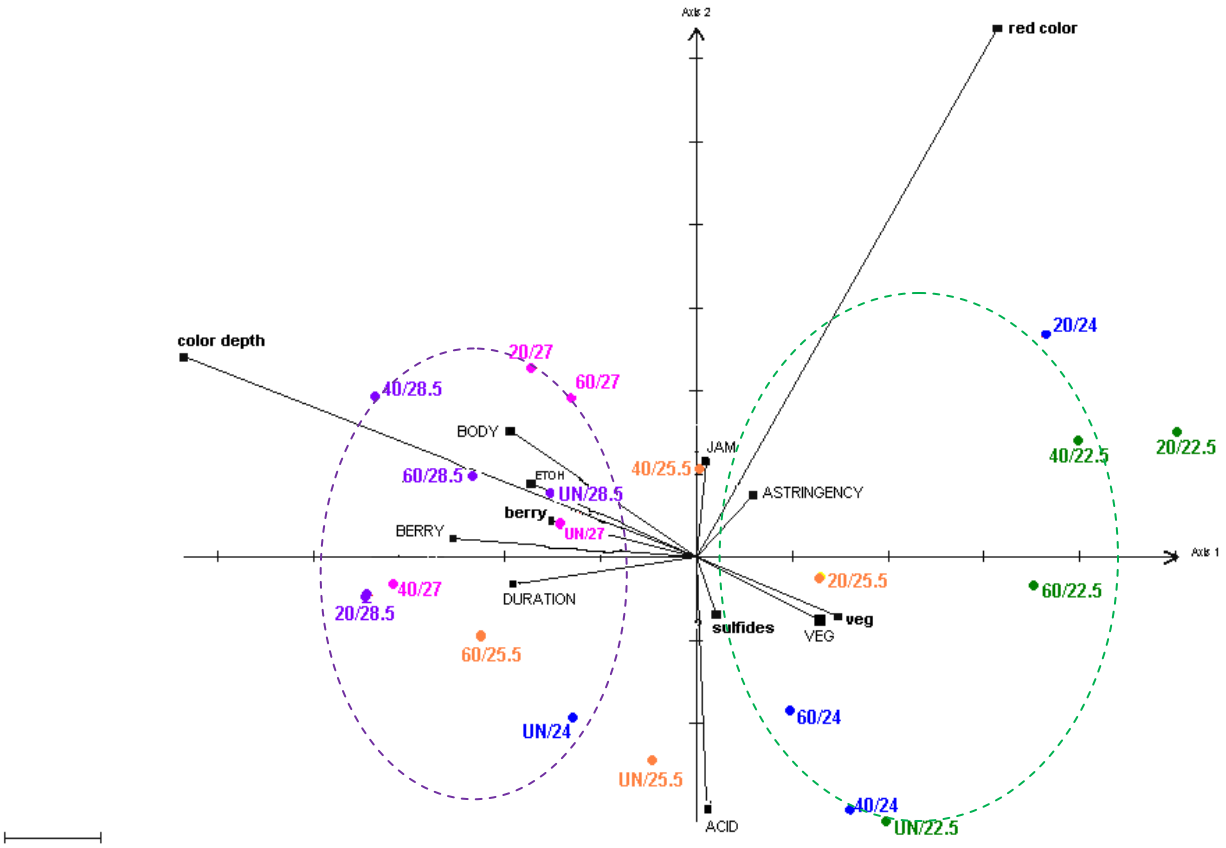


Figure 5.2: Principal component diagram (PCA) of wines from the 20 crop load and °Brix at harvest treatments in 2005. Attributes and their associated axis are in black: lower case lettering represents aroma attributes and uppercase represents flavor attributes. Axis 1: 50.8 % Axis 2: 23.7 %

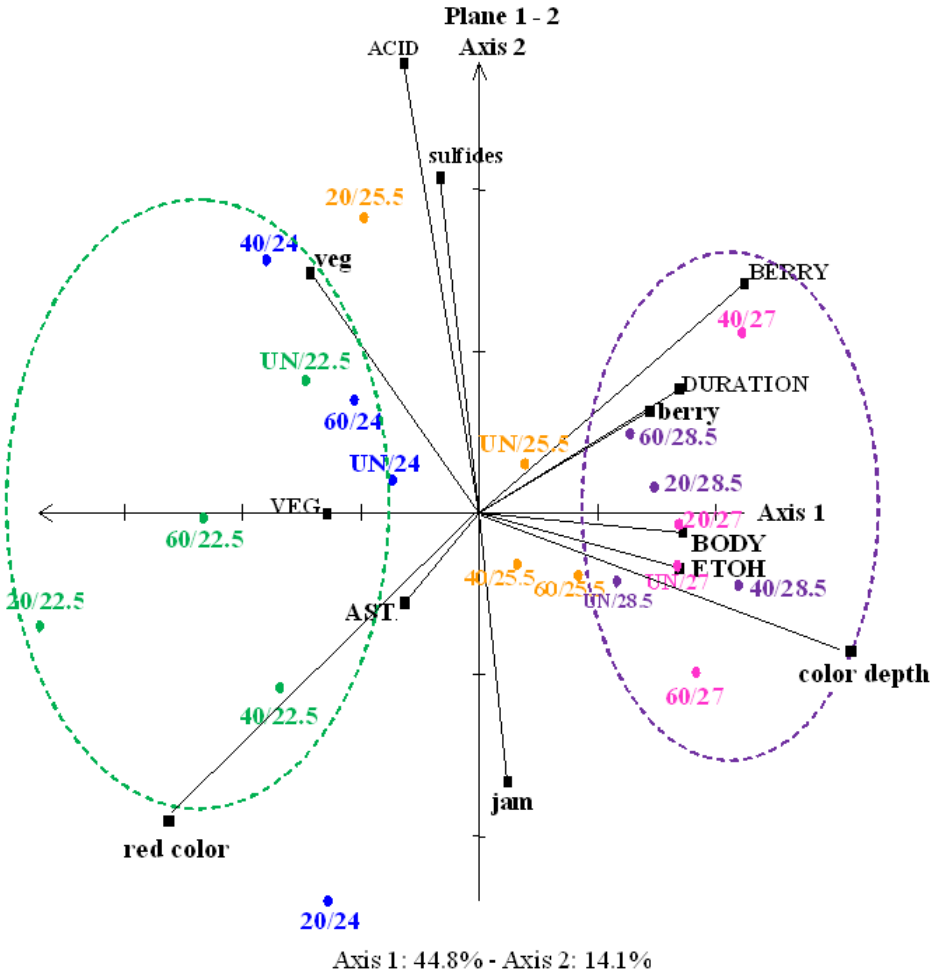


Figure 5.3: Principal component diagram of wines from the 20 crop load and °Brix at harvest treatments in 2006. Attributes and their associated axis are in black: lower case lettering represents aroma attributes and uppercase represents flavor attributes.

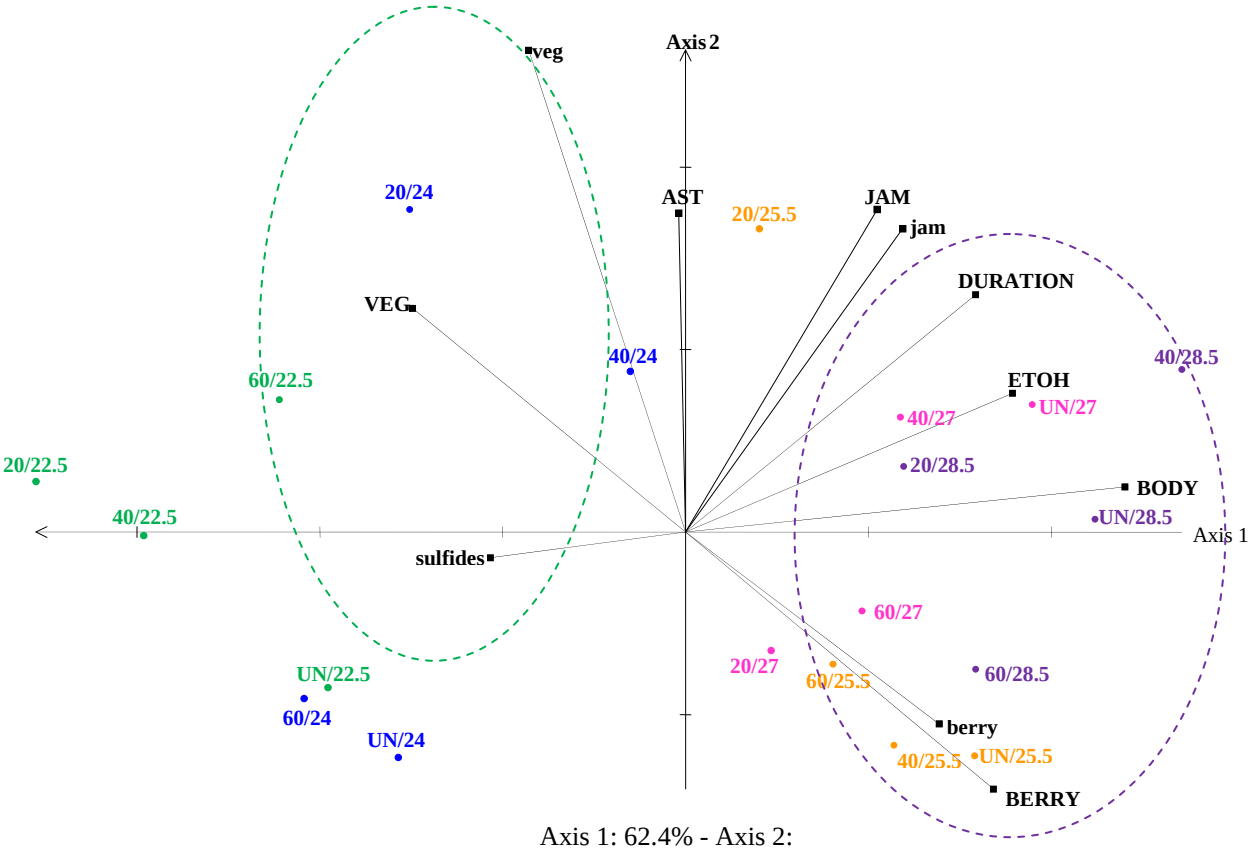


Figure 5.4: Principal component diagram of wines from the 20 crop load and °Brix at harvest treatments in 2007. Attributes and their associated axis are in black: lower case lettering represents aroma attributes and uppercase represents flavor attributes.

5.4.4 Scaled Attribute Ratings (Australian Panel)

The scaled attribute ratings were conducted on the 2005 treatment wines at the University of Adelaide, South Australia and are presented in Table 5.14. Crop load did not significantly affect any attribute ratings. However, °Brix at harvest had a significant effect on flavor intensity, body and quality ($p<0.001$) in that ratings increased as °Brix at harvest increased. Moreover, the highest ratings were consistently at the 28.5 °Brix target. For the attribute 'quality' there were no significant differences between wines harvested at the 27.0 and 28.5 °Brix target; however, the

28.5 °Brix target had a higher rating. In addition, a significant interaction between crop load and °Brix occurred with the flavor intensity, body and quality ratings.

Table 5.14: Effect of crop load and target °Brix at harvest on four attributes rated on 2005 treatment wines by an Australian panel.

Treatment	flavor intensity	vegetative	body	quality
Crop load				
20	5.0	4.4	4.6	4.5
40	5.0	4.5	4.6	4.5
60	5.2	4.3	4.8	4.7
UN	5.0	4.5	4.6	4.4
	ns	ns	ns	ns
Brix				
22.5	4.4 a	4.5	4.1 a	4.1 a
24.0	4.8 b	4.3	4.3 b	4.3 ab
25.5	5.0 b	4.4	4.7 c	4.5 b
27.0	5.3 c	4.3	5.0 d	4.8 c
28.5	5.7 d	4.5	5.3 e	5.1 c
	***	ns	***	***
Interaction	***	ns	**	**

Means with columns separated by different letters differ significantly at p<0.001 by LSD. *, **, ***, ns indicate significance at 0.05, 0.01, 0.001 and not significant

5.4.5 Consumer Testing

A likability rating including 75 consumers was conducted on the 2006 treatment wines (Table 5.15 and Figure 5.5). Pertinent results of the survey administered to obtain demographics of the consumers studied are as follows. The consumer group included 17 different countries of birth, however 50 % were born in Australia and all were Australian residents. Fifty four % of respondents spend less than $20 AUD, on average, on a bottle of wine and 80 % spend less than $25 on a bottle. The most preferred price range was $15 - $19.99 AUD, with 38 % of respondents in this range. Seventy six % of respondents reported that they consume wine at least “a few times a week”. Shiraz was the favorite grape variety of respondents, followed by Cabernet Sauvignon.

Significant differences occurred due to °Brix at harvest. Wines which were harvested at the 22.5 °Brix target scored significantly lower than those harvested at higher °Brix targets. There were no differences due to crop load. The interaction showed that overall, the highest rating was in the 40/27.0 treatment followed by the 20/27.0; however, neither were statistically different from each other, even though they were significantly different from both the 60 cl and UN (Figure 5.5).

Table 5.15: Effect of crop load and target °Brix at harvest on the likability rating of 2006 treatment wines. Ratings are based on the 1-9 hedonic scale.

Treatment	2006 likability score
Crop load	
20	4.6
40	4.7
60	4.6
UN	4.6
	ns
Brix	
22.5	4.3 a
24.0	4.6 b
25.5	4.7 b
27.0	4.8 b
28.5	4.8 b
	**
Interaction	ns

Means with columns separated by different letters differ significantly at p<0.001 by LSD. *, **, ***, ns indicate significance at 0.05, 0.01, 0.001 and not significant respectively

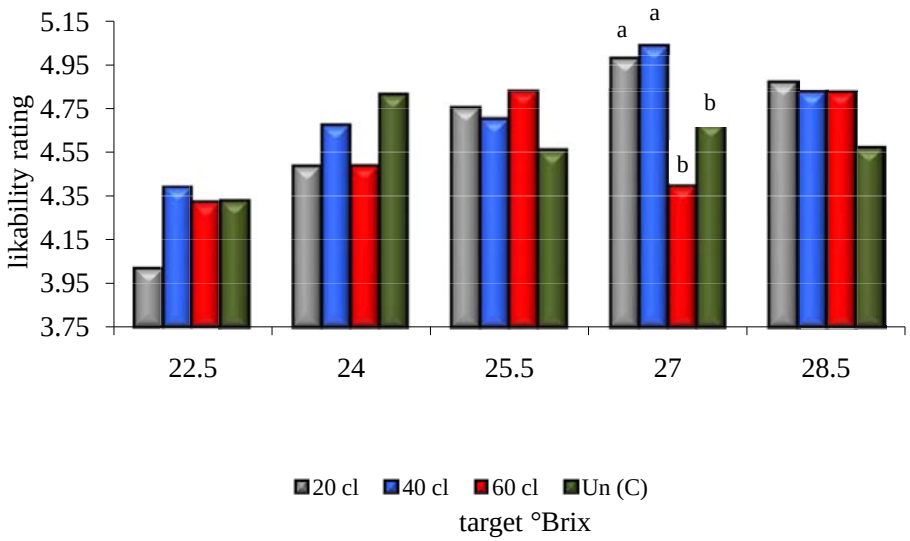


Figure 5.5: The effects of crop load and °Brix at harvest on consumer likability rating in 2006 treatment wines.

5.5 Discussion

5.5.1 Difference Testing

Results of the difference testing indicated that there were more differences in the lower °Brix targets, particularly at 22.5 °Brix and to some extent 24.0 °Brix relative to the higher °Brix targets. Most likely this was due to a large inconsistency between individual berries within a single cluster. Degrees °Brix for the overall must can measure near a target °Brix; however, the actual °Brix per berry within a given cluster is known to differ substantially (Kasimatis *et al.* 1975, Glynn and Boulton 2001, Tarter and Keuter 2005). The collective work of Coombe has reported that each berry develops independently—this may derive as early as anthesis. Therefore, this asynchrony must be managed through adequate sampling populations (Coombe 1992). Furthermore, color per berry and extraction potential may have been variable due to different paces of ripening within each berry, particularly at lower maturity. Moreover, panelists

informally declared that color differences were their biggest indicator for deciphering between wines in the triangle test. Color is known to be a primary indicator for judgments made on food or beverage products (Lawless and Heymann 1998, Delwiche 2004) and especially for wine (Gawel 1997, Parr *et al.* 2003). These results suggest that greater differences exist among grapes harvested at lower maturities. Additionally, the triangle test proved a successful method for analyzing fermentation replications and facilitating blending decisions among the treatment wines.

5.5.2 Expert Panel

Expert quality score: The 'expert' panel in the present study was consistent with previous research defining wine expertise. Wine experts have been defined as demonstrating the conceptual knowledge of typical features which go together with a specific wine style (Parr *et al.* 2003). This includes categorical structure assisting experts to recall (Hughson and Boakes 2002) and match (Solomon 1991) wine descriptions, and prototypes of wine styles (Parr *et al.* 2003).

Crop load influenced expert panel wine score significantly in 2006 and 2007, but overall the effects of crop load were not as substantial as differences due to °Brix at harvest. Certainly, crop reduction did not improve wine score. Therefore, treatments which underwent crop reduction (i.e. 20 cl, 40 cl, and 60 cl) were not elevated to a higher quality wine program and/or price point. In fact, the unthinned treatment had more optimal scores relative to the lower crop loads. These results support those of Zamboni *et al.* (1996) who reported that tasting panels preferred wines made from vines with higher bud numbers at pruning for the variety Sauvignon.

The expert panel wine scores became significantly lower (better) as °Brix at harvest increased. There were no significant increases in wine score past 27.0 °Brix, suggesting that optimal wine

quality can be achieved at 27.0 °Brix rather than undergoing extended ripening to 28.5 °Brix. The effects of °Brix at harvest were not as considerable in 2005. This may have been due to slower ripening, overall, in 2005. On average, 2007 had the lowest wine scores among all treatments and therefore better wine quality relative to 2005 and 2006. Once again, this highlights that overall quality is largely influenced by seasonal effects.

The interaction between crop load and °Brix at harvest on expert wine score was significant each year. By and large, the combination of a higher crop load and higher °Brix at harvest resulted in a more optimal wine score. In fact, the best wine scores for 2005, 2006 and 2007, were in the UN/27.0, UN/28.5, and UN/27.0, respectively. The interaction is presented as a parallel linear regression in Figure 5.6 and demonstrates that wine score became lower (better) as °Brix at harvest increased in all crop loads even though trends in crop load were slightly different between years.

The expert panel wine score signifies the commercial placement of each treatment wine. Treatment wines that scored between 8.5 to 7.0 were suitable for the super premium price point brand produced by J. Lohr Vineyards and Wines. Treatment wines achieving scores below 7.0 are considered suitable for the ultra premium price points. Although none of the treatments achieved ultra premium scores, in all likelihood this was primarily a factor of the quality potential of the site rather than solely due to vineyard treatments and/or practices. There were many treatments which did not achieve scores at or below 8.5 and were thereby designated for wine products placed at a lower price point tier. The differences in expert quality scores conclude that although no treatment wines achieved *ultra premium* status, specific crop load x target °Brix treatments caused resulting wines to achieve *premium* quality status. Moreover,

these data illustrate that improving viticultural practices can improve wine quality, regardless of site. However, the potential quality for a given site (e.g. climate and soil) may be a limiting factor for achieving *ultra* premium wine quality.

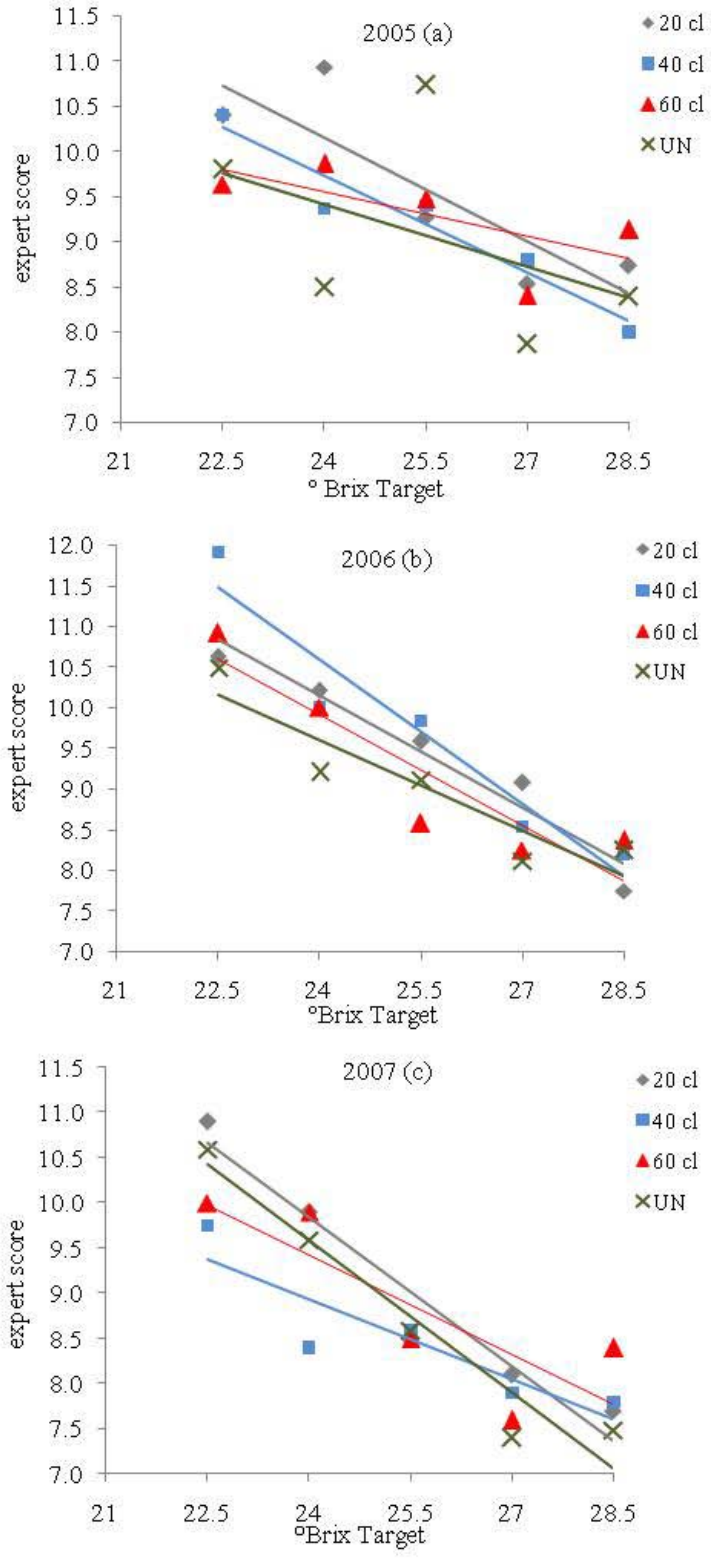


Figure 5.6: Parallel linear regression of the interaction between crop load and °Brix at harvest on the expert panel wine score in three years.

Treatment effects on expert panel attribute ratings:

Vegetative: Undoubtedly, crop reduction to the 20 cl per vine crop load increased the vegetative rating by the expert panel. Extended ripening to 27.0 or 28.5 °Brix had a very significant effect on the reduction of the vegetative attribute in treatment wines rated by the expert panel. Most likely, the additional time on the vine, facilitated by extended ripening, reduced the compounds attributing to the vegetative characteristics such as methoxypyrazines. Methoxypyrazines can have a significant impact on Cabernet Sauvignon as they are known to cause undesirable vegetative flavors in wine (Lacey *et al*. 1991, Noble *et al*. 1995, Wilkinson *et al*. 2006). In addition, increased shading within the canopy and fruiting zone is known to increase the chance of unripe herbaceous character development in the fruit (Haselgrove *et al*. 2000) and thus affects wine flavor.

These sensory results are substantiated by the methoxypyrazine data presented in Chapter 4 which illustrated that IBMP decreased as °Brix at harvest and DAV increased. Furthermore, the data showed higher initial IBMP levels in the lower crop loads and would explain why the 20 cl received a higher vegetative rating relative to the other crop loads. Chapman *et al.* (2004) also reported that concentrations of the compound 2-methoxy-3-isobutylmethoxypyrazine (MIBP) increased as bud number (hence crop load) decreased, and positively correlated with bell pepper intensity ratings.

In addition, the increased ethanol and subsequently higher body rating in wines which underwent extended ripening may have masked and/or lessened the intensity of compounds contributing to the vegetative attribute. Ethanol-induced palate warmth and perceived viscosity may indirectly affect aroma and flavor perception; however, these interactions have not been thoroughly

investigated (Delwiche 2004). The interaction which occurred in 2006 is displayed in Figure 5.7 and indicates that the 20 cl had the highest vegetative ratings at each target °Brix level. This was in contrast with the higher crop loads (i.e. 60 cl and UN) which generally had lower vegetative ratings at each °Brix target. The vegetative rating by the expert panel is further supported by the descriptive analysis and scaled attribute ratings which had similar trends. Moreover these results support those of Chapman *et al*. 2004 who reported decreased "veggie" attributes as crop load increased.

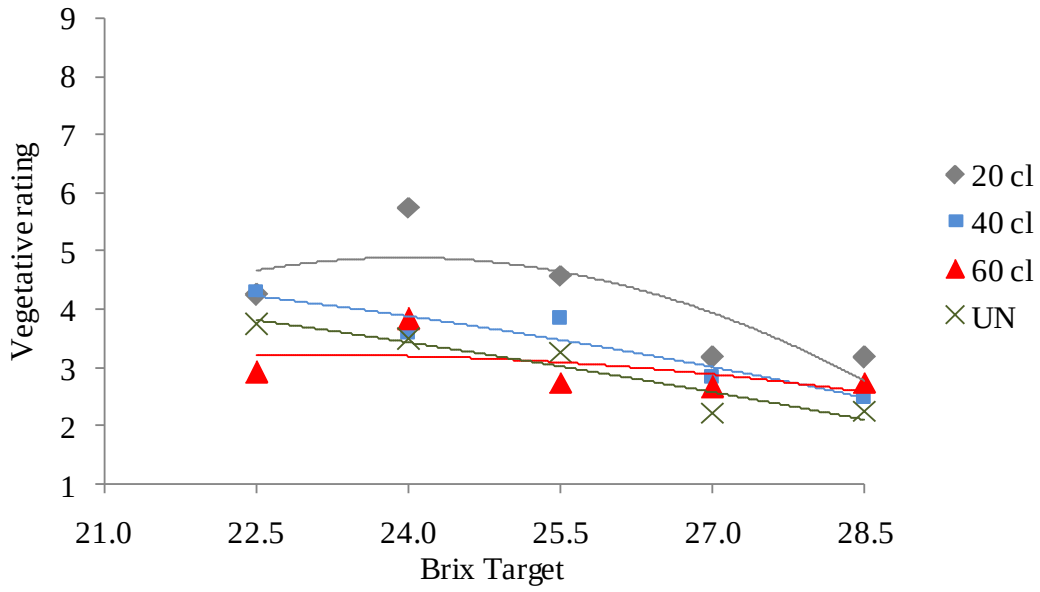


Figure 5.7: The interaction between crop load and °Brix at harvest on the vegetative rating of treatment wines by the expert panel in 2006.

Body: The body attribute was only significantly affected by crop load in 2007. In that year, the 40 cl was rated significantly greater relative to the 20 cl and 60 cl but was still statistically similar to the UN. There was not a consistent trend in the body rating due to crop load between the two years; however, body significantly increased as °Brix at harvest increased. It is likely that body ratings increased, in part, as a result of higher ethanol in wines which had higher °Brix

at harvest. The sensory perception of body or viscosity of a wine is known to be influenced by the amount of alcohol (ethanol) within the wine (Nurgel and Pickering 2005, Gawel *et al.* 2007). The interaction between crop load and °Brix showed different trends among years (Figure 5.8 a, b). The most inconsistent interaction was in the 40 cl which had contrasting patterns between 2006 and 2007 and the 60 cl at the 28.5 °Brix target in 2007. Unfortunately, the interactions do not provide any repeated trends, and may be more related to perception differences among the panelists in rating this attribute.

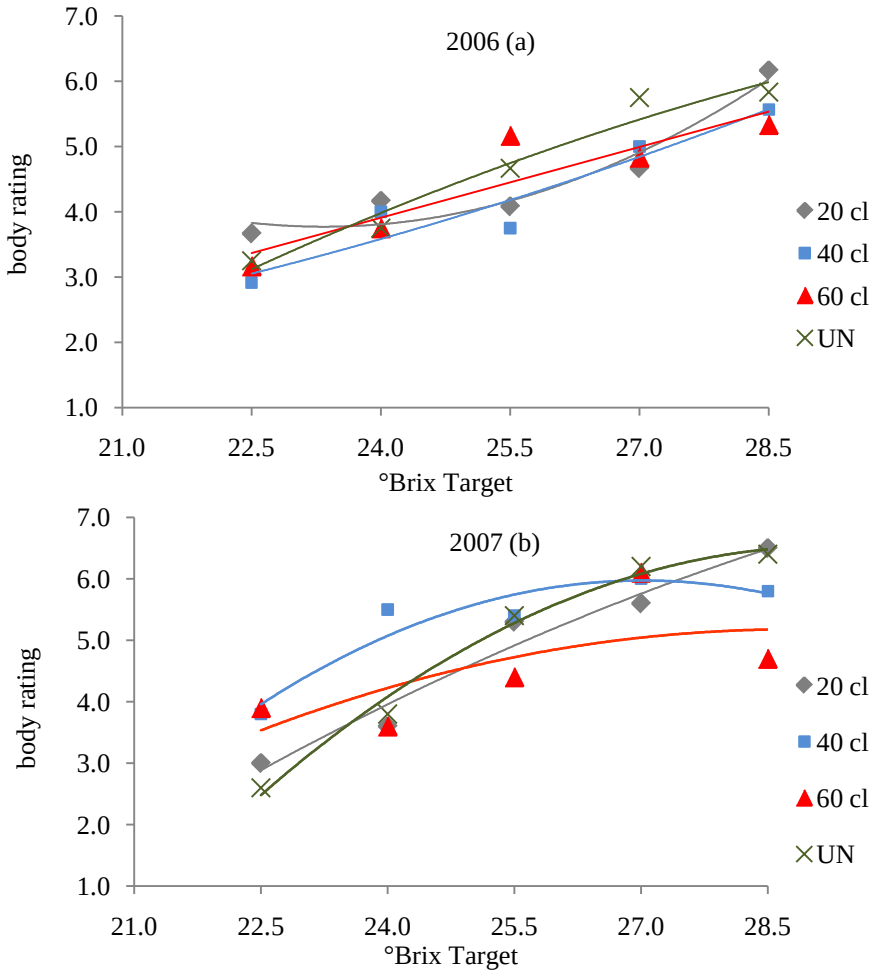


Figure 5.8: The interaction of crop load and °Brix at harvest on the attribute body rated by the expert panel.

Flavor intensity: Flavor intensity in the UN was significantly greater than all other crop loads in 2006. This pattern differed in 2007; although significant differences occurred overall, there were more similarities between crop loads—as indicated by mean separation. Therefore, the 2007 data do not provide strong conclusions for the effect of crop load on flavor intensity—as rated by the expert panels. However, these results demonstrate that crop reduction to the 20 cl did not increase flavor intensity relative to all other crop loads. Furthermore, higher °Brix at harvest increased flavor intensity ratings and thereby supports the practice of extended ripening for improvement of flavor intensity. The interactions in 2006 and 2007 are presented in Figure 5.9 a, b; however, there were no similar trends among both years. Once again, the interaction may have been due to judge variability in rating the attribute flavor intensity.

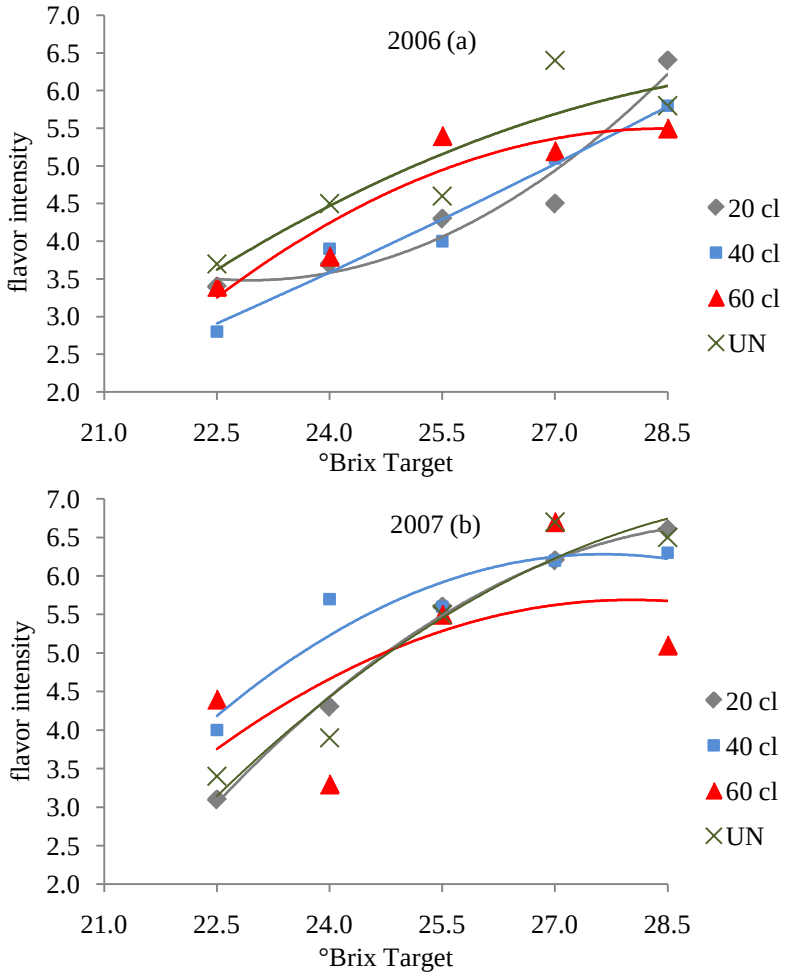


Figure 5.9: The interaction of crop load and °Brix at harvest on the attribute flavor intensity rated by an expert panel.

Quality 2006: The 2006 treatment interaction on the attribute 'quality' is presented in Figure 5.10. At the 22.5 °Brix target, the 20 cl, 60 cl and UN were all rated similarly. However, as °Brix at harvest increased, the UN rated higher in quality relative to the 20 cl and 40 cl until the final °Brix target. This interaction suggests that the UN was perceived to be of higher quality at the lower °Brix targets, compared with the lower crop loads at earlier °Brix targets. The UN/27.0 and 20/28.5 obtained the highest quality rating overall. Generally, better wine quality was perceived for the higher crop loads at the moderate °Brix targets (i.e. 24.0, 25.5, and 27.0).

Quality ratings in the lower crop loads only surpassed the 60 cl and UN with extended ripening to 28.5 °Brix.

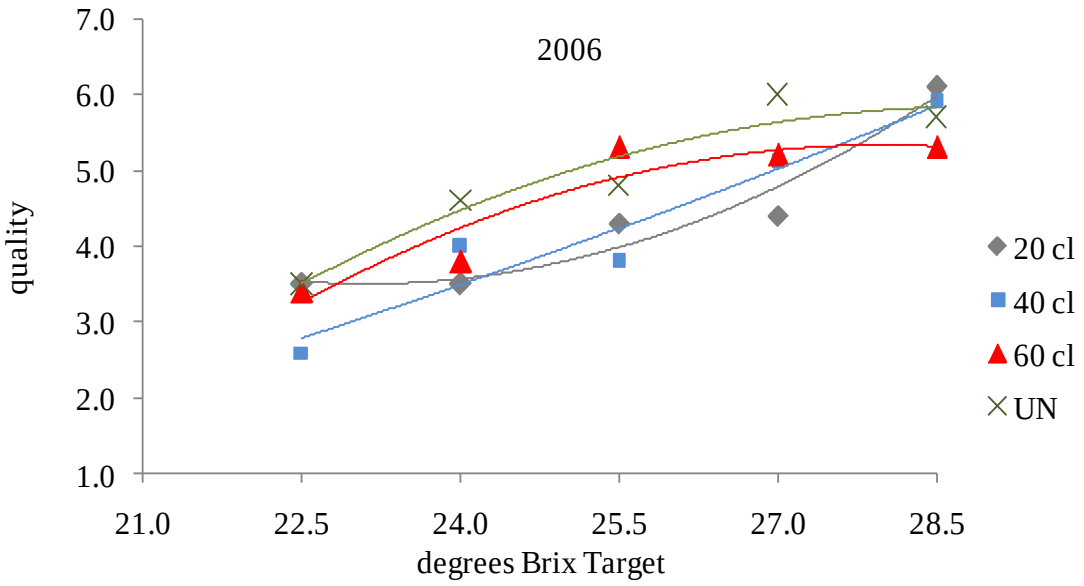


Figure 5.10: The interaction of crop load and °Brix at harvest on ratings for the attribute quality in 2006 treatment wines.

5.5.3 Descriptive Analysis

Analysis of variance indicated that judges were a significant source of variation in the data each year. This indicates that the judges may have used the scales differently although rated the wines similarly relative to the group. Panelists commonly use scales differently and/or stay toward the middle section of the scale (Meilgaard *et al.* 2007). The wine by judge (W x J) interactions suggest that judges did not always use the terms similarly relative to the entire group. Perhaps the significant interactions between W x J and judge by replication (J x R) can be explained by the high variability among the judges. Chapman *et al.* (2004) also reported that judges were a significant source of variation in the descriptive analysis testing on Cabernet Sauvignon wines

made from different crop yields. Furthermore, no individual attribute was significant due to the W x J interaction in *all* years. However, the attributes color density, ethanol, berry, jam and body were significant due to the W x J interaction in two of the three years. Previous studies using DA generated and rated similar descriptors of Cabernet Sauvignon. For example, Heymann and Noble (1987) used berry aroma, berry by mouth and vegetative by mouth, in addition to other similar descriptors not used in my study. Additionally, Chapman *et al.* 2004 used vegetative aroma, vegetative flavor, astringency, red/black berry, jam aroma and fruity flavor as key attributes for assessing yield differences on subsequent wine sensory.

Treatment effects on descriptive analysis: Degrees Brix at harvest was certainly a driving factor in how treatment wines associated with particular attributes. Treatment wines made from grapes harvested above 25.5 °Brix were strongly correlated to the more fruity aromas and flavors such as berry and jam: as opposed to the lower °Brix targets which correlated with less desirable flavors and aromas such as vegetative, sulfides, acid and/or astringency. The best color depth was achieved with higher °Brix at harvest and red color intensity decreased as °Brix at harvest increased. Wine color is known to influence its acceptance and consumer preference (Somers and Evans 1974, Boido *et al.* 2006). Increased color intensity as identified by sensory analysis has been linked with greater consumer acceptability (Del Pozo-Insfran *et al.* 2006). Extended ripening also significantly increased the mouthfeel components of treatment wines such as body, duration, and ethanol burn.

Crop load influenced the descriptive analysis, but to a lesser extent. The higher crop loads were more associated with the desired flavor profile beginning at the 25.5 °Brix target compared with the 20 cl which required extended ripening to a minimum of 27.0 °Brix in order to obtain some

desirable wine characteristics. Furthermore, higher crop loads had a more optimal vine balance, as indicated by the Y/P ratios and increased PAR—discussed in Chapter 3. Presumably, treatments with more optimal vine balance positively affected the flavor profile in subsequent wines. Generally, the combination of higher crop load with extended ripening to 27.0-28.5 °Brix produced the most desirable sensory outcome in treatment wines analyzed by descriptive analysis. These results concur with previous research by Chapman *et al.* (2004) who reported that increased crop load increased the intensity of fruity attributes and concomitantly decreased "veggie" attributes in Cabernet Sauvignon. This is also supported by Bravdo *et al.* (1985) who showed a positive correlation between crop load and wine quality.

The direct effects of crop load and °Brix at harvest on sulfide aroma was inconsistent, but generally wines at the 22.5 and 24.0 °Brix target rated higher in sulfides. There were significant negative correlations between sulfide aroma and, berry aroma, berry flavor, and jam aroma in 2006 and 2007. Furthermore, a significant positive correlation existed between sulfide and vegetative aroma in 2005 and with vegetative flavor in 2007. These data suggest that sulfides were more prevalent to the judges in lower °Brix wines. Sulfides could have been suppressed in the higher °Brix wines due to increased berry aroma and flavor; however, it is more likely that increased vegetative aromas confounded the sulfide ratings. For example, vegetative aroma was misperceived as sulfide aroma; therefore, wines with higher levels of vegetative aromas i.e. the lower °Brix targets, may have affected sulfide intensity ratings. Practical observation has indicated that panelists can easily confuse sulfide and vegetative aromas (S. Langstaff 2006 personal communication, S. Peck 2008 personal communication).

Seasonal effects on descriptive analysis: Although each season had variable rainfall, temperature fluctuations, and average yield, trends in the descriptive analysis were similar among all three years. The averages for attribute ratings were consistent from year to year—no one year had distinctively higher ratings for any attributes relative to all years.

The 2006 season had fewer differences among the vegetative flavor and aroma characteristics relative to 2005 and 2007. In fact, there were no significant differences found in treatment wines due to vegetative aroma or flavor attributes. In 2007, a greater majority of treatments at the 25.5 °Brix targets were correlated with berry aroma and flavors than in previous years—particularly treatments 60/25.5, 40/25.5, and UN/25.5. Prior to 2007, 60/25.5, and 40/25.5 were the only treatments moderately close to either berry aroma or flavor. The 2007 season had the lowest average yield among the three years and faster °Brix ripening. It is conceivable that this contributed to the 40 cl, 60 cl, and UN all correlating with the desirable attributes at moderate °Brix targets. Interestingly, 20/25.5 still remained near the astringent and vegetative attributes in 2007. These results suggest that wines harvested at moderate °Brix levels (i.e. 25.5) can achieve a desirable flavor profile if the crop load is in balance.

5.5.4 Scaled Attribute Ratings (Australian panel)

Results of the scaled attribute ratings showed that as °Brix at harvest increased, ratings for the attributes flavor intensity, body and quality increased. Generally, these data were consistent with results from the descriptive analysis and expert panel in 2005 and it was therefore deemed unnecessary to duplicate these ratings in 2006 and 2007. However, the use of a primarily Australian panel was an interesting comparison to the other sensory results which exclusively used American panelists.

A major dissimilarity occurred in the vegetative ratings relative to those of the expert panel and DA, in that results from the Australian panel had no significant differences due to crop load or °Brix at harvest. Panelists for the scaled attribute ratings were all Australian residents which may account for the lack of differences in the vegetative attribute. Possibly this was influenced by a greater familiarity with green flavor characteristics in Australian Cabernet Sauvignon as compared with California Cabernet Sauvignon. For example, Australian Cabernet Sauvignon is known to have a stylistic amount of green characters which have been described as 'mint' or 'eucalyptus' flavor. The chemical compound 'cineole' is known to play a significant role in eucalyptus character (Herve *et al.* 2003). Van Leeuwen *et al.* (2007) reported that 40 % of the studied wines contained above threshold levels of cineole; suggesting that eucalyptus may be a perceptually significant flavor in Australian wines. Subsequently, Saliba *et al.* (2009) reported that moderate intensity of eucalyptus character in red wine should not be considered a taint—and the consumer rejection threshold was at 27.5 ppb. This is considerably higher than the detection threshold previously reported at 3.2 ppb (Herve *et al.* 2003). In addition, anecdotal evidence has suggested that characters such as: 'herbaceous', 'vegetative', 'grassy', 'capsicum', and 'green' are distinctive in characterizing Cabernet Sauvignon from the Margaret River Region of Australia (Wilkinson *et al.* 2006); considered to be a "definitive champion region" for Cabernet Sauvignon production (Mann 2008). Some consumers may even consider moderate intensities of eucalyptus character preferable to no eucalyptus character (Saliba *et al.* 2009). Therefore, it is conceivable that consumers who were familiar with this typifying character in Australian Cabernet Sauvignon would have a greater familiarity with 'greener' flavors and a higher level of acceptability. According to Lichtenstein and Fischhoff (1977), product familiarity is based on a person's self report of how much he/she knows about the product. Additionally, Coupey *et al.*

(1998) stated that people generally have well-formed, articulated values; and can retrieve an appropriate response to preference-elicitation questions.

Differences among the other three attributes further suggest that extended ripening improved wine quality. Moreover, these ratings demonstrate that panelists associate higher wine quality with higher flavor intensity and body—as also demonstrated by the expert panel. Generally, these data concur with the other sensory results in this experiment, with the exception of the inconsistency that occurred in the scaled attribute ratings of the Australian panel relative to the expert panel for the attribute 'vegetative'. The work of Langstaff *et al.* (1991) suggested that 'body' is an abstract term due to its multidimensional nature (Gawel 1997). Additionally, Gawel *et al.* (2007) reported that higher ratings of the term body were commonly associated with higher ratings of flavor.

The interaction of crop load and °Brix at harvest on flavor intensity, body and quality show that the UN and 60 cl consistently had higher ratings at the first two target °Brix levels (i.e. 22.5 and 24.0) whereas the 20 cl and 40 cl were consistently lower (Figures 5.11 a, b, c). All crop loads increased in ratings as °Brix at harvest increased however the 20 cl and 40 cl rated consistently higher than the 60 cl and UN at the 28.5 °Brix target. In fact, the 40 cl had the highest ratings at the 28.5 °Brix target for all three attributes: flavor intensity, body and quality. Worth noting is that quality ratings on the UN were variable between target °Brix and exhibited a large reduction at the 25.5 °Brix target. Overall, panelists stayed within a narrow range (between 3.5-5.5) on the 1-9 scale. This central tendency is known to be common for panelists using the 9 point scale (Lawless and Heymann 1998, Resurreccion 1998, Yeh *et al.* 1998, Villanueva *et al.* 2005,

Meilgaard *et al.* 2007). Certainly, these results demonstrate that extended ripening significantly increased the quality and associated attributes ‘body’ and ‘flavor intensity’ in treatment wines.

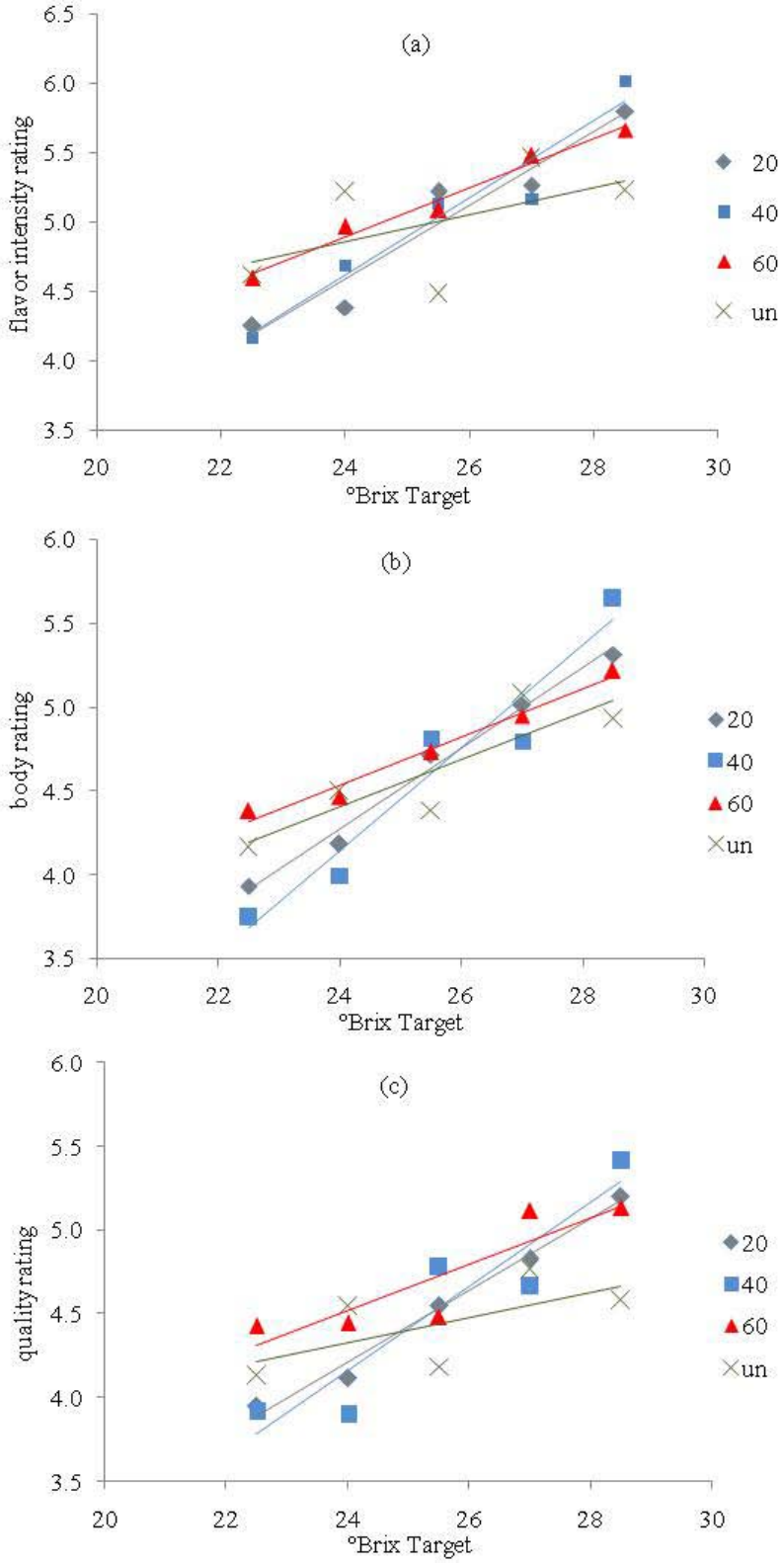


Figure 5.11: Parallel linear regression of the interaction of crop load and °Brix at harvest on three attributes in 2005 treatment wines rated by an Australian panel.

5.5.5 Consumer Testing

The consumer testing demonstrated that the only significant differences in likability were between treatments harvested at the 22.5 °Brix target and those at the higher °Brix targets (i.e. 24.0-28.5 °Brix). The key attributes driving this outcome according to the DA were vegetative aroma and flavor in all years and astringency in 2005 and 2006. Interestingly, there were no statistical differences due to crop load. This data conflicts with the expert panels, which clearly varied in quality ratings due to both crop load and °Brix. However, these consumer acceptability results highlight that crop reduction and extended ripening to levels near 27-28.5 °Brix did not significantly improve consumer likability. It must be noted that the consumer testing was only conducted on one vintage, and may be influenced by the demographics of the panelists. The consumers were primarily Australian and were all Australian residents. As a result, consumers were accustomed to different stylistic Cabernet Sauvignon wines within the Australian markets as compared with the styles of California Cabernet Sauvignon. Research supports that many Australian wines are known for a characteristic eucalyptus and/or herbaceous flavor (*Wilkinson et al.* 2006, Van Leeuwen *et al.* 2007) which is generally not a common attribute of the American Style. Unfortunately, no scientific studies exist comparing the preferences of Australian consumers to American consumers for Cabernet Sauvignon; therefore, this remains a theory until further investigation.

Consumer preferences of Australian 'main styles' Shiraz and Cabernet Sauvignon were tested by Lattey *et al.* (2007). The study concluded that consumers have different likes and dislikes, particularly for sensory properties related to green flavors and astringency.

The dissimilar results between likability ratings and expert panel scoring in my experiment, suggest that consumer preference was strongly influenced by demographics i.e. Australian based

consumers versus American based experts. These outcomes should be considered for commercial export of wines. Furthermore, Frøst and Noble (2002) reported that individual preferences have a stronger influence on likability than wine knowledge or sensory expertise. Consumer sensory testing has potential to improve industry knowledge of what constitutes wine quality. Therefore, including consumer testing in sensory analysis of viticultural and enological experiments is recommended for future research.

5.6 Conclusions

a) Difference testing indicated that more differences were detected among grapes harvested and fermented at lower °Brix levels such as 22.5 and 24.0 °Brix. In most cases, the three fermentation replications were similar enough to be blended for descriptive analysis testing. The triangle test proved a reliable method for difference testing. It is speculated that the increased differences detected in lower °Brix levels were due to a greater variability in ripeness among berries within a cluster.

b) Wines made from higher crop load and higher °Brix at harvest were rated as the most optimal quality by the expert panel. Treatments from low °Brix and low crop load had the least optimal quality rating. The unthinned crop load averaged best for wine quality among the three years as did wines harvested at 27.0 and 28.5 °Brix. The 2007 season had the most optimal wine scores relative to the three years, indicating that season remains influential on overall wine quality.

c) Results from the expert ratings in both 2006 and 2007 indicated a strong positive relationship between optimum quality score and the attributes 'body' and 'flavor intensity'.

In contrast, there was a strong negative relationship between optimum quality score and the attribute 'vegetative'.

d) Descriptive analysis had similar trends among all years and proved a reliable and useful tool for analyzing the sensory effects of viticultural treatments. The principal component analysis illustrated a major divide in the flavor profiles of wines from grapes harvested at lower °Brix (i.e. 22.5-24.0) and those harvested at higher °Brix (i.e.27.0-28.5). Wines made from lower °Brix correlated with a less desirable flavor profile including the attributes: vegetative, astringent, sulfides, red color and acid. The higher °Brix wines correlated with a more desirable profile including the following attributes: berry, body, duration and color depth. Vines with a greater crop load achieved or approached a more desirable flavor profile at lower °Brix targets. Vine balance and rate of ripening most likely influenced this interaction.

e) Attribute ratings by the Australian panel resulted in no significant differences due to crop load on the attributes flavor intensity, vegetative, body or quality. However, °Brix at harvest significantly affected the ratings of flavor intensity, body and quality, in which attribute ratings increased as °Brix at harvest increased. Dissimilarities between these results and those of the expert panel are speculated to be a result of an Australian panel as compared with an American panel and the stylistic differences in Cabernet Sauvignon between the two markets.

f) Consumer testing indicated significant differences due to °Brix at harvest; however, there were no differences due to crop load. The lack of differences in these results, considering the large differences in the other sensory tests, was likely influenced by the majority of

consumers being of Australian origin and residency. Hence, a greater acceptability of green characters due to familiarity with the flavor profile of Australian Cabernet Sauvignon relative to California Cabernet Sauvignon. However, this remains a theory based on speculation due to limited scientific studies. Further investigation of viticultural effects on the sensory attributes of primary importance to consumers is recommended.

Chapter 6: EFFECTS OF CROP LOAD AND LATE SEASON IRRIGATION ON EXTENDED RIPENING

6.1 Introduction and Experimental Aims

A number of experiments have indicated that irrigation of grapevines has a significant effect on grape yield and on certain compositional factors such as: °Brix, pH and TA and thereby affects wine quality (Bravdo *et al.* 1984, Bravdo *et al.* 1985, Hepner and Bravdo 1985, Matthews and Anderson 1989, Esteban *et al.* 1999, Kennedy *et al.* 2002, Roby *et al.* 2004).

As the practice of extended ripening becomes more common, grape growers, viticulturists and winemakers are searching for practical ways to mitigate the negative effects of extended ripening—particularly berry weight loss, sluggish fermentations and high alcohol wines.

Premature arrest of fermentation constitutes one of the most challenging problems in wine production (Bisson 1999). Juice soluble solids (e.g. °Brix) at harvest will render the final alcohol in subsequent wines; however, the addition of water to must, prior to yeast inoculation, may decrease high soluble solids to conventional levels if effectively calculated and employed. The addition of water prior to fermentation creates potential negative perception issues with wine consumers, whereas blending or de-alcoholization of wines adds production costs and can introduce wine quality risks (Grant 2006).

Berry weight loss due to extended ripening is significant and is primarily caused by berry dehydration (Rogiers *et al.* 2000, Bisson 2001, Battany 2005 and Grant 2005). The losses in berry weight correlate with financial losses for growers paid on dollars per tonne. There is speculation that yield loss can be mitigated through soil moisture management (Dokoozlian 2006, Grant 2006). However, questions about irrigation amounts, timing and effects on subsequent wine quality still remain.

There is a widespread perception in the wine industry that irrigation close to harvest has a dilution effect on fruit and is thereby detrimental to wine quality, although little scientific evidence exists which substantiates this perception (Keller *et al.* 2006). It is widely known that following veraison (Stage III) the phloem becomes the major route of transport into the berry (Hrazdina *et al.* 1984, Mullins *et al.* 1992, Greenspan 1994); therefore phloem sap becomes the primary water source for the berry after veraison (Keller *et al.* 2006). However, the work of Bondada *et al.* (2005) demonstrated the xylem to be functional in the later stages of ripening when an appropriate gradient was artificially imposed on berries—introducing the possibility that back- flow from the berry to vine could account for berry weight loss (Tillbrook and Tyerman 2008). Additionally, Sanchez *et al.* (2006) and Mendez-Costabel (2007) studied the effects of late season deficit irrigation on Merlot grapes in the San Joaquin Valley of California and Sonoma County—indicating significant affects on yield components and vine physiology measurements.

However, there are still many unanswered questions regarding water relations between the berry and vine during the late stages of ripening. The cause and level of xylem flow cessation at ripening and its potential functionality is still unclear (Creasy *et al.* 1993, Keller *et al.* 2006) and remains an important research topic (Tillbrook and Tyerman 2008). Practical management practices are necessary in order for growers and wineries to achieve economic sustainability when using the practice of extended ripening. The notion of maintaining peak berry weight by increasing applied irrigation in the vineyard and concurrently decreasing soluble solids to acceptable levels would potentially alleviate the major problems associated with extended ripening. Furthermore, this practice could improve relations between growers and winemakers near harvest rather than increasing tension.

The aims of this experiment were to:

1. Monitor changes in berry composition due to different crop loads and irrigation levels.

2. Investigate whether crop load, late season irrigation, or the interaction of the two affect vegetative growth and yield components.

3. Test whether fruit and wine composition are affected by crop load, late season irrigation or both; including significant changes in sensory analysis and wine quality parameters of resultant wines.

Hypotheses tested were that (1) increased irrigation in late season increases yield and (2) increased irrigation in late season reduces berry total soluble solids and wine alcohol (%).

6.2 Materials and Methods

The effect of crop load and irrigation during the later stages of fruit ripening was examined in field-grown Cabernet Sauvignon grapevines—clone 8 on 1103 Paulsen rootstock. The experiment comprised 2.47 ha (6.1 acres) on ≤ 2 % slope.

6.2.1 Experimental Design

The experimental design was a 2 x 2 factorial with two irrigation levels and two crop loads. The crop loads were a thinned treatment (TH) and an unthinned (UN). Vines were thinned at E-L 31, (generally near 30 days post bloom) by removing approximately 15 clusters per vine in order to leave approximately 45 clusters per vine.

Vines were drip irrigated at the commercial standard deficit irrigation amount (0.70 ETc) until the fruit reached approximately 20 °Brix. The irrigation treatments were imposed at 22 °Brix.

Both crop loads were irrigated at two different irrigation levels, a standard regulated deficit irrigation practice (SD) and double the standard practice (DI). In addition, the DI included a 24 hour irrigation applied 48 hours prior to harvest—consistent with the commercial post harvest irrigation practice. The actual irrigation hours are presented in Appendix 11; generally the SD was irrigated at 40 % of ETc and the DI was at 80 % ETc for the period between 22 °Brix to harvest. The crop load x irrigation treatments were replicated four times in the field. Replications were comprised of three consecutive rows—data was only taken from the middle row of each replication. Irrigation treatments were imposed at 22 °Brix. Treatments were harvested when the SD irrigation in each crop load reached 27.0 °Brix.

Designated panel sites were set up equidistant from each other within the row and consisted of three panels of three vines each per treatment replication. These sites were used for data collection including: leaf water potential, yield components and growth data.

6.2.2 Leaf Water Potential

Leaf water potential (LWP) was measured using a pressure chamber manufactured by Soil Moisture Instruments, Corp. model 3000. Leaf water potential was measured beginning at solar noon. All LWP readings were taken at the designated panel sites and in randomized order each week. A fully exposed leaf approximately 4-5 leaves from the shoot tip was cut at the petiole, immediately put into a plastic bag and transferred into the pressure chamber. The pressure chamber was then sealed and nitrogen gas allowed to enter through the gas exchange valve until first signs of water appeared on the cut tip of the petiole. Pressure (bars) was then recorded. Three leaves per treatment replication were measured and averaged. LWP was measured on the same day each week.

6.2.3 Yield Components

Berry weight development: Berry weight was monitored on a weekly basis beginning at 22 °Brix for all treatment replications. Berries were collected from each treatment replication in zip lock bags (see berry sampling protocol, Appendix 1). One hundred berries were randomly selected from each collection sample and weighed to determine the average per berry weight. A Fisher Scientific accu 2202 scale was used for berry weight measurements.

Berry weight at harvest: Berry weight was measured at harvest by collecting berries on each treatment one hour prior to the harvesting of fruit for fermentation. The same berry sampling protocol was followed as was used for berry development. One hundred berries were randomly selected from each collection sample and weighed to determine average berry weight. A Fisher Scientific accu 2202 scale was used for berry weight measurements.

Berries/cluster: The number of berries/cluster was calculated from average harvest berry weight and cluster weight for each treatment i.e. berries/cluster = average cluster wt (g)/average berry wt (g).

Clusters/vine: Clusters were counted as fruit was harvested from each panel vine and ***mean cluster weight*** (g) calculated as [fruit per vine (kg)/clusters per vine] x 1000 (g)/(kg).

Yield/vine: Yield per vine (kg) was measured at harvest on the designated panel vines. All clusters per vine were cut and weighed in the field then put in the macro bin with the entire row for fermentation.

Tonnes/hectare: Yield, in terms of tonnes (t) per hectare (ha), was calculated from the yield/vine measurements in the panel vines using the following equation:

Tonnes/ha = (kg/vine x 1922 vines/ha) ÷ 1000 kg/tonne

6.2.4 Vine Growth and Canopy Density

Ceptometer: Leaf area index (LAI) and Photosynthetically Active Radiation (PAR) were measured on the vine canopy using an AccuPar L-80 Ceptometer by Decagon. Measurements were taken at veraison and harvest, and followed the same protocol for each data set (Appendix 2).

Mean cane weight: The total ***shoots per vine*** were counted before pruning and mean cane weight was calculated as pruning weight.vine^{-1}/total canes.vine^{-1}.

Pruning weights: Pruning was done in early January of each subsequent season and pruning weights were measured to investigate changes in vegetative growth and to calculate the yield/pruning ratio (Y/P). The designated panel vines in each treatment were used. Pruned canes from each vine were gathered, tied together and weighed with a Berkley hanging scale.

6.2.5 Fruit and Wine Composition

Berry development: The °Brix, pH and TA development was monitored weekly—beginning at the 22 °Brix target—using the same berry sampling protocol described above in 'berry weight development'. Degrees Brix was measured using the Anton Paar 35n density meter with significant digits to the tenth place. Titratable acidity was measured by a standard hand titration using 50 mL of deionized water, and indicator dye and Sodium Hydroxide at 1 N solution (Appendix 5). Juice pH was measured using an Orion 720A pH meter.

Harvest juice chemistry: Juice samples for harvest juice chemistry were taken just after crushing. Degrees Brix was measured using an Anton Paar DMA 35n density meter. Juice pH and TA were measured on a Tim 865 titration manager by Radiometer analytical autotitrator.

Yeast assimilable nitrogen: The yeast assimilable nitrogen (YAN) was measured using a Randox Enzymatic kit. The protocol for preparation of NOPA and Ammonium is described in Appendix 8.

Ethanol: Wine ethanol (ETOH) was analyzed on the FOSS Wine Scan. The preparation protocol is described in Appendix 9.

6.2.6 Wine Color and Phenolics

Wine color and phenolics were analyzed using the Shimadzu UV-1700 pharma spec spectrophotometer. Wine samples were analyzed at Absorbance (280 nm) for ***total phenols*** and at Absorbance (420 nm) and Absorbance (520 nm) on press and post malolactic fermentation wines. ***Color density*** was obtained by combining the values from the Absorbance (420 nm) and Absorbance (520 nm) measurements. i.e. Absorbance (420 nm) + Absorbance (520 nm) = Color Density (CD) Sample preparation procedure is described in Appendix 7. Wine ***Hue*** is expressed as the ratio between Absorbance (420 nm) and Absorbance (520 nm) A(420nm)/A(520nm).

6.2.7 Wine Sensory

Expert panel: Treatment wines were scored by the 10 expert panelists in January of each year following the previous vintage. The panelists consisted of J. Lohr Vineyards and Wines management who annually score and categorize commercial wines to determine their market placement including: price point and brand within the J. Lohr portfolio. Expert panelists

included: Vice President (VP) of Winemaking, Red Winemaker, Enologist, Associate Enologist, VP Sales and Marketing, President/Proprietor, Vineyard Managers (2), Viticulturists (3), and other associated management (2).

Treatment wines were taken directly from barrels two days prior to the sensory testing. Wine bottles followed the standard practice of J. Lohr Winery wherein, they were treated with 25 ppm of Sulfur Dioxide ($SO_{2)}$ and 1 ppm of Copper Sulfate ($CuSO_4$) as a standard practice to suppress any sulfides which can be present in young wines. Wines were scored based on the grading system presented in Table 6.1. Panelists had a minimum of 3 previous years experience with the scoring system and therefore were familiar to the scoring system. Treatment wines were presented in four flights of four wines, unidentified and in randomized order for each panelist. All wines were evenly poured to 44.4 mL per glass. Panelists were instructed to taste wines at a comfortable pace, break when needed, and score wines based on the letter grade (e.g. A-1) which was later converted to the equivalent number score.

Table 6.1: Commercial wine scoring system used for expert panel to grade treatment wines into market value price point. Note: Wine quality and price point increase as score decreases.

Letter Grade		Score Value	Price Point
A	1	**1**	>$50.00 USD
	2	**2**	
	3	**3**	
B	1	**4**	$50.00 - 30.00 USD
	2	**5**	
	3	**6**	
C	1	**7**	$20.00-16.00 USD
	2	**8**	
	3	**9**	
D	1	**10**	$10.00-8.00 USD
	2	**11**	
	3	**12**	
E	1	**13**	$5.00-3.00 USD (no J.Lohr program)
	2	**14**	
	3	**15**	

Flavor intensity rating: Subjective opinion on increased irrigation during late ripening has been thought to 'dilute' the flavors of resultant wines (Keller *et al.* 2006). To address this, flavor intensity ratings were done by the expert panel concurrent with their scoring of treatment wines to consider whether the expert score was weighted by the amount of flavor intensity within the treatment wines. Wines were scored using the 1-9 hedonic scale.

Difference testing: The triangle test was conducted for the 2007 vintage to test whether any significant differences could be detected in wines due to treatment effects. Procedures from Lawless and Heymann (1998) were followed. Each of the two crop loads were tested against the different irrigation regimes and for all field replications. For example, the UN/SD and UN/DI were difference tested for each of its four field replications. Each session included a total of six panelists who tasted four flights each. Panelists were wine industry professionals and were

consistent among all sessions. Sessions were held daily for one week. Wines were presented in randomized order for the six panelists and for each direction of the triangle test. Panelists took a forced break of one minute between directional tastings. Wines were poured to 44.4mL per glass. Panelists were told that two wines in each flight were the same; identify the wine which was different.

6.2.8 Statistical Analysis

All statistical analysis was conducted using GenStat 10th edition. All interactions were analyzed by a general two way analysis of variance ANOVA. The treatment structure was crop load x irrigation and the blocking structure by replication. Mean separation was done by LSD and Duncan's multiple range test for means at $p<0.05$.

6.3 Results

6.3.1 Leaf Water Potential

The irrigation treatments caused substantial differences in vine water status as indicated by the midday LWP—crop load did not have a large effect on LWP (Figures 6.1 (a, b), 6.2).

In 2007, LWP became progressively less negative with both irrigation treatments over time although the DI for both crop loads had considerably higher LWP, i.e. less negative, than the SD on all measurement dates. Although fewer LWP measurements were taken in 2006 the data clearly demonstrate that the DI affected vine water status, resulting in a higher LWP. There was, however, one exception on 26-Oct-2006 when the TH/SD and TH/DI were quite similar. The 9-Nov-2006 measurement (Figure 6.1 b), when the UN/SD and UN/DI were similar, was taken just after harvest and reveals the change in water status upon the removal of crop.

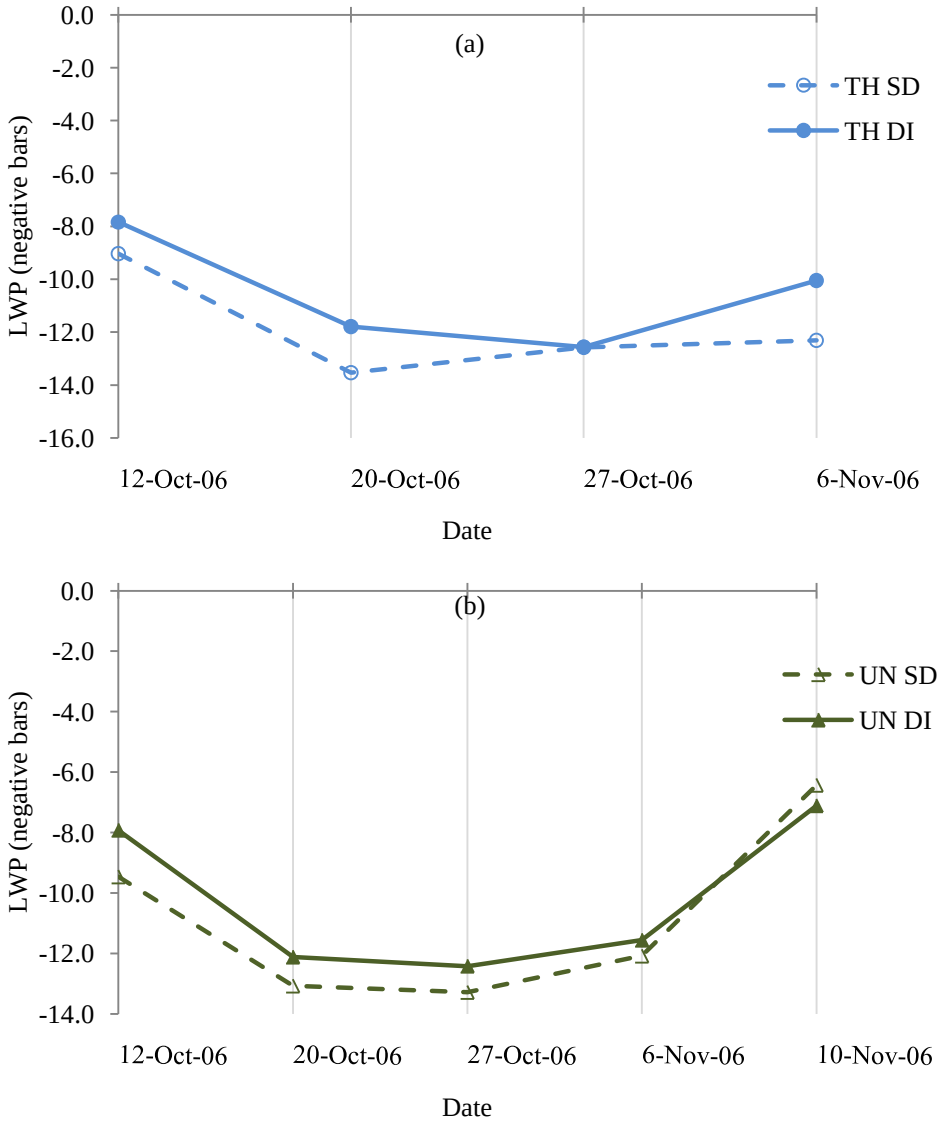


Figure 6.1: The effects of increased irrigation late season on leaf water potential (LWP) for two crop loads in 2006. Figure 6.1(a) represents the thinned crop load (TH); Figure 6.1(b) represents the unthinned (UN) crop load.

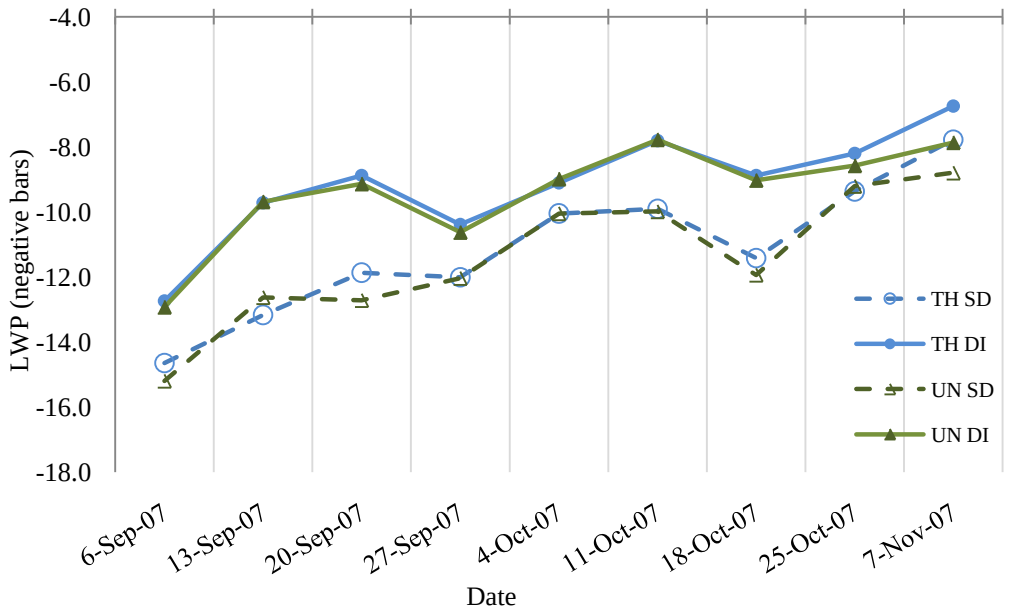


Figure 6.2: The effects of increased irrigation late season on leaf water potential (LWP) for two crop loads in 2007.

6.3.2 Yield Components

Berry weight development: Berry weight development was monitored on a weekly basis from approximately 1-2 weeks after the onset of irrigation treatments in 2006 and 2007, (Figures 6.3 and 6.4, respectively). Berry weight was consistently greater for the DI relative to the SD for both crop loads and in each year. The UN/DI was significantly higher at the final measurement in both 2006 and 2007 (Figures 6.3 b and 6.4 b) and was higher relative to the UN/SD by 9 % and 11 % in 2006 and 2007, respectively. The most notable effect of the DI occurred for the UN in 2007, particularly for the final three weeks of berry weight measurements. Beginning just after 11-Oct-07, the UN/DI and UN/SD were divergent, demonstrating that the SD decreased in berry weight, while the DI maintained a large proportion of its initial berry weight. Interestingly,

Figures 6.3 (a) and 6.4 (a) illustrate that the TH/DI did not maintain its berry weight as effectively as the UN/DI.

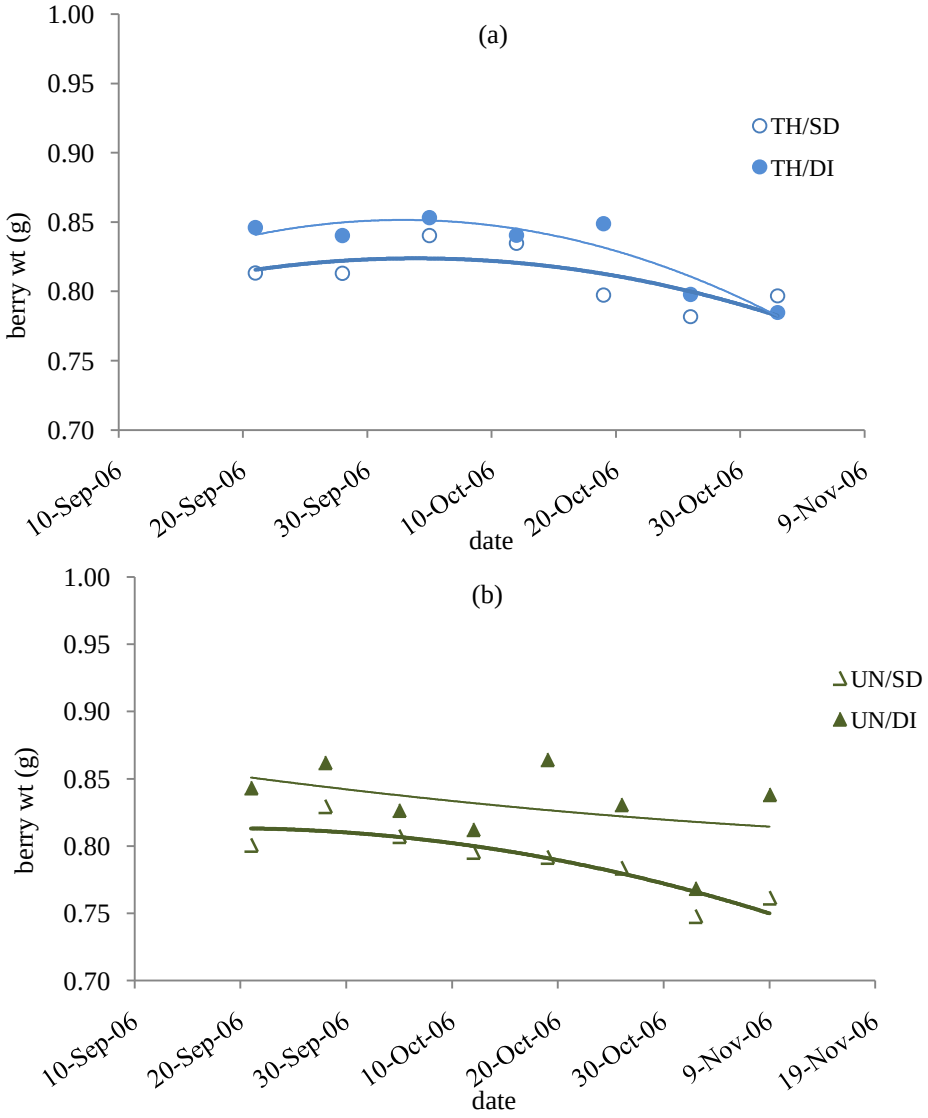


Figure 6.3: The effects of crop load and increased irrigation late season on berry weight development during ripening in 2006. Figure 6.3 (a) represents the thinned (TH) crop load and Figure 6.3 (b) represents the unthinned (UN). The bold line represents the DI.

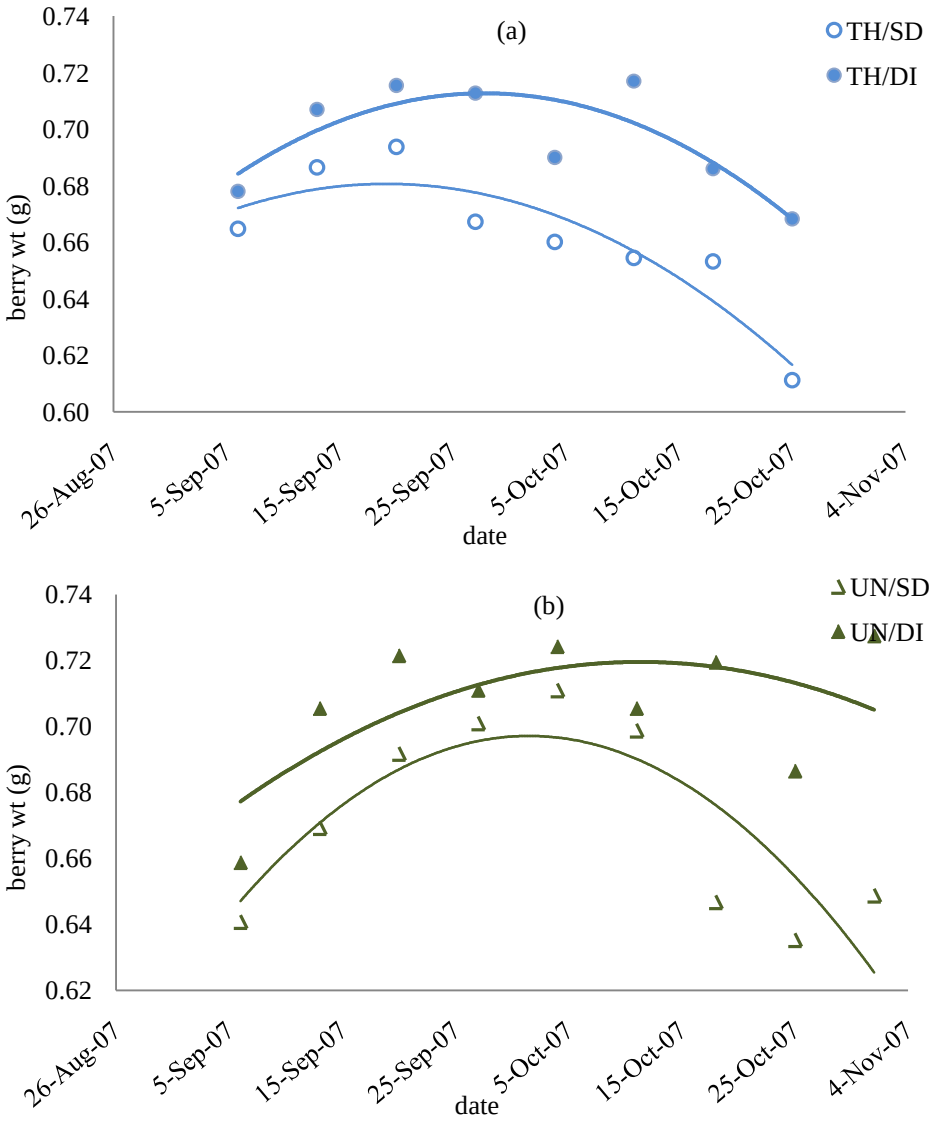


Figure 6.4: The effects of crop load and increased irrigation late season on berry weight development during ripening in 2007. Figure 6.4 (a) represents the thinned crop load and Figure 6.4 (b) represents the unthinned. The bold line represents the DI.

Berry weight at harvest: The effects of irrigation and crop load on berry weight at harvest are presented in Table 6.2. Berry weight was only significantly different in 2007. The unthinned (UN) had a significantly greater berry weight on average relative to the thinned (TH) in 2007.

The DI increased berry weight in both years, but was significantly greater in 2007 ($p<0.001$). There were no significant interactions.

Table 6.2: Effect of late season irrigation and crop load on berry weight at harvest.

	berry weight at harvest (g)		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned	0.79	0.64 a	0.72
Unthinned	0.80	0.66 b	0.73
	ns	*	
Irrigation			
Standard (SD)	0.78	0.58 a	0.68
Double Irrigation (DI)	0.81	0.71 b	0.76
	ns	***	
Interaction	ns	ns	

Means within columns separated with different letters differ significantly at $p<0.001$ or 0.05 by LSD. *, **, ***, ns, indicate significance at $p<0.05$, 0.01, 0.001, or not significant respectively.

Berries/cluster: There were no significant differences in berries per cluster due to crop load, irrigation or the interaction of the two (Table 6.3).

Table 6.3: Effect of increased irrigation late season on berries per cluster in two crop loads.

	berries/cluster		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned (TH)	166	164	165
Unthinned (UN)	157	170	164
	ns	ns	
Irrigation			
Standard (SD)	163	161	162
Double Irrigation (DI)	160	173	167
	ns	ns	
Interaction	ns	ns	

*, **, ***, ns, indicate significance at $p<0.05$, 0.01, 0.001, or not significant respectively.

Clusters/vine: The effects of irrigation and crop load on clusters per vine (cl/vine) are presented in Table 6.4. The UN had significantly more clusters/vine in both 2006 and 2007 relative to the TH ($p<0.001$). Clusters per vine were unaffected by irrigation each year. A significant interaction occurred between crop load and irrigation in 2007.

Clusters per vine were noticeably greater, in 2007 relative to 2006. The overall average cl/vine was 41 and 61 for 2006 and 2007, respectively. On average, clusters per vine were reduced by approximately 20 % in the TH.

Table 6.4: Effect of increased irrigation late season on clusters per vine in two crop loads.

	clusters/vine		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned (TH)	37 a	55 a	46
Unthinned (UN)	46 b	69 b	58
	***	***	
Irrigation			
Standard (SD)	40	62	51
Double Irrigation (DI)	43	61	52
	ns	ns	
Interaction	ns	*	

Means within columns separated with different letters differ significantly at $p<0.001$ *, **, ***, ns, indicate significance at $p<0.05$, 0.01, 0.001, or not significant respectively.

Mean cluster weight: Mean cluster weight was measured in both years (Table 6.5) although significant differences only occurred in 2007, when the TH had a significantly lower mean cluster weight relative to the UN. The DI treatment resulted in a significantly greater mean cluster weight than the SD. There were no significant interactions.

Table 6.5: Effect of increased irrigation late season on mean cluster weight in two crop loads.

	mean cluster weight (g)		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned (TH)	130.7	103.5 a	117.1
Unthinned (UN)	125.1	111.0 b	118.1
	ns	*	
Irrigation			
Standard (SD)	126.4	100.7 a	113.6
Double Irrigation (DI)	129.4	113.8 b	121.6
	ns	**	
Interaction	ns	ns	

Means within columns separated with different letters differ significantly at $p<0.01$ or 0.05 *, **, ***, ns, indicate significance at $p<0.05$, 0.01, 0.001, or not significant respectively.

Yield/vine: Crop load and irrigation both affected fruit yield per vine (kg) (Table 6.6). Fruit yield per vine in the UN was significantly greater than the TH in both 2006 and 2007. On average the UN had 1.4 kg of fruit per vine more than the TH. Fruit per vine increased for the DI relative to the SD in both years with the most significant increase in 2007. Overall, the TH decreased in yield/vine by approximately 15 % relative to the UN. Yield per vine was higher on average in 2007 due to increased clusters/vine.

Table 6.6: Effect of increased irrigation late season on fruit per vine (kg) in two crop loads.

	yield/vine (kg)		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned (TH)	4.6 a	5.4 a	5.0
Unthinned (UN)	5.5 b	7.7 b	6.6
	**	***	
Irrigation			
Standard (SD)	4.9 a	6.2 a	5.6
Double Irrigation (DI)	5.3 b	6.9 b	6.1
	*	***	
Interaction	ns	ns	

Means within columns separated with different letters differ significantly at p<0.001, 0.01 or 0.05 *, **, ***, ns, indicate significance at p<0.05, 0.01, 0.001, or not significant respectively.

Tonnes/ha: The effect of crop load and irrigation on tonnes per hectare (t/ha) is presented in Table 6.7. The t/ha due to cluster thinning were significantly different each year. Moreover, the UN consistently had significantly more than the TH. On average, there was a 22 % reduction (i.e. 2.8 t/ha decrease) due to cluster thinning in the TH. The DI increased t/ha but was only significant in 2007. A significant interaction occurred in 2007.

Table 6.7: Effect of increased irrigation late season on tonnes per hectare (ha) in two crop loads.

	tonnes/ha		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned (TH)	9.3 a	10.9 a	10.1
Unthinned (TH)	11.1 b	14.7 b	12.9
	**	***	
Irrigation			
Standard (SD)	9.8	12.1 a	11.4
Double Irrigation (DI)	10.6	13.5 b	11.7
	ns	**	
Interaction	ns	*	

Means within columns separated with different letters differ significantly at p<0.001 or 0.01 *, **, ***, ns, indicate significance at p<0.05, 0.01, 0.001, or not significant respectively.

6.3.3 Vine Growth

PAR and LAI: Photosynthetically active radiation (PAR) and leaf area index (LAI) within the fruiting zone was measured using the ceptometer in 2007 (Table 6.8). Neither crop load nor irrigation caused significant differences. The LAI decreased from veraison to harvest as did the percent (%) of above canopy PAR in the fruiting zone. There were no significant interactions.

Table 6.8: Effect of crop load and increased irrigation late season on photosynthetically active radiation (PAR) and leaf area index (LAI) on the canopy light environment measured using the ceptometer at veraison and harvest in 2007. PAR is expressed as the percent (%) of above canopy PAR within the fruiting zone.

	PAR and LAI			
Treatment	**LAI *ver***	**LAI *har***	**PAR ver (%)**	**PAR har (%)**
Crop load				
Thinned (TH)	3.7	2.0	34.0	41.0
Unthinned (UN)	3.8	1.9	30.0	39.0
	ns	ns	ns	ns
Irrigation				
Standard (SD)	3.6	1.9	32.0	39.0
Double Irrigation (DI)	3.9	2.0	34.0	40.0
	ns	ns	ns	ns
Interaction	ns	ns	ns	ns

*, **, ***, ns, indicate significance at $p<0.05$, 0.01, 0.001, or not significant respectively.

Shoots/vine: Shoots per vine were not significantly different due to crop load, irrigation or the interaction of the two in either year (Table 6.9). There were on average, five more shoots per vine in 2007 as compared with 2006.

Table 6.9: Effect of increased irrigation late season on shoots per vine in two crop loads.

	shoots per vine		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned (TH)	34	39	37
Unthinned (UN)	34	39	36
	ns	ns	
Irrigation			
Standard (SD)	34	39	37
Double Irrigation (DI)	34	39	37
	ns	ns	
Interaction	ns	ns	

*, **, ***, ns, indicate significance at $p<0.05$, 0.01, 0.001, or not significant respectively.

Mean cane weight: The effects of crop load and irrigation on mean cane weight are presented in Table 6.10. Differences in mean cane weight due to crop load were not significant in 2006 or 2007. Irrigation significantly affected mean cane weight in 2007, whereby the DI was significantly greater than the SD irrigation treatment. There were no significant interactions in either year. Mean cane weight differed considerably between the two seasons. The average mean cane weight in 2006 was 58 g compared with 31 g in 2007.

Table 6.10: Effect of increased irrigation late season on mean cane weight (g) in two crop loads.

	mean cane weight (g)		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned (TH)	58.1	30.8	44.5
Unthinned (UN)	57.4	30.4	43.9
	ns	ns	
Irrigation			
Standard (SD)	57.1	27.5 a	57.1
Double Irrigation (DI)	58.4	33.6 b	58.4
	ns	**	
Interaction	ns	ns	

Means within columns separated with different letters differ significantly at $p<0.01$
*, **, ***, ns, indicate significance at $p<0.05$, 0.01, 0.001, or not significant respectively.

Pruning weights: The effects of crop load and irrigation on pruning weight per vine are presented in Table 6.11. Crop load did not significantly affect pruning weight per vine in either year. The DI increased pruning weight in both years, but was significantly greater than the SD only in 2007 ($p<0.01$). There were no significant interactions.

The average pruning weight was higher in 2006 compared with 2007. Pruning weight averaged 2.0 and 1.2 kg/vine in 2006 and 2007, respectively.

Table 6.11: Effect of increased irrigation late season on pruning weights (kg/vine) of two crop loads.

	pruning weight (kg/vine)		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned (TH)	2.0	1.2	1.6
Unthinned (UN)	2.0	1.2	1.6
	ns	ns	
Irrigation			
Standard (SD)	1.9	1.1 a	1.9
Double Irrigation (DI)	2.0	1.3 b	2.0
	ns	**	
Interaction	ns	ns	

Means within columns separated with different letters differ significantly at p<0.01 *, **, ***, ns, indicate significance at p<0.05, 0.01, 0.001, or not significant respectively.

Y/P Ratio: The yield to pruning weight ratios (Y/P) for crop load, irrigation, and the interactions are presented in Table 6.12. Crop load had a significant effect on Y/P in 2007, but was not significant in 2006. The UN had a higher Y/P ratio relative to the TH. There were no significant differences in the Y/P ratio due to irrigation. Moreover, there were no significant interactions between crop load and irrigation. The Y/P ratios were higher overall in 2007 than in 2006.

Table 6.12: Effect of increased irrigation late season on the Y/P ratio in two crop loads.

	Y/P ratio		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned (TH)	2.4	4.8 a	2.4
Unthinned (UN)	2.4	6.5 b	2.4
	ns	***	
Irrigation			
Standard (SD)	2.4	5.9	4.2
Double Irrigation (DI)	2.4	5.4	3.9
	ns	ns	
Interaction	ns	ns	

Means within columns separated with different letters differ significantly at p<0.001 *, **, ***, ns, indicate significance at p<0.05, 0.01, 0.001, or not significant respectively.

6.3.4 Berry Development

Degrees Brix: The °Brix development within the four crop load x irrigation treatments is presented in Figures 6.5 and 6.6 for 2006 and 2007, respectively. There were similar patterns in °Brix development for both crop loads; yet the greatest differences occurred between irrigation methods. The SD irrigation at each crop load had higher °Brix relative to the DI—this pattern remained throughout ripening until harvest.

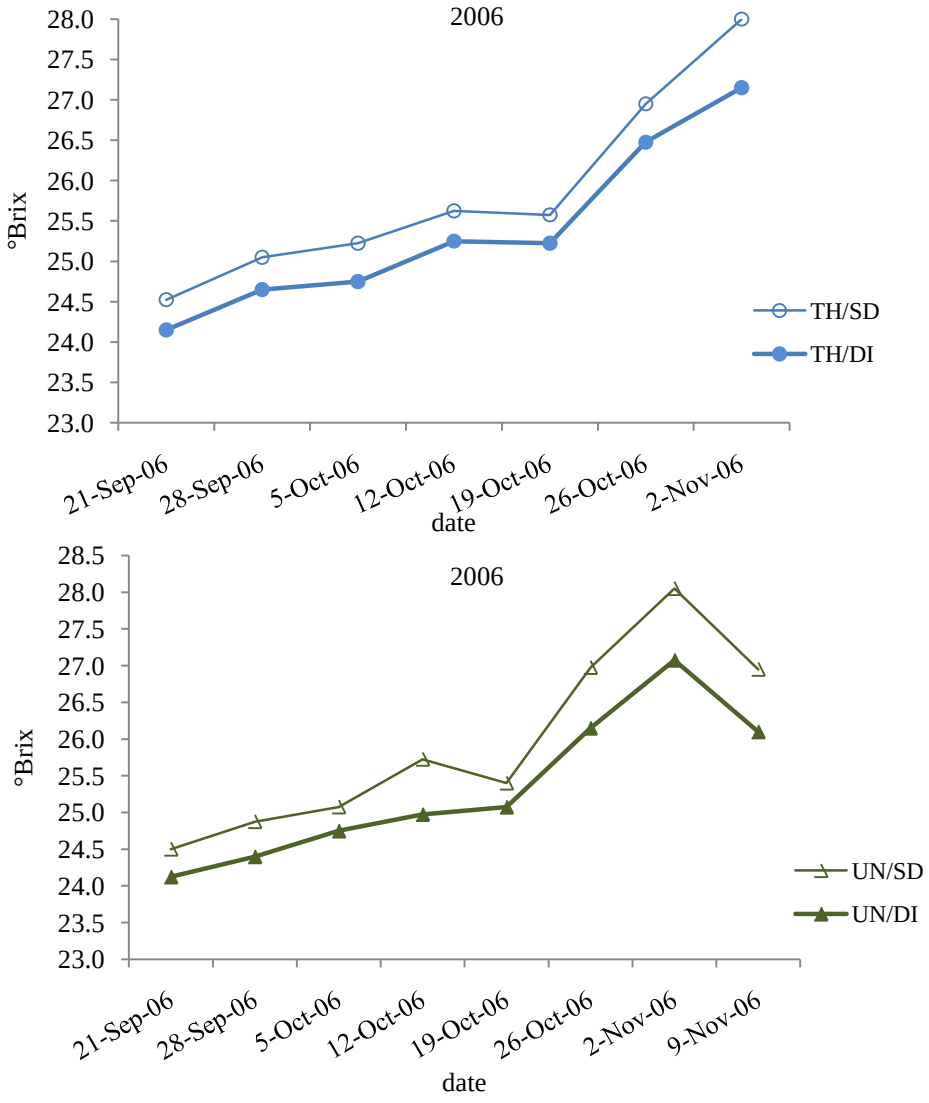


Figure 6.5: Crop load and irrigation effects on °Brix development during the later stages of ripening in 2006. Figure 6.5 (a) is the thinned (TH) crop load and Figure 6.5 (b) is the unthinned (UN). The DI was initiated on 6-Sep-2006.

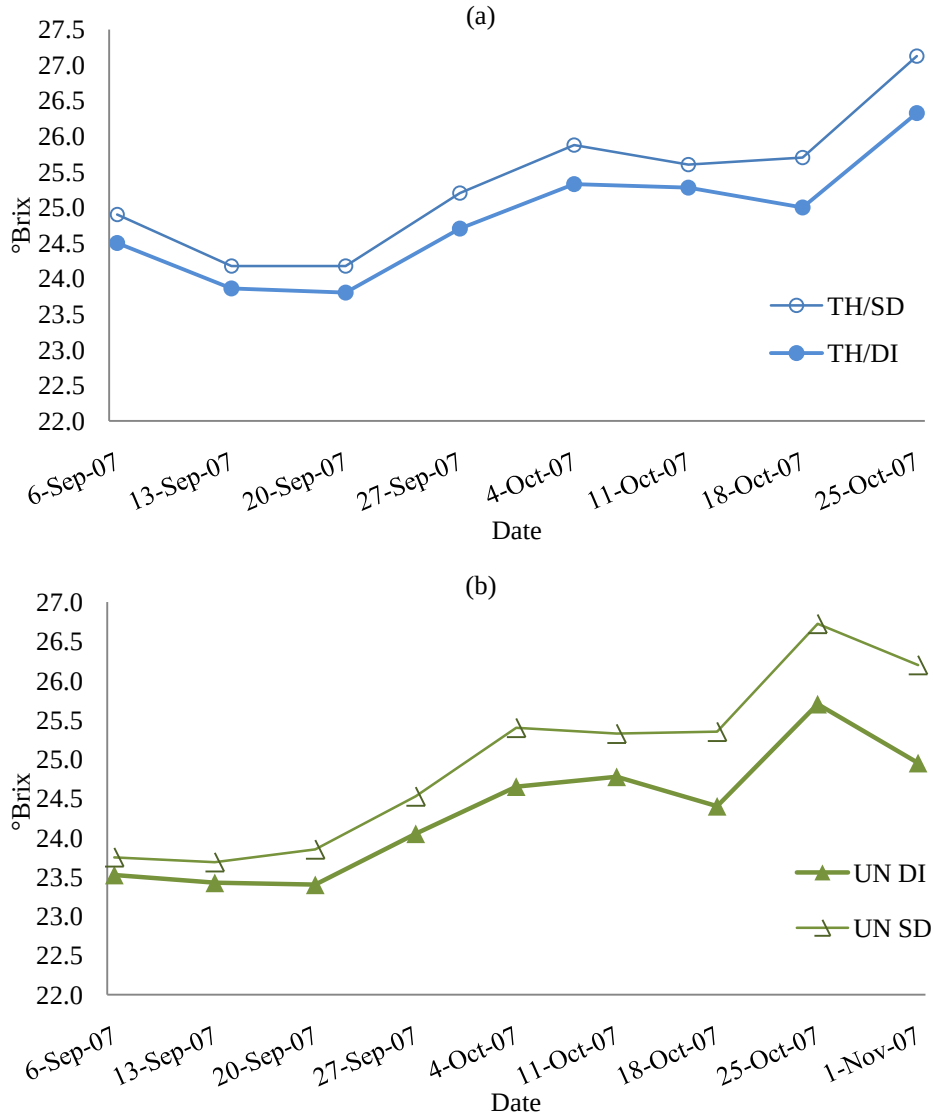


Figure 6.6: Crop load and irrigation effects on °Brix development during the later stages of ripening in 2007. Figure 6.6 (a) is the thinned (TH) crop load and Figure 6.6 (b) is the unthinned (UN). The DI was initiated on 30-Aug-2007.

Sugar (g) per berry: The absolute berry sugar content was calculated from berry weight and °Brix development data (Figures 6.7 and 6.8) to illustrate the effects of crop load and irrigation on sugar (g)/berry during the ripening period. The overall trend established that the DI at both crop loads had more sugar (g)/berry relative to the SD. The only exception was in 2006 when the TH/DI began decreasing slightly after 30-Oct-06.

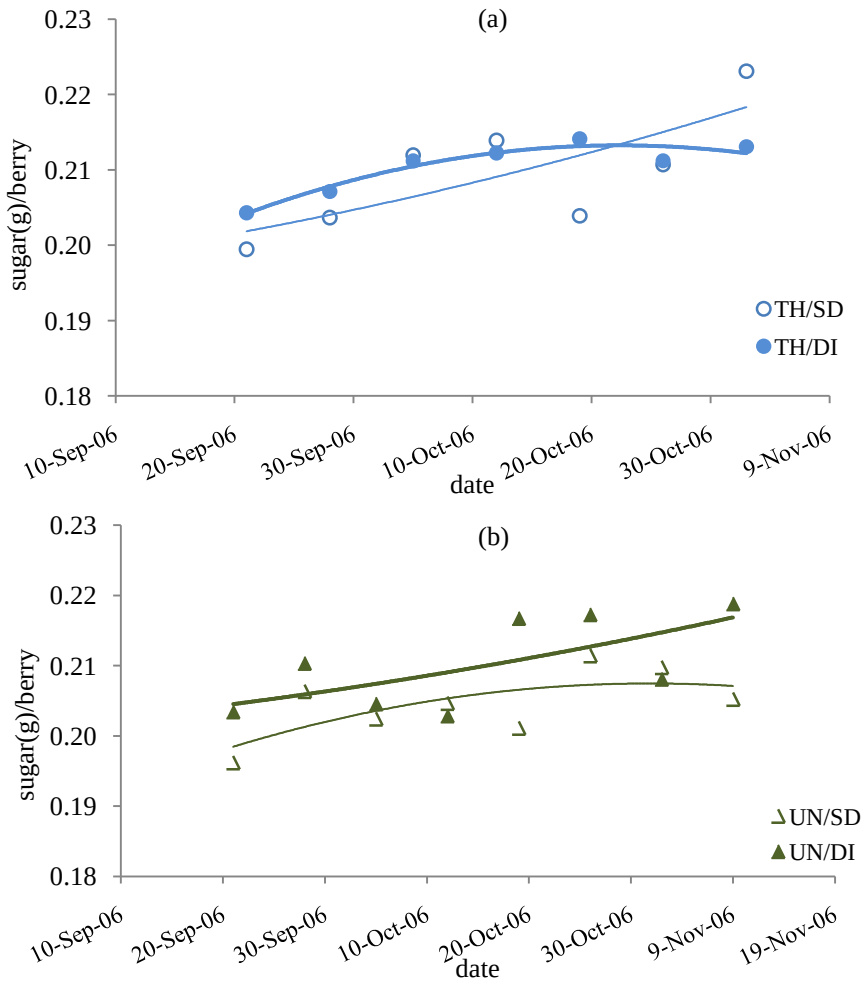


Figure 6.7: Crop load and irrigation effects on sugar (g)/berry during the later stages of ripening in 2006. Figure 6.7 (a) is the thinned (TH) crop load and Figure 6.7 (b) is the unthinned (UN). The bold line represents the DI.

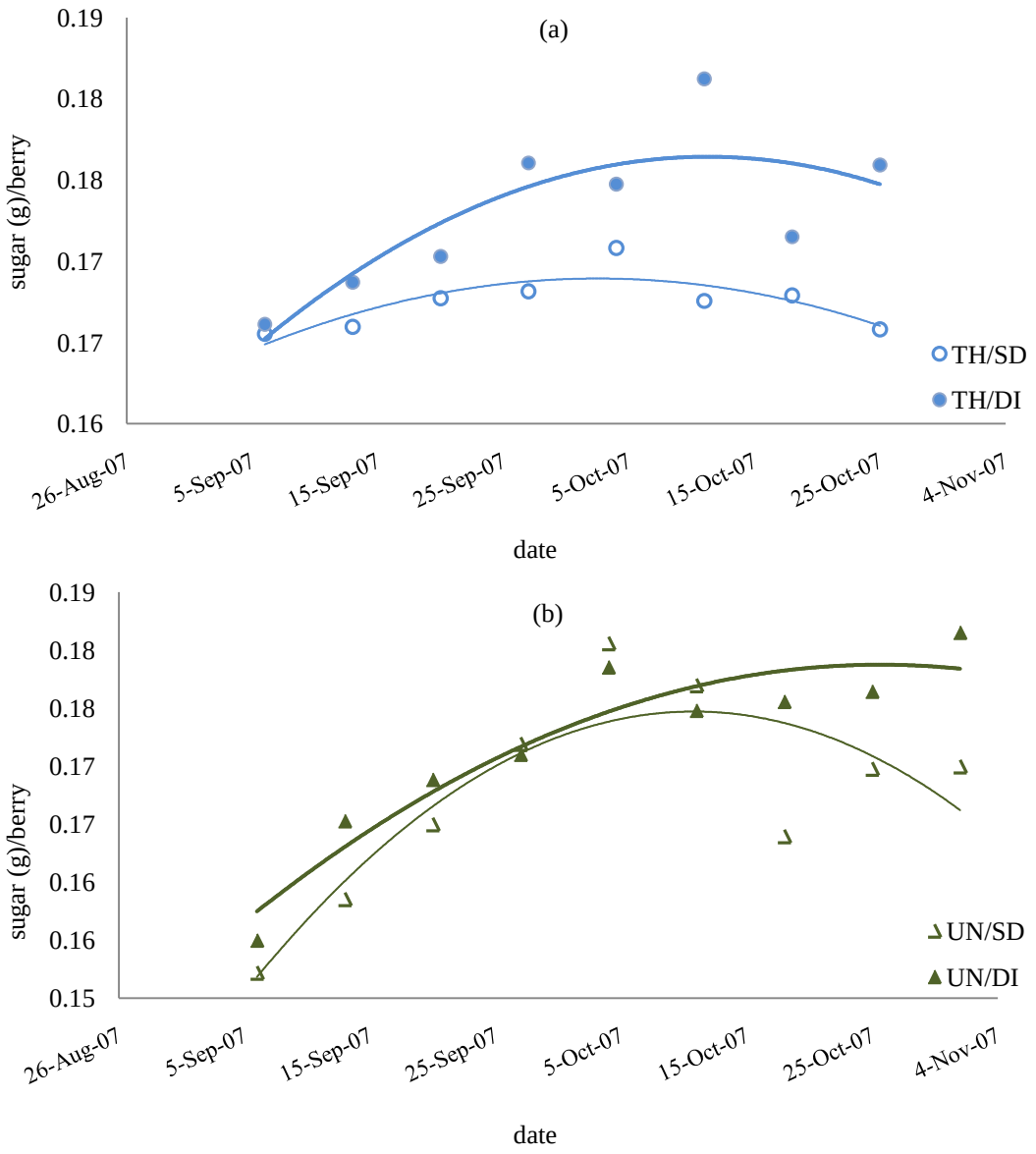


Figure 6.8: Crop load and irrigation effects on sugar (g)/berry during the later stages of ripening in 2007. Figure 6.8 (a) is the thinned (TH) crop load and Figure 6.8 (b) is the unthinned (UN). The bold line represents the DI.

pH development: The pH development in berries was measured weekly for each treatment (Figures 6.9 and 6.10). There were similar patterns in pH development between crop loads, although the UN had a lower pH at the first measurement (i.e. 21-Sept-06) and generally had a lower pH at each sampling date relative to the TH. The DI resulted in a lower pH in both crop

loads relative to the SD irrigation. In 2006, the UN/DI was substantially lower than the UN/SD on most measurements (Figure 6.9 b) whereas the TH/DI and TH/SD were not always substantially different, but did follow a similar trend.

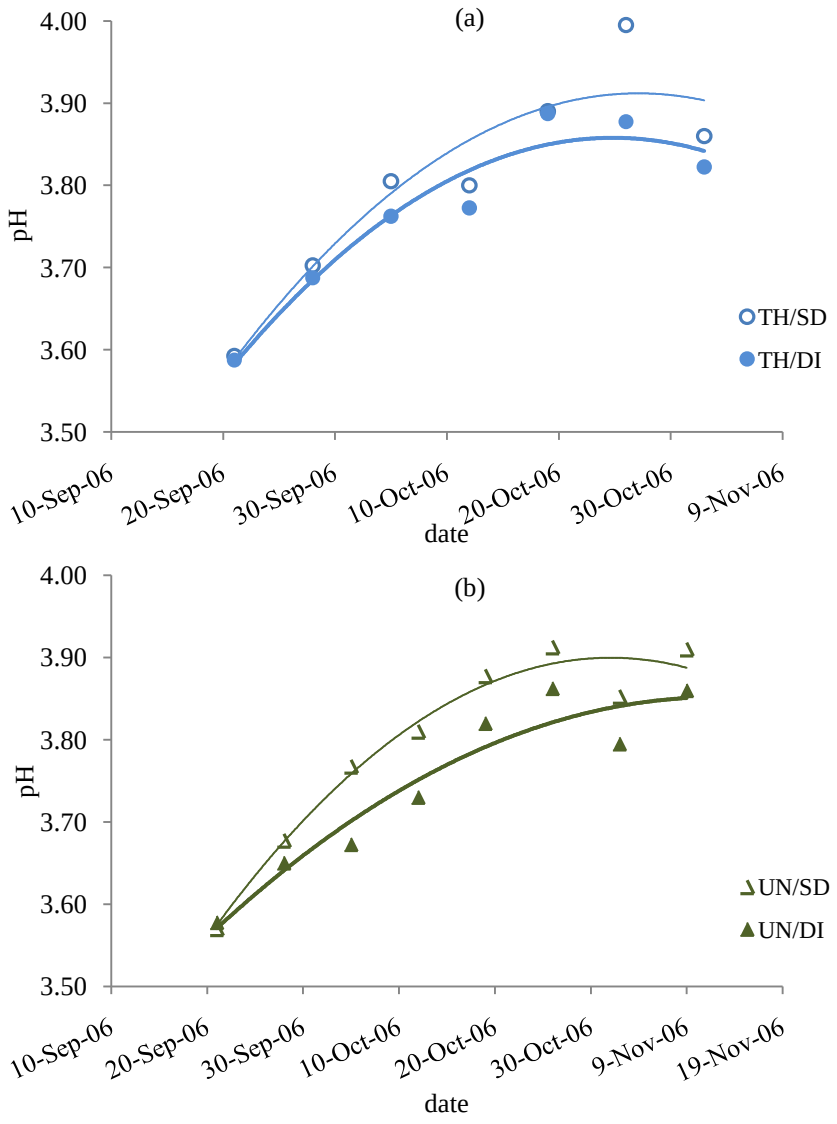


Figure 6.9: Crop load and irrigation effects on pH development during the later stages of ripening in 2006. Figure 6.9 (a) is the thinned (TH) crop load and Figure 6.9 (b) is the unthinned (UN). The bold line represents the DI.

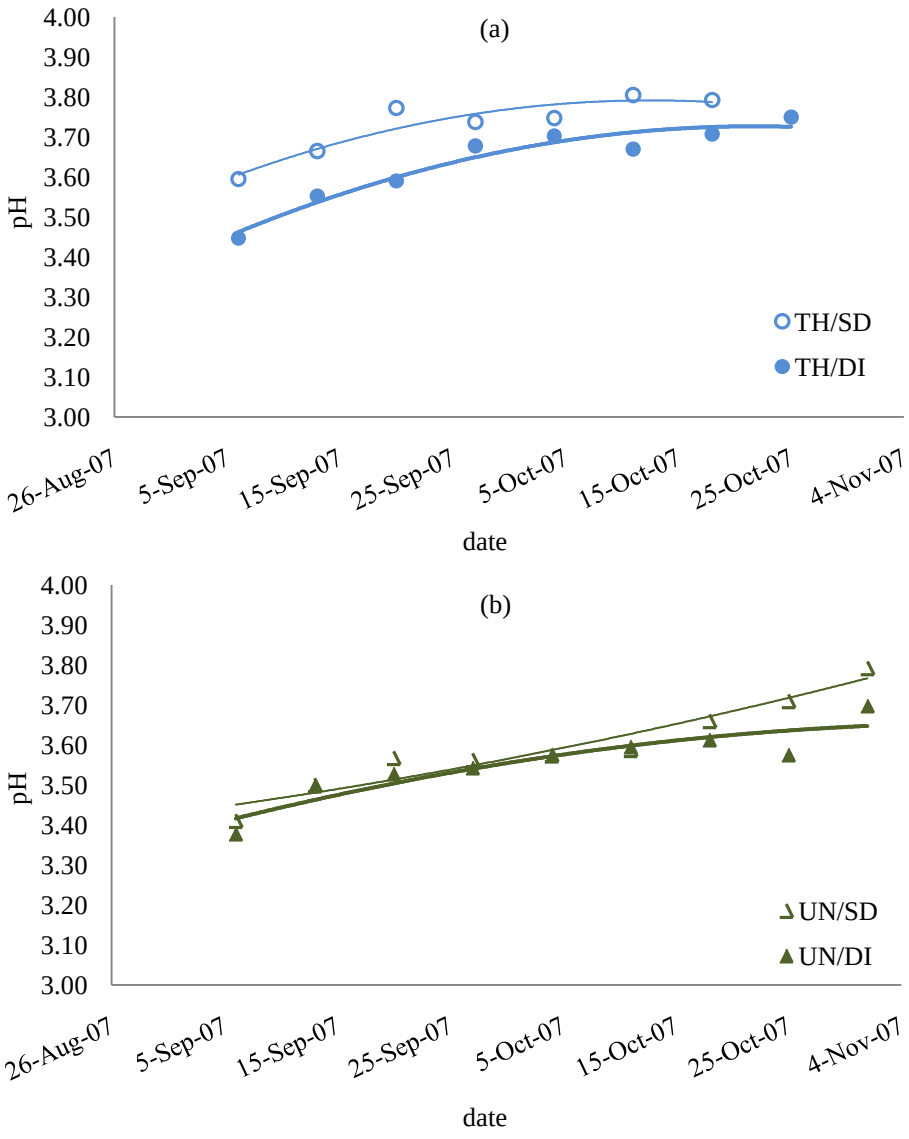


Figure 6.10: Crop load and irrigation effects on pH development during the later stages of ripening in 2007. Figure 6.10 (a) is the thinned (TH) crop load and Figure 6.10 (b) is the unthinned (UN). The bold line represents the DI.

TA development: The TA was measured on a weekly basis to analyze the effects of irrigation and/or crop load on TA development (Figures 6.11 and 6.12). Certainly, all treatments decreased in TA as ripening occurred. However, the overall trend illustrates that TA was lower for the SD

relative to the DI. This difference appeared to be more prominent for the UN as compared with the TH.

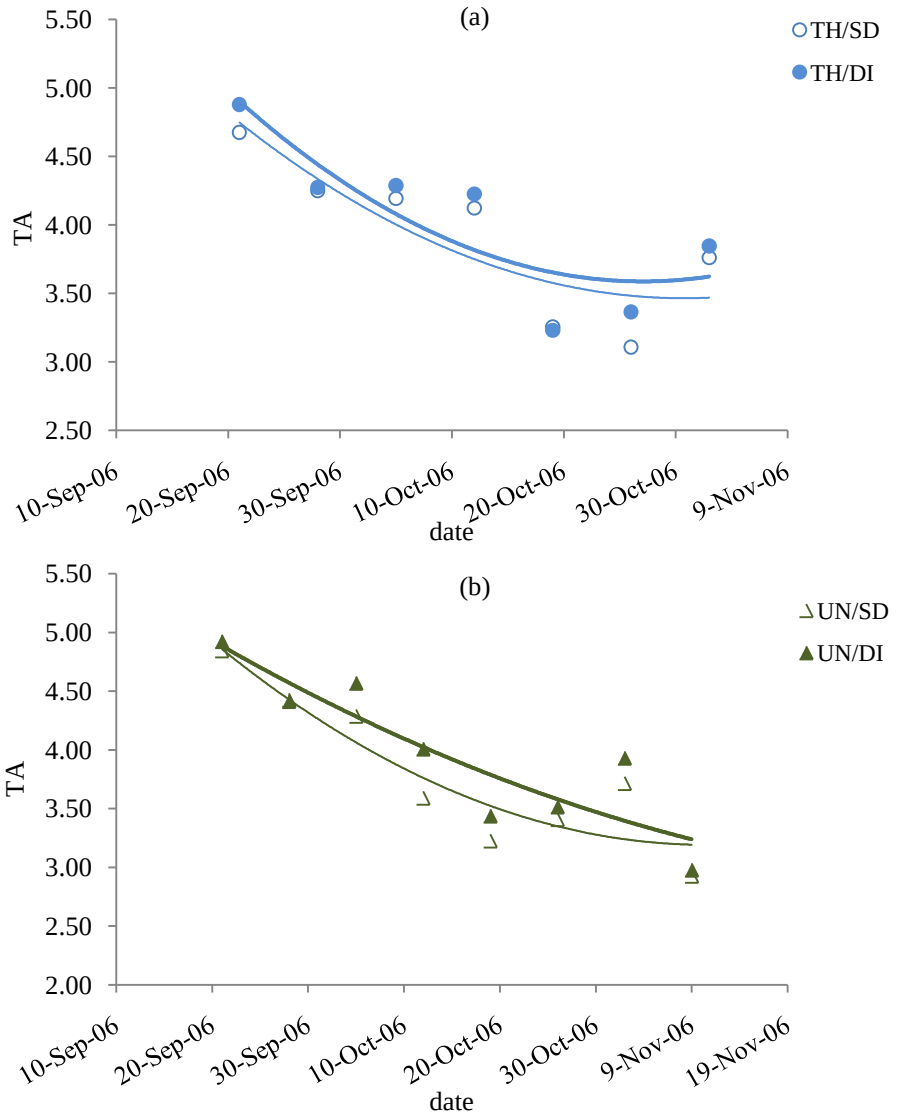


Figure 6.11: Crop load and irrigation effects on TA development during the later stages of ripening in 2006. Figure 6.11 (a) is the thinned (TH) crop load and Figure 6.11 (b) is the unthinned (UN). The bold line represents the DI.

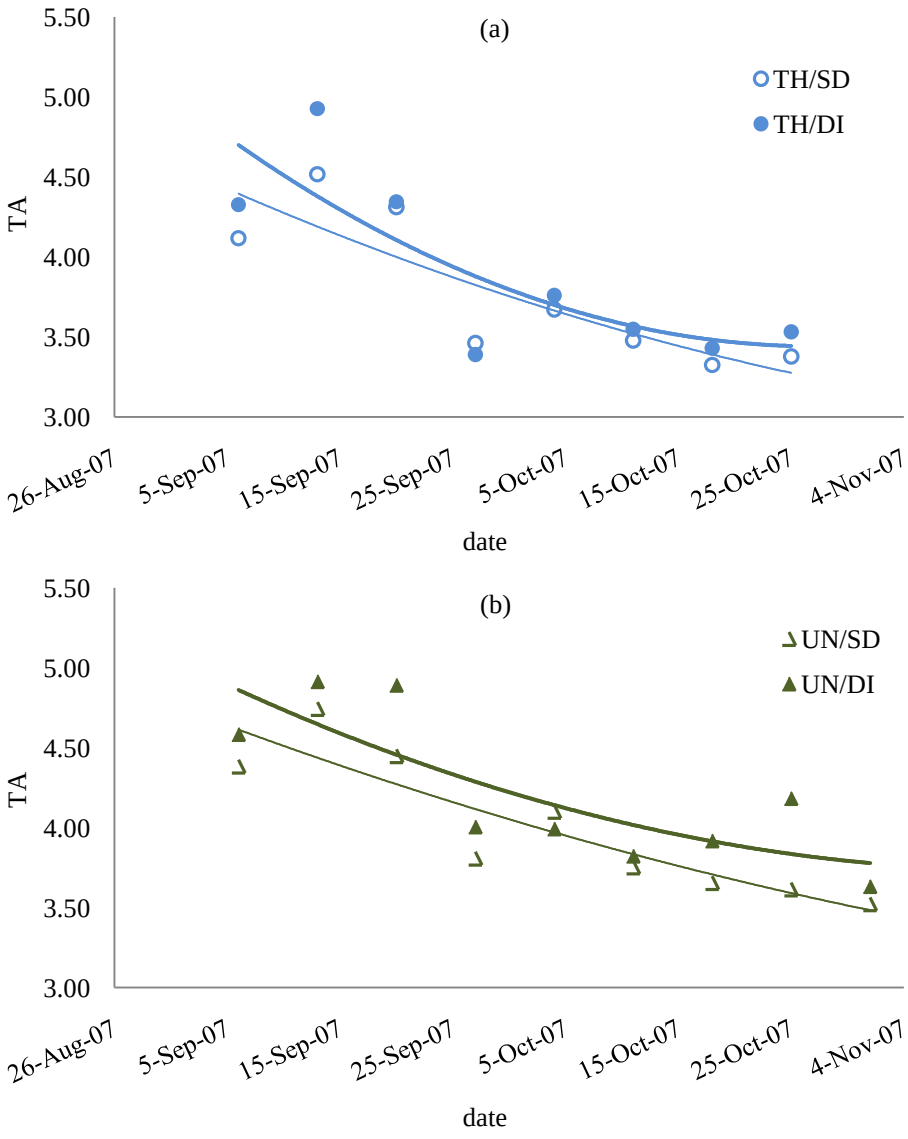


Figure 6.12: Crop load and irrigation effects on TA development during the later stages of ripening in 2007. Figure 6.12 (a) is the thinned (TH) crop load and Figure 6.12 (b) is the unthinned (UN). The bold line represents the DI.

6.3.5 Fruit and Wine Composition

Degrees Brix at harvest: Both crop load and irrigation had a significant effect on °Brix at harvest in each year (Table 6.13). Degrees Brix was significantly affected by crop load, although the pattern changed between 2006 and 2007. Degrees Brix in the UN was significantly greater than

the TH in 2006. In contrast, the TH was significantly greater than the UN in 2007 although the grand mean for both years were the same. The SD irrigation had significantly greater °Brix at harvest in both years and a grand mean of 27.3 °Brix as compared with the DI which averaged 26.7 °Brix.

Table 6.13: Effect of crop load and increased irrigation during the later stages of ripening on juice soluble solids (°Brix) at harvest.

	Brix *juice*		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned (TH)	27.6 a	26.5 a	27.0
Unthinned (UN)	27.9 b	26.1 b	27.0
	**	**	
Irrigation			
Standard (SD)	28.1 a	26.5 a	27.3
Double Irrigation (DI)	27.4 b	26.1 b	26.7
	***	**	
Interaction	ns	ns	

Means within columns separated with different letters differ significantly at $p<0.001$ or 0.01 by LSD. *, **, ***, ns, indicate significance at $p<0.05$, 0.01, 0.001, or not significant respectively.

pH juice: The pH in juice at harvest was significantly affected by crop load and irrigation (Table 6.14). The TH was significantly lower than the UN in 2006 but was significantly greater than the UN in 2007. The grand mean was greater for the TH. The SD had a significantly greater pH than the DI in both years, although the grand mean was the same for the SD and DI. A significant interaction occurred in 2006.

Table 6.14: Effect of crop load and increased irrigation during the later stages of ripening on juice pH at harvest.

	pH *juice*		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned (TH)	3.87 a	3.78 a	3.80
Unthinned (UN)	3.90 b	3.68 b	3.70
	**	***	
Irrigation			
Standard (SD)	3.90 a	3.74 a	3.80
Double Irrigation (DI)	3.88 b	3.72 b	3.80
	*	*	
Interaction	**	ns	

Means within columns separated with different letters differ significantly at p<0.001, 0.01 or 0.05 by LSD. *, **, ***, ns, indicate significance at p<0.05, 0.01, 0.001, or not significant respectively.

TA juice: There were significant differences in juice TA at harvest due to both crop load and irrigation (Table 6.15). The changes in TA due to crop load were different for 2006 and 2007. The TH had significantly greater TA than the UN in 2006 and in contrast was significantly lower than the UN in 2007. The grand mean was greatest for the TH crop load (4.22 g/L) as compared with the UN at (4.18 g/L); however, this would not constitute a substantial difference in terms of practical winemaking. The DI had a higher TA than the SD in both years, although it was only significantly higher in 2007.

Table 6.15: Effect of crop load and increased irrigation during the later stages of ripening on titratable acidity (TA) in juice at harvest.

	TA *juice (g/L)*		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned (TH)	4.46 a	3.97 a	4.22
Unthinned (UN)	3.81 b	4.55 b	4.18
	***	***	
Irrigation			
Standard (SD)	4.12	4.16 a	4.14
Double Irrigation (DI)	4.16	4.36 b	4.26
	ns	*	
Interaction	*	ns	

Means within columns separated with different letters differ significantly at p<0.001 or 0.05 by LSD. *, **, ***, ns, indicate significance at p<0.05, 0.01, 0.001, or not significant respectively.

YAN: The yeast assimilable nitrogen (YAN) was measured on juice at harvest (Table 6.16). Crop load had a significant effect on YAN in 2007. The TH had a significantly greater YAN than the UN. There were no significant differences due to irrigation or the interaction of crop load and irrigation.

Table 6.16: Effect of crop load and increased irrigation during the late stages of ripening on YAN in juice at harvest.

	YAN (mg/L)		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned (TH)	181.6	156.5 a	169.1
Unthinned (UN)	191.4	138.9 b	165.2
	ns	*	
Irrigation			
Standard (SD)	180.5	148.5	164.5
Double Irrigation (DI)	192.5	146.9	169.7
	ns	ns	
Interaction	ns	ns	

Means within columns separated with different letters differ significantly at p<0.05 by LSD. *, **, ***, ns, indicate significance at p<0.05, 0.01, 0.001, or not significant respectively.

Wine Ethanol: Ethanol (ETOH) was measured on press wine (Table 6.17) and was consistently greater in the TH crop load although only significantly greater in 2007. There were no significant differences between the SD and DI. Furthermore, no significant interactions occurred.

Table 6.17: Effect of crop load and increased irrigation during the late stages of ripening on Ethanol (ETOH) in juice at harvest.

	ETOH (%)		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned (TH)	15.1	14.3 b	15.1
Unthinned (UN)	14.6	14.0 a	14.6
	ns	*	
Irrigation			
Standard (SD)	14.8	14.1	14.5
Double Irrigation (DI)	14.9	14.2	14.5
	ns	ns	
Interaction	ns	ns	

Means within columns separated with different letters differ significantly at $p<0.05$ by LSD. *, **, ***, ns, indicate significance at $p<0.05$, 0.01, 0.001, or not significant respectively.

6.3.6 Wine Color and Phenolics

Total phenols: Total phenols were measured on treatment wines after the completion of malolactic fermentation and are presented in Table 6.18. The TH crop load was consistently greater for total phenols, although only significant in 2007 ($p<0.001$). There were significant differences due to irrigation each year. The SD was significantly greater than the DI in total phenols and averaged 1.95 nm greater than the DI for the two years. A significant interaction occurred in 2007 ($p<0.05$).

Table 6.18: Effect of crop load and increased irrigation during the late stages of ripening on total phenols in wine post malolactic fermentation (*pml*).

	total phenols *pml* (nm)		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned (TH)	38.1	43.0 b	40.6
Unthinned (UN)	37.5	40.9 a	39.2
	ns	***	
Irrigation			
Standard (SD)	38.4 b	43.3 b	40.8
Double Irrigation (DI)	37.2 a	40.6 a	38.9
	*	***	
Interaction	ns	*	

Means within columns separated with different letters differ significantly at $p<0.001$ or 0.05 by LSD. *, **, ***, ns, indicate significance at $p<0.05$, 0.01, 0.001, or not significant respectively.

Color density: Color density and its individual components of A(420 nm) and A(520nm) absorbance were measured in post malolactic wine (Table 6.19). Overall, crop load did not have a significant impact on color density. Neither color density nor A(520 nm) were significantly different due to crop load. The A(420 nm) was significantly affected by crop load in 2007 ($p<0.05$) but not in 2006. Irrigation had a significant effect on color density, A(420 nm) and A(520 nm) in both 2006 and 2007. Color density, including the A(420nm) and A(520 nm) was consistently greater in the SD relative to the DI. There were no significant interactions.

Table 6.19: Effect of crop load and increased irrigation during the later stages of ripening on color density A(420 nm) + A(520 nm) in wine post malolactic fermentation (*pml*).

	A(420 nm) *pml*		A(520 nm) *pml*		CD nm *pml*	
Treatment	**2006**	**2007**	**2006**	**2007**	**2006**	**2007**
Crop load						
Thinned (TH)	2.75	3.34 b	3.23	5.08	5.98	8.42
Unthinned (UN)	2.74	3.19 a	3.30	4.90	6.04	8.09
	ns	*	ns	ns	ns	ns
Irrigation						
Standard (SD)	2.84 b	3.47 b	3.40 b	5.38 b	6.23 b	8.84 b
Double Irrigation (II)	2.66 a	3.06 a	3.13 a	4.60 a	5.79 a	7.66 a
	***	***	**	***	*	***
Interaction	ns	ns	ns	ns	ns	ns

Means within columns separated with different letters differ significantly at p<0.001, 0.01 or 0.05 by LSD. *, **, ***, ns, indicate significance at p<0.05, 0.01, 0.001, or not significant respectively.

Hue: Wine hue was not consistently affected by crop load or irrigation (Table 6.20). The UN was significantly lower than the TH in 2006, but not significant in 2007. In contrast, there were significant differences between the SD and DI in 2007 but not in 2006. There was no significant interaction in any year.

Table 6.20: Effect of crop load and increased irrigation during the late stages of ripening on hue in wine post malolactic fermentation (*pml*).

	hue *pml*		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned (TH)	0.85 b	0.66	0.76
Unthinned (UN)	0.83 a	0.65	0.74
	*	ns	
Irrigation			
Standard (SD)	0.84	0.64 a	0.74
Double Irrigation (DI)	0.85	0.67 b	0.76
	ns	*	
Interaction	ns	ns	

Means within columns separated with different letters differ significantly at p<0.05 by LSD. *, **, ***, ns, indicate significance at p<0.05, 0.01, 0.001, or not significant respectively.

6.3.7 Wine Sensory

Expert rating: An expert panel was used to score the treatment wines made from the crop load and irrigation treatments (Table 6.21). Crop load did not have a significant effect on the wine quality score. The SD irrigation did score lower, and therefore better in wine quality and potential price point relative to the DI in both years. A significant interaction occurred in 2006 but not in 2007.

Table 6.21: Effect of crop load and increased irrigation during the late stages of ripening on expert wine score.

	expert score		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned (TH)	9.6	8.1	8.9
Unthinned (UN)	9.6	8.5	9.1
	ns	ns	
Irrigation			
Standard (SD)	9.5 a	7.9 a	8.7
Double Irrigation (DI)	9.7 b	8.6 b	9.2
	**	*	
Interaction	**	ns	

Means within columns separated with different letters differ significantly at $p<0.01$ or 0.05 by LSD. *, **, ***, ns, indicate significance at $p<0.05$, 0.01, 0.001, or not significant respectively.

Flavor intensity: The expert panel rated the flavor intensity of each treatment wine in both 2006 and 2007 (Table 6.22). There were no significant differences due to crop load, irrigation or the interaction of the two.

Table 6.22: Effect of crop load and increased irrigation during the late stages of ripening on the sensory score for the attribute flavor intensity.

	flavor intensity		
Treatment	**2006**	**2007**	***Grand Mean***
Crop load			
Thinned (TH)	4.3	5.2	4.7
Unthinned (UN)	4.2	5.6	4.9
	ns	ns	
Irrigation			
Standard (SD)	4.4	5.1	4.8
Double Irrigation (DI)	4.1	5.6	4.9
	ns	ns	
Interaction	ns	ns	

*, **, ***, ns, indicate significance at $p<0.05$, 0.01, 0.001, or not significant respectively.

Difference test: The triangle test was employed to test differences due to crop load and irrigation (Table 6.23). There were no significant differences between the SD and DI irrigation for the TH crop load. For the UN, significant differences were detected between the UN/SD and UN/DI only for replication four (R4)—there were no significant differences for all other replications. The percent correct out of a possible 24 showed that only the triangle test on R4 UN/SD versus R4 UN/DI had greater than 50 % correct.

Table 6.23: Results of difference testing on wines made from crop load and irrigation treatments in 2007.

	triangle test 2007		
Treatment	Total correct (out of 24)	Sig.	Percent correct (%)
R1 TH SD vs. R1 TH DI	11	ns	45.8
R2 TH SD vs. R2 TH DI	8	ns	33.3
R3 TH SD vs. R3 TH DI	8	ns	33.3
R4 TH SD vs. R4 TH DI	9	ns	37.5
R1 UN SD vs. R1 UN DI	9	ns	37.5
R2 UN SD vs. R2 UN DI	10	ns	41.7
R3 UN SD vs. R3 UN DI	11	ns	45.8
R4 UN SD vs. R4 UN DI	15	*	62.5

*, ns, indicate significance (Sig.) at $p<0.05$, or not significant respectively.

6.4 Discussion

6.4.1 Treatment Effects on Leaf Water Potential

Water potentials in vascular plants can be measured by means of a pressure chamber (Scholander *et al.* 1965). The graphs of mid day leaf water potential (LWP) demonstrate the vine response to the increased irrigation for both crop loads. On average the LWP of DI vines was 1.0 and 2.0 bars higher in 2006 and 2007, respectively. Regardless of crop load, the SD averaged 11.3 bars in both 2006 and 2007 whereas the DI averaged 10.4 and 9.3 bars, respectively. These results support those of Sanchez *et al.* (2006) and Mendez-Costabel (2007) who reported progressively higher stem water potentials in well irrigated vines (i.e. 1.2 ETc) and consequently higher photosynthesis. Roby *et al.* (2004) suggested that the optimal midday LWP for sugar accumulation was between -1.2 and -1.4 MPa i.e. -12 and -14 bars. Leaf water potential of vines irrigated at 100 % of ETc is generally at -10 bars or higher (Williams 2001). Additionally, it is recommended to initiate the season's first irrigation at -12 bars for red grape varieties (Williams

2001) as this is considered the onset of vine stress (L.E. Williams, personal communication, 2002). Generally, the level of stress gauged by mid-day LWP between: -10 to -12 bars, -12 to -14 bars, -14 to -16 bars and above -16 bars is considered mild stress, moderate stress, high stress, and severe stress, respectively (Bogart 2005). Therefore, the SD reached "moderate stress" on three dates in 2006, and both "high' and "moderate" stress for the first three dates in 2007. In 2007, all treatments became progressively less negative i.e. less stressed, as the season progressed towards harvest. This may be explained by cooler average temperatures and shorter days as harvest approached.

6.4.2 Treatment Effects on Yield Components

Berry weight development: Berry growth and final weight at harvest are known to be directly related to vine water status (Hardie and Considine 1976, Esteban *et al.* 1999, Rogiers *et al.* 2006a). Certainly, the additional irrigation in the DI maintained higher berry weight relative to the SD suggesting that either the DI had a greater flow of water into the berry, or lost less water due to berry dehydration, or both. In addition, these results support Rogiers *et al.* (2001), Bondada *et al.* (2005), Keller *et al.* (2006), and Rogiers *et al.* (2006a) who reported that the xylem remains qualitatively connected to the berry late in development. Additionally, Tilbrook and Tyerman (2008) demonstrated that the vascular connection remains vital—and therefore hydraulic conductivity of xylem cells could be regulated by the activity of aquaporins.

Crop load and irrigation had an interactive effect on berry weight (Table 6.24). The UN/DI maintained the highest berry weight among all treatments in each year and, on average, maintained 8 % greater peak berry weight relative to all other treatments. Furthermore, in 2007 the UN/DI had its greatest berry weight at the final measurement. These results raise a

challenging question as to what exactly caused the UN/DI to maintain more berry weight than the TH/DI. Based on the research of Tilbrook and Tyerman (2008) it is likely that the hydraulic connectivity was more intact relative to the other treatments and/or had the least amount of backflow. Water 'backflow' from the berry back to the stem has been shown to occur in water stressed vines (Lang and Thorpe 1989) and through measured hydraulic conductivity in Shiraz berries (Tyerman *et al.* 2004). Additionally, the work of Bondada *et al.* (2005), Keller *et al.* (2006) and Tilbrook and Tyerman (2008) qualitatively demonstrated that backflow can occur from the berry to vine; however, there are large quantitative differences between varieties. In this experiment, the SD irrigation imposed a greater water stress on the vine, as indicated by the LWP data, and this suggests a lower leaf transpiration rate—although this was not measured. It is plausible that reduced evaporation in the berry and/or less leaf transpiration would have increased backflow of water from the berry to vine and would therefore explain the greater reduction in berry weight in the SD treatments. In contrast, Rogiers *et al.* (2004) reported greater losses in Shiraz berries of standard irrigated vines compared with deficit irrigated vines after maximum volume was attained, with a concluding hypothesis that changes in transpiration rate may have been greater in berries of the larger volume (i.e. standard irrigated). These outcomes highlight the need for further examination of crop load effects on the hydraulic relations between the vine and berry—particularly for the variety Cabernet Sauvignon.

Table 6.24: The interaction of crop load and irrigation during the later stages of ripening on the percent (%) change in berry weight from peak berry weight to final berry weight.

	% change berry weight		
Treatment	**2006**	**2007**	***Grand Mean***
TH/SD	-7.1	-11.6	**-9.4**
UN/SD	-8.4	-8.6	**-8.5**
TH/DI	-8.0	-8.2	**-8.1**
UN/DI	-2.3	+0.4	**-1.0**

Seasonal differences did occur. The average berry size, among all treatments was considerably greater in 2006 relative to 2007. Generally, treatments began to decrease at an earlier date in 2006 relative to 2007.

Berry weight at harvest: Berry size was reported by Williams (2001) to reach its maximum with irrigation regimes which met 75 % (i.e. 0.75) of full ETc. In the present experiment the DI regime during the late stages of ripening had a major effect on berry weight. The implementation of the DI just after 20 °Brix did in fact sustain berry weight. Given that the increased irrigation was implemented from 20 °Brix it is unlikely that berry size *per se* increased—rather, a greater proportion of peak berry weight was maintained with the increased irrigation. These results are in agreement with Sanchez *et al.* (2006) who reported that increasing irrigation level from 0.7 to 1.2 % ETc during late ripening caused significantly higher maximum yields and berry weight for Merlot. Mendez-Costabel (2007) reported similar results on Cabernet Sauvignon grown in Sonoma. Additionally, these results support the proposition by Keller *et al.* (2006) that berries remain hydraulically connected to the vine beyond veraison.

Crop load affected berry weight but to a lesser extent. In 2007, the UN had a significantly greater berry weight at harvest relative to the TH. This could have been due to a more intact

hydraulic connection influenced by the rate of ripening or perhaps was influenced by being harvested one week later and thereby receiving one extra irrigation set. Although these berry weight results are conflicting between the two years, many crop load studies have reported conflicting results for changes in berry size (Weaver and Pool 1969, Kaps and Cahoon 1989, Chapman *et al.* 2004, Keller *et al.* 2005).

Overall, the treatments had greater effects on berry weight in 2007 than 2006. This may have been due to weather differences specific to the individual season such as winter rainfall and temperature during ripening; or could have been a carryover effect from the 2006 treatments. Additionally, the irrigation set applied pre harvest was substantially greater in 2007. The overall irrigation strategy was based on the ETc calculation, but in 2007 we decided to increase the amount of irrigation hours/liters of the final pre-harvest irrigation to be more consistent with a commercial post-harvest irrigation practice. In all probability this increase influenced berry weight at harvest and presents a potential irrigation strategy to mitigate berry weight loss. Growers could essentially apply what would have been their post harvest irrigation, pre harvest—thus maintaining similar annual pumping costs.

Berries/cluster: Neither crop load nor irrigation treatments affected berries per cluster. The number of berries per cluster is influenced by the number and size of the inflorescence primordia determined during the previous season (Mullins *et al.* 1992) and the number of flowers which develop into berries (McCarthy 1997) e.g. percent fruit set. The 2006 results are explainable given that the treatments were not implemented until post fruit set and at 22 °Brix for crop thinning and irrigation, respectively. Additionally, Goodwin and Jerie (1989) reported no differences in berries per cluster for bud break to bloom deficit-irrigated Chardonnay within the

first year. The data reported here indicate that the 2006 treatments did not affect berries per cluster in the following year (2007).

Clusters/vine: The cl/vine data confirms that crop thinning was implemented successfully each year by the labor crew and did cause a significant difference ($p<0.001$). The TH/SD had significantly more clusters per vine than the TH/DI. This interaction occurred in 2007 only, and perhaps is explained by a change in bud fruitfulness. The growth data indicated a significant increase in pruning weight/vine and mean cane weight which may have increased shading—although this theory is not supported by the ceptometer data which was 'ns' in all years.

Mean cluster weight: Mean cluster weight was surprisingly greater for the TH in 2007—this contrasted with 2006—and mean cluster weight in thinned vines of the crop load/extended ripening experiment (presented in Chapter 3). Overall, these data suggest that crop load in *this* experiment (crop load x irrigation) had less influence on mean cluster weight than in the main experiment (crop load x target °Brix at harvest). Possibly, this was influenced by the actual number of clusters thinned and/or the timing of thinning. The DI increased mean cluster weight by 8 g/cluster on average and was of greatest significance in 2007. The 2007 irrigation schedule included a longer pre-harvest irrigation set and most likely influenced the higher mean cluster weight.

Yield/vine: Crop load differences occurred as a result of the cluster thinning—confirmed by the significant differences in yield/vine between the TH and UN. The fruit weight per vine in the DI was significantly greater in 2007—a 10 % increase relative to the SD. Overall, the 2007 season had more significant differences in yield components between the DI and SD relative to 2006.

There were no significant differences in yield between the TH/SD and TH/DI. However, the UN/DI had significantly higher yield/vine than the UN/SD indicating that the UN vines maintained more weight (i.e. kg fruit/vine). The irrigation effects on yield are consistent with Rogiers *et al.* (2004), Sanchez *et al.* (2006), Mendez-Costabel (2007) and Intrigliolo and Castel (2008) who all reported significant yield increases due to increased irrigation. Additionally, many studies have shown that yield decreases as water availability decreases (Smart *et al.* 1974, Hardie and Considine 1976, Stevens *et al.* 1996, McCarthy 1997, Esteban *et al.* 1999). Although measurements were not taken, the data suggest that the UN had better conductivity from the vine to the berry; or perhaps a bigger sink effect and therefore more substrate from the source. These findings and theories warrant further research on the interaction of crop load and irrigation during the later stages of ripening.

Tonnes per hectare: There were significant increases in tonnes per hectare (t/ha) due to both crop load and irrigation. Interestingly, the DI increased t/ha relative to the SD, and most noticeably in 2007. The interaction in 2007 suggests that increased irrigation in the UN (i.e. UN/DI) was more effective than in the TH (i.e. TH/DI) (Figure 6.13). The TH/DI was only 0.30 t/ha greater than the TH/SD as compared with the UN/DI which was 1.3 t/ha greater than the UN/SD. This represents a 4 % increase for TH/SD relative to TH/DI compared with a 16 % increase for UN/SD relative to UN/DI. Moreover, this interaction demonstrates that the UN crop load was affected more by the increased irrigation than the TH crop load. These results concur with those of Mendez-Costabel (2007) who showed that yield increased by 10.5 % and 10.7 % in Merlot and Cabernet Sauvignon vines, respectively, which received irrigation above 100 % ETc—and relative to vines irrigated at a lower % of ETc—during late ripening.

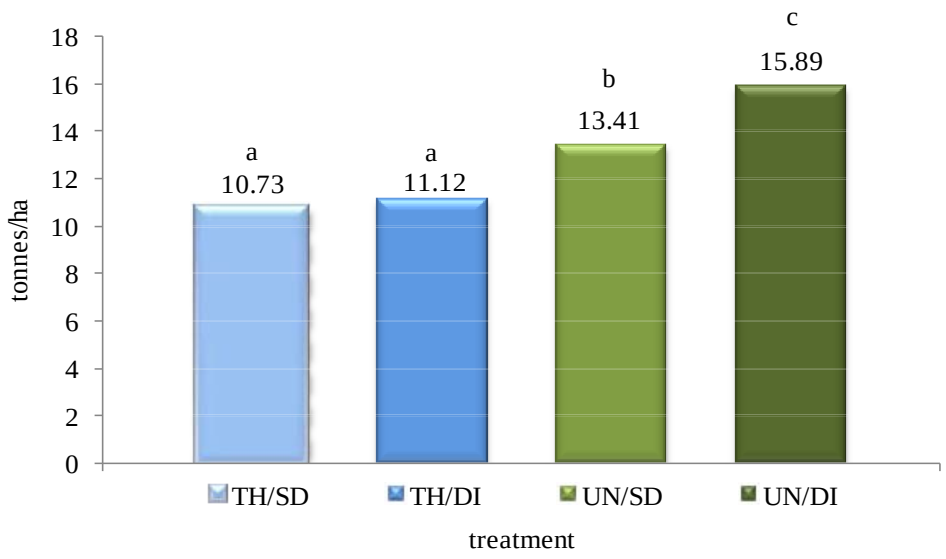


Figure 6.13: The interaction of crop load and irrigation on tonnes per ha in 2007. Columns with different letters differ significantly by LSD.

6.4.3 Treatment Effects on Vine Growth

PAR and LAI: Overall, the ceptometer measurements indicated that PAR in the fruiting zone was not significantly altered due to the crop load and/or irrigation treatments. Additionally, LAI did not change as a result of the treatments. These results conflict with the pruning weight and mean cane weight data, which increased in the DI relative to the SD in 2007. The work of Stevens *et al.* (1995) reported that reduced shoot growth correlated with increased water stress indices and lower stem water potential—the onset of irrigation treatments in their experiment was based on soil moisture rather than ripening level and may explain the different outcomes. In 2007, LAI and PAR were measured at harvest but pruning weight and mean cane weight were measured at winter pruning in January. It is conceivable that the large pre harvest irrigation on the DI, followed by crop removal, initiated post harvest shoot growth in the DI and would thereby explain the contradicting results between LAI and PAR, and winter pruning weights.

Additionally, there were no signs of yield component compensation due to crop thinning which suggests few differences in the partitioning of grapevine resources between vegetative growth and reproductive growth. Furthermore, the work of Mendez-Costabel (2007) showed that increased irrigation during late ripening—to levels above 100 % ETc—had no effect on total vine leaf area.

Shoots/vine: Shoots per vine were not different in either 2006 or 2007 which confirms that the pruning and shoot thinning practices were consistent among treatments. Therefore, differences due to shoots/vine can be eliminated as the basis for changes in growth or yield components among treatments. However there was a slight difference in shoots per vine between seasons in that 2007 averaged 5 shoots per vine more than in 2006. This difference was most likely the result of basic human error in shoot thinning among the labor crews.

Mean cane weight: Crop load did not have a significant effect on mean cane weight and demonstrates that crop thinning did not increase canopy growth. These results differed from the other experiment (described in Chapter 3) which had increased mean cane weight as clusters per vine increased. The timing of the thinning was near or just past E-L 31. It is likely that a greater reduction in crop load would have resulted in increased mean cane weight.

Interestingly, the DI significantly increased mean cane weight in 2007 but not in 2006. This suggests that the effects of increased irrigation during the later stages of ripening on mean cane weight may be dependent on the seasonal characteristics, or there was a carry-over effect due to the previous year's increased irrigation. Becker and Zimmerman (1984) reported reduced starch concentrations in potted vines that were deficit irrigated compared with well irrigated potted vines.

Pruning weights: Moreover, crop load did not have a great effect on pruning weight per vine in this experiment. Although differences in clusters per vine were significant, it was not enough to significantly alter growth.

However, irrigation did impact pruning weights in the second year of the experiment. Pruning weights were consistent with mean cane weight in that the DI was significantly greater than the SD in 2007 although no differences occurred in 2006. It is likely that the increased irrigation promoted additional growth of laterals, thereby resulting in both increased cane weight and overall pruning weight per vine. What remains unknown is *when* the increased growth occurred and why only 2007 was affected. Possibly growth increased at the onset of irrigation treatments (i.e. 22 °Brix) and gradually increased shoot growth, or may have resulted from the large pre-harvest irrigation set, or both. As mentioned previously, both years followed the same irrigation strategy except that a larger pre harvest irrigation was applied to the DI in 2007 in order to emulate commercial post harvest irrigation and increase practicality of the experiment. The additional pre harvest irrigation on the DI, in conjunction with the fruit harvested from the vine just 48 hours later, resulted in a luxury of water within the soil profile—and thereby could have increased lateral shoot growth. Pruning weights were measured the following winter therefore the grapevines had time to utilize the additional water within the soil profile until complete dormancy.

An alternative explanation is that in 2007 the overall averages for mean cane weight and pruning weights were considerably lower relative to 2006. Perhaps using the ETc calculation for 2007 was less precise than in 2006 due to the smaller canopy—assumed by the reduced pruning and cane weights. The ETc calculation uses the same crop coefficient (Kc) based on growing degree

days rather than the *actual* growth and leaf area. The smaller canopy in 2007 would have required less water, but was calculated using the standard Kc for the growing degree days.

Carbohydrate reserve accumulation depends on the rate of photosynthesis and the partitioning of that photosynthate between shoot, root and fruit growth and storage (Howell 2001). The work of Sanchez *et al.* (2006) and Mendez-Costabel (2007) reported increased photosynthesis and greener leaves due to increased irrigation in the later stages of ripening. Although these parameters were not measured in the current study, it is conceivable that photosynthesis was affected by the different irrigation regimes and could explain the increased growth in the DI. Furthermore, previous studies have indicated that reduced shoot growth, pruning weights and/or leaf area reduce carbohydrate reserves (Candolfi-Vasconcelos and Koblet 1990, Bennett *et al.* 2005). Further research is required in this area to gain a better understanding of irrigation effects on grapevines during the later stages of ripening and its potential effects on vine physiology and growth.

Yield/pruning ratio: The yield to pruning weight ratio was—significant due to crop load—only in 2007. This is surprising given that other yield components were significantly different between the TH and UN. This outcome concurs with Intrigliolo and Castel (2008) who reported no changes in Y/P and leaf area/yield ratios in irrigated and non irrigated Tempranillo, although vegetative growth increased. The UN had a significantly higher ratio in 2007 and suggests a better balanced vine. A ratio between 5-10 is considered within the optimal range (Bravdo *et al.* 1984, 1985, Dry *et al.* 2004). Generally vines in warmer climates are thought to be optimal near the higher values (Dry *et al.* 2004). These ratios demonstrate that unthinned vines can be within the optimal Y/P range and balanced. Furthermore, crop thinning is not always a necessary practice.

The Y/P ratios in each treatment were quite different for 2006 and 2007 (Table 6.25). The 2007 Y/P ratios were closer to optimal signifying better vine balance; however, the Y/P in 2006 indicates overly vigorous and under cropped vines among all treatments.

Table 6.25: The interaction of crop load and irrigation during the later stages of ripening on Y/P ratio in two seasons.

	Y/P	
Treatment	**2006**	**2007**
TH/SD	2.4	5.1
UN/SD	2.4	6.6
TH/DI	2.5	4.5
UN/DI	2.4	6.4

6.4.4 Crop Load and Irrigation Effects on Berry Development

Sugar ripening: Degrees Brix development shows that the SD irrigation consistently had higher °Brix than the DI. The berry weight development curves demonstrated that the SD had consistently lower berry weight relative to the DI. Possibly, there was a dehydration effect in the SD rather than an actual increase of sugar synthesis and the higher °Brix in the SD irrigation was a result of the 'concentration' effect rather than new synthesis of sugar in leaves translocated into the berry. This proposition is supported by the sugar per berry (g) curves which illustrated different responses to the °Brix development curves during the later stages of ripening.

Generally, the DI had more sugar (g)/berry relative to the SD—this was particularly apparent in 2007. In contrast, °Brix development demonstrated that the SD was consistently higher than the DI (Figures 6.4 and 6.5). Furthermore, these results support the proposition that increased °Brix in the SD was mainly due to berry dehydration and a concentration effect. Additionally, sugar weight per berry in the SD started to decline at an earlier date than the DI. The only exception

was for the TH in 2006 where the DI declined in sugar (g)/berry approximately two weeks before the SD. Various studies have also shown that the amount of sugar (g)/berry was always greatest in grapevines with the most water supplied throughout the season (Van Zyl 1984, Hepner and Bravdo 1985, Matthews and Anderson 1989, Mendez-Costabel 2007). Furthermore, the divergent patterns of the SD and DI increased as harvest approached. On average, sugar per berry was 6 % greater in the DI relative to the SD on the final measurement in 2007, in contrast with °Brix which averaged 3 % higher in the SD relative to the UN. Although berry water content (e.g. berry moisture) was not measured in the present experiment, the work of Sanchez *et al.* (2006) demonstrated that the rise in soluble solids (°Brix) was accompanied by an approximate 6 % decrease in berry moisture for two different irrigation treatments. Collectively, these results suggest that increased °Brix in the SD irrigation treatment was more likely a result of the 'concentration effect' due to berry dehydration. This contrasts with the DI which maintained more berry weight due to less dehydration, and consistently lower °Brix. These results support the concept that water flow post veraison is mainly via the phloem (Greenspan *et al.* 1994); and that increased water stress following veraison reduces phloem sugar loading (Wang *et al.* 2003).

pH and TA development: The SD irrigation at both crop loads, generally, had a higher pH and lower TA relative to the DI in the development curves—however the differences were not great. Bravdo *et al.* (1985) reported that juice pH was quite similar among different irrigation treatments. Neither irrigation nor crop load had a major effect on pH development in 2007—this is consistent with the findings of Sanchez *et al.* (2006). Additionally, Ginestar *et al.* (1998) reported that pH and TA concentration were unaffected by different post veraison irrigation treatments on Shiraz. Presumably the DI had more water per berry; therefore, it is conceivable

that this influenced pH and TA. Treatment effects did not alter results enough to warrant considerable changes for winemaking in practical terms.

6.4.5 Fruit and Wine Composition

Degrees Brix at harvest: Crop load was responsible for significant differences in °Brix at harvest, but was not consistent between years. The UN was harvested a week later than the TH each year and may have contributed to the higher °Brix in 2006. In 2007 the UN not only had one more week to ripen, but received a larger final irrigation relative to the final irrigation in 2006—and therefore may have caused the UN to be lower in °Brix at harvest in 2006 relative to 2007. Overall, the grand mean was the same which suggests that the differences due to crop load were not great across both years of the experiment.

Irrigation had a clear effect on °Brix at harvest in that the DI decreased °Brix at harvest although it only decreased by 0.6 °Brix, on average, between the two years.

pH and TA at harvest: Titratable acidity and pH are of great importance for juice stability and are commonly used as quality parameters primarily due to their influence on subsequent wine color and microbiological stability (Boulton 1980).

The effects of crop load on pH and TA of juice at harvest were conflicting between years. Juice pH was highest in the TH in 2006 and in contrast significantly lower relative to the UN in 2007. The grand mean of both years demonstrated that, overall, pH was highest in the TH relative to the UN. The effects of irrigation indicated that pH in the SD was significantly higher relative to the DI. Although significant, the pH differences between the SD and DI would not be

considered different enough in “practical” terms to be treated differently from a winemaking perspective. The same would apply for pH differences due to crop load in 2006.

Still, these results support those of Esteban *et al.* (1999) who reported higher pH in non irrigated relative to irrigated Tempranillo grapes. However, they conflict with those of Chapman *et al.* (2005) who reported pH was lowest in minimally irrigated vines as compared with standard and double irrigated Cabernet Sauvignon. In addition, Sanchez *et al.* (2006) reported no differences in pH in Merlot vines irrigated at 0.7 or 1.2 % ETc from 20 °Brix to harvest. Furthermore, results on the effects of irrigation and pH in juice are conflicting. It appears that timing of irrigation and actual amounts relative to the soil moisture of the site—and perhaps variety—play an integral role in these outcomes. Additionally, limited studies exist on irrigation specific to the later stages of ripening—and including extended ripening. The interaction in 2006 revealed that the UN/SD had a significantly higher pH relative to all other treatments (Figure 6.14) although the interaction was not significant in 2007. Again, the magnitude of these differences may not be relevant from a practical winemaking standpoint.

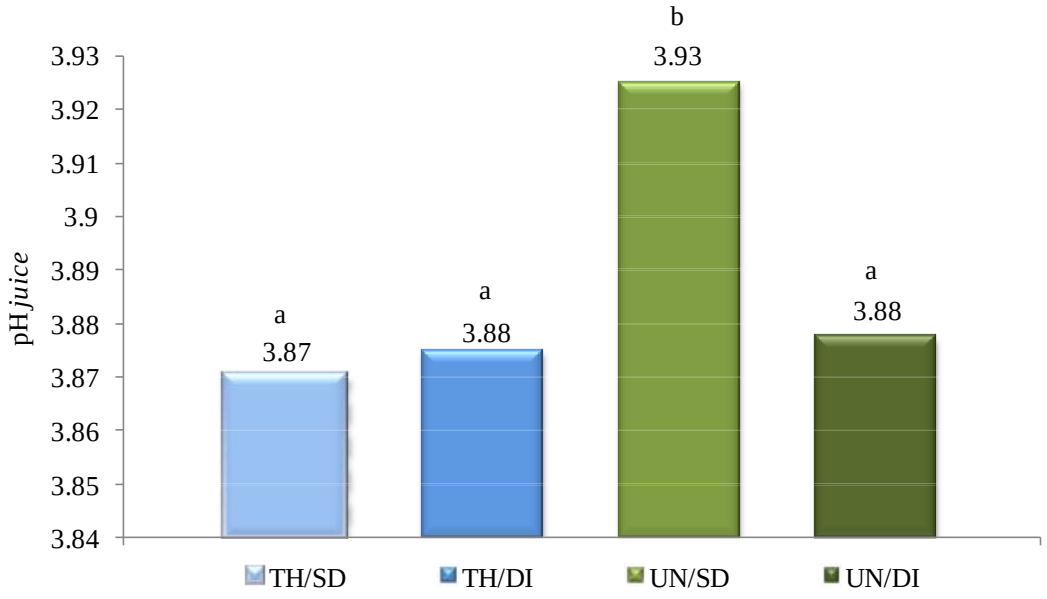


Figure 6.14: The interaction of crop load and irrigation on juice pH in 2006. Columns with different letters differ significantly at $p<0.01$.

Juice TA was highest in the TH in 2006 then lowest in 2007, $p<0.01$ and 0.001, respectively. On average the TH had a higher TA relative to the UN, but only by 0.10 g/L. Kliewer and Weaver (1971) reported that TA was lowest in berries from vines with the least crop load; concomitantly pH was highest in the lowest crop loads.

The DI was consistently higher in TA relative to the SD, although only significant in 2007 and overall averaged only 0.10 g/L higher than the SD. These results conflict with Chapman *et al.* (2005) who reported that TA was highest in the minimal irrigation treatment and lowest in the double irrigation treatment of Cabernet Sauvignon; however, the irrigation methods are most likely not comparable with those of the present experiment. The significant interaction in 2006 demonstrates that the UN/SD and UN/DI were both significantly lower in TA relative to the TH/SD and TH/DI (Figure 6.15). In addition the UN/SD had a significantly lower TA relative to all other treatments in 2006. These data concur with the TA development which consistently

demonstrated that TA was lower in the SD among both crop loads. Once again, although significant differences occurred in TA, some of the differences are not relevant from a practical standpoint—particularly because acid adjustments are commonly made prior to fermentation.

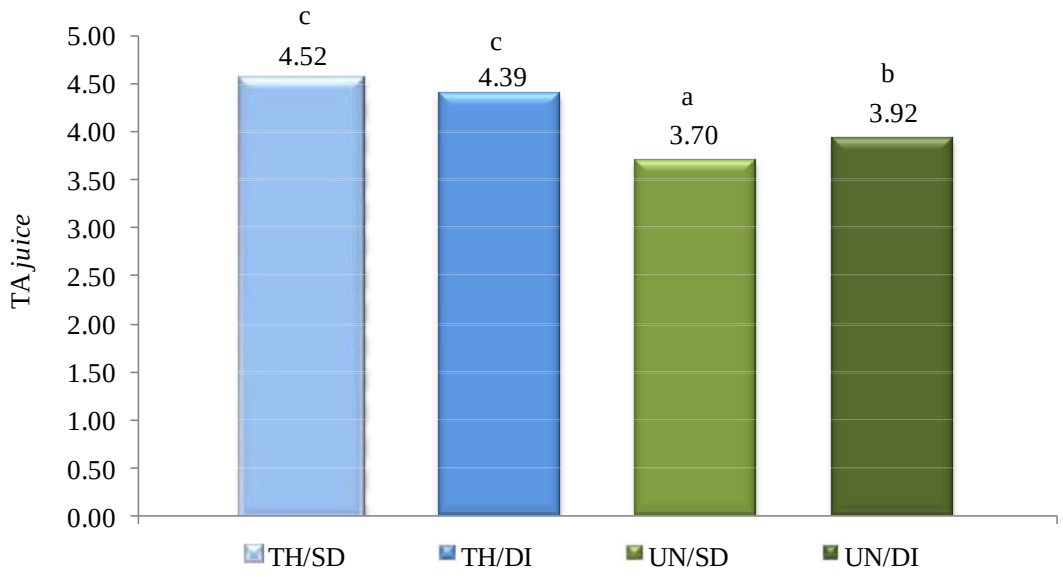


Figure 6.15: The interaction of crop load and irrigation on juice TA in 2006. Columns with different letters differ significantly at $p<0.05$.

YAN: Treatment effects on YAN were minor and only one significant difference occurred. The TH had significantly higher YAN relative to the UN in 2007 ($p<0.05$), but overall the treatments did not have a major impact on YAN. Given that the entire experimental plot received the same pre bloom nitrogen fertilization, it is conceivable that crop thinning impacted on YAN relative to the unthinned. Nitrogen application in the vineyard is known to increase nitrogenous compounds in the associated berries and subsequent must (Bell and Henschke 2005) and combined with the crop reduction may explain why the TH had a significantly greater YAN in 2007. Nitrogen compounds in grapes and wines have been shown to increase in under-cropped vines (Sinton *et*

al. 1978), and for Cabernet Sauvignon vines crop thinned by 1/3 and 2/3 and relative to unthinned vines (Ough and Nagaoka 1984).

Optimal YAN for fermentation ranges between 330 to 530 mg YAN/L depending on yeast strain and sugar content of the must (Jiranek *et al.* 1995a). However, it is generally agreed that adequate fermentation can still proceed at suboptimal rates as low as 150 mg YAN/L (Henschke and Jiranek 1993). Therefore, most treatments in 2007, with the exception of the TH, were suboptimal for fermentation. YAN in 2006 met the optimal range in all treatments and highlights that individual season characteristics can impact YAN.

Wine ethanol: Ethanol content has been identified as a potential cause of stuck or sluggish fermentations (Casey and Ingledew 1985, Boulton *et al.* 1996, Bisson 1999). Additionally, wines sold with greater than 14.0 % alcohol require a higher excise tax in the United States—and thus have financial implications. Interestingly in this experiment, there were no significant treatment effects on ETOH in 2006. In contrast, 2007 was significantly different due to both crop load and irrigation. The UN/DI in 2007 had the most favorable outcome with an ETOH of 13.8 %, indicating that increased irrigation during late ripening can mitigate the negative effects of extended ripening—i.e. high alcohol wines. Additionally, crop load may play an integral role. Due to the conflicting results between seasons, it appears that crop load and irrigation are capable of affecting ETOH, but the magnitude of the effects may depend on seasonal characteristics.

6.4.6 Effects of Crop Load and Irrigation on Wine Color and Phenolics

Overall, total phenols were more affected by irrigation than crop load, although there were differences due to crop load in the second year. In general, the TH x SD irrigation had the

highest total phenols. The interaction in 2007 (Figure 6.16) illustrates that the TH/SD had significantly more total phenols relative to the other treatments. The UN/DI was lowest in total phenols and there were no differences between the TH/DI and UN/SD. Sanchez *et al.* (2006) demonstrated that no differences in total phenols occurred in Merlot vines at two irrigation levels during late ripening, or in Cabernet Sauvignon under similar irrigation in Sonoma (Mendez-Costabel 2007). However, the volume of water applied in terms of ETc was higher than in my experiment and may explain the different outcomes.

It appears that increased irrigation combined with no crop thinning caused a less favorable environment for the development of phenols; however, differences in total phenols were not large in "real terms" even though statistically significant differences were found. Previous research shows that wines made from shaded fruit contain lower total phenols (Joscelyne *et al.* 2007, Ristic *et al.* 2007) and wines made from well exposed fruit contained greater levels of phenolics and color (Mazza *et al.* 1999). The growth data indicated that the DI had significantly greater mean cane weight and pruning weight per vine in 2007 which could have increased shading in the fruiting zone thereby decreasing total phenols within the berries—however the LAI and PAR data does not support this. Alternatively, the SD irrigation initiated a greater water stress on the vine, as indicated by LWP measurements, and perhaps caused basal leaves to senesce earlier thereby allowing more light into the fruiting zone. In addition, water stress has been shown to directly affect vine metabolism and thereby increase berry phenolics—particularly anthocyanins (Roby *et al.* 2004, Bindon *et al.* 2008a, 2008b)

The precise cause of increased phenols remains unresolved, but the SD irrigation did slightly increase phenolics relative to the DI. However, these differences, alone, would not have changed wine quality enough to affect the wine program placement. Wine phenolics are critically

important to the quality of all wines (Peynaud *et al.* 1996) and determining the effects of wine phenolics on wine sensory presents a broad challenge—but continues to warrant further research. In addition, few studies exist which investigate the effects of late season irrigation and/or extended ripening on subsequent wine phenols.

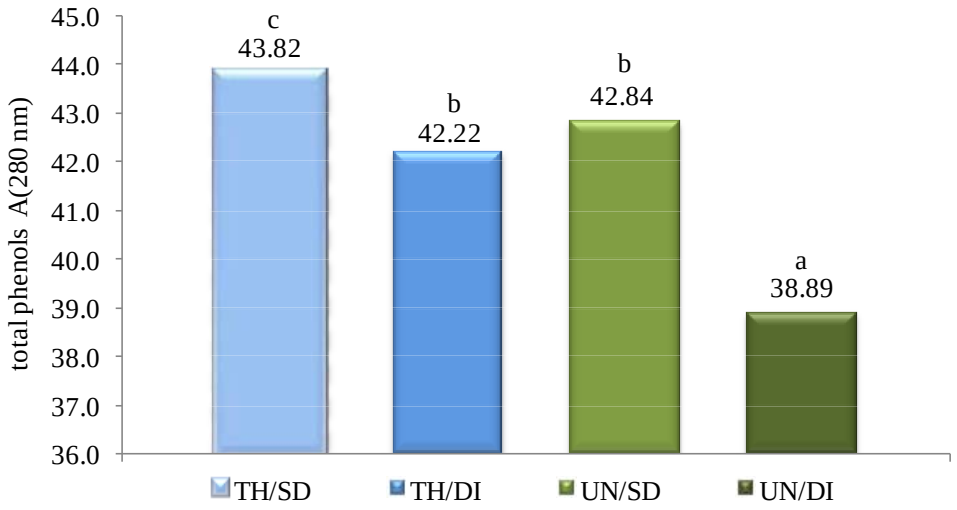


Figure 6.16: The interaction of crop load and irrigation on total phenols in 2007. Columns with different letters differ significantly at $p<0.05$.

The SD irrigation positively affected color density including its components: A(420 nm) and A(520 nm). Color density was highest for the SD irrigation treatment each year and increased CD in the SD relative to the DI, by 11 % on average among the two years. These results conflict with those of Bravdo *et al.* (1985) who reported the highest wine color in Cabernet Sauvignon grapes which received the most frequent irrigations, and with Mendez-Costabel (2007) who showed no effect of increased irrigation during late ripening, on wine color (i.e. anthocyanins or total phenols). My results are more consistent with Intrigliolo and Castel (2008) who reported on irrigated versus non-irrigated Tempranillo and found that irrigation reduced color intensity by 18 % relative to the non irrigated treatment, although large seasonal variations occurred.

Additionally, Freeman and Kliewer (1983) reported reduced wine color in irrigated versus non irrigated vines but speculated that the results may be due to either direct or indirect effects of the irrigation treatments. With regard to crop load, Freeman and Kliewer (1983) also reported that severe pruning, and thus lower yields, had no consistent effect and often reduced wine color. This work supports color density findings in the current study which were 'not significant' due to crop load and had different trends between years.

Previous research supports that wine color parameters can be indirectly affected by other factors such as berry size and perhaps fruit exposure. However, LAI and PAR data in this study suggest that differences were *not* due to cluster microclimate. Ginestar *et al.* (1998) reported an interesting dissimilarity between total phenols and anthocyanins measured on a per berry basis versus per mg/g berry mass. When measured per berry, the 'dry' (lowest irrigation) treatment had the lowest amount of anthocyanins and total phenols. In contrast, the dry treatment was highest in the concentration of anthocyanins and total phenols when expressed as mg/g berry mass and was concluded to be due to smaller berries in the dry treatment and greater shading in the higher irrigation treatments. Certainly berry size was greater in the DI; however, CD was only measured on wine, not berries.

Wine hue was only marginally affected by crop load and/or irrigation and did not have a consistent trend among years. Generally the lowest hue was in the UN/SD irrigation. These results demonstrate that crop load and irrigation did not cause large differences for the A(420 nm) and A(520 nm) absorbance and may explain the lack of consistency between the two years.

6.4.7 Effects of Crop Load and Irrigation on Wine Sensory

The expert ratings consistently demonstrated that the highest quality wines were from the SD irrigation treatments regardless of crop load, and there were no differences due to crop load *per se*. An interaction occurred (Figure 6.17) and was significant in 2006; indicating that the lowest (i.e. best) wine scores were from the UN/SD in each year. The highest (worst) expert scores were consistently from the DI but alternated between the TH and UN from 2006 to 2007, respectively. These results conflict with those of Bravdo *et al.* (1985) who reported the best wine quality scores were for Cabernet Sauvignon grapes which received the most frequent irrigations. The 2007 scores were better overall relative to 2006, yet all treatments (all years) would be designated for the same wine program and price point based solely on the commercial scoring at J. Lohr Winery.

There were no significant differences in the flavor intensity ratings even though there were significant differences for the expert scores. In contrast to these results, Reynolds *et al.* (2007) reported increased flavor intensities, as demonstrated through descriptive analysis attributes, and concluded that irrigation did not have a detrimental effect on sensory attributes. In fact, irrigation increased the intensity of many desirable attributes; however, this study was on Chardonnay in a cool, humid climate and may not be comparable to Cabernet Sauvignon in a hot climate.

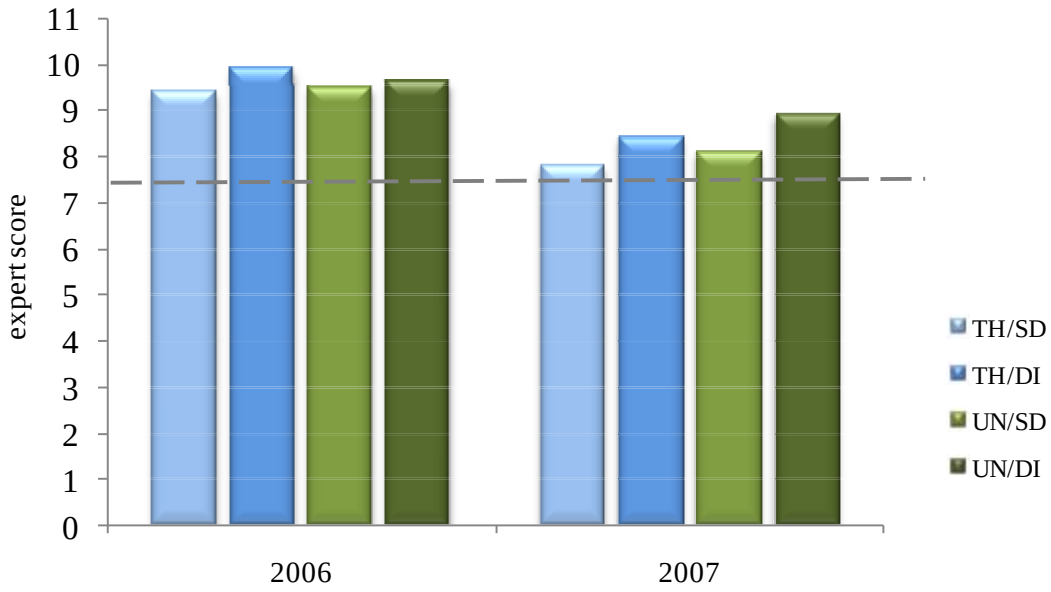


Figure 6.17: The interaction of crop load and irrigation on expert score for two years. Note: grey line represents the minimum score to meet quality expectation for dedicated wine brand. Note: wine quality increases or is more optimal as score decreases.

Results from the difference testing indicated that, overall, differences due to the crop load and irrigation treatments were only significant in one replication. The UN/SD versus UN/DI in replication four (R4) were significantly different from each other. However, after the difference testing, panelists commented that R4 UN/SD had obvious sulfide aromas which were integral in differentiating between the two treatment replications. This may explain why all other treatment replications were 'not significant'. Presumably, the treatments implemented prior to harvest were not the cause of differences detected in resulting wines; rather, the development of sulfides during cellaring caused the differences. Furthermore, the great majority of treatments had fewer than 50 % correct, demonstrating that for all treatment replications, except for R4 UN/SD versus R4 UN/DI, panelists correctly identified differences on less than half of the triangle tests. These results are similar to those of Sanchez *et al.* (2006) who reported that increased irrigation during extended ripening did not have a significant impact on wine sensory properties in their two year

study. Moreover, Mendez-Costabel (2007) showed that trained panelists were unable to identify significant differences in wines made from vines with different irrigation (post veraison) and harvested at four different maturity levels.

In summary, the wine sensory data indicate that although irrigation treatment wines were significantly different in the expert scoring, they were not significantly different in flavor intensity or in the difference testing. Additionally, there were no significant differences or definitive patterns indicating changes in sensory attributes due to crop load. Furthermore, these wines were all designated for the same commercial wine program at J. Lohr Winery, and thus merit the same price point.

6.5 Conclusions

a) Irrigation affected leaf water potential indicating that irrigation can affect the grapevine during the later stages of ripening and through to harvest. This supports previous research that transport of water via the xylem and/or the phloem remains functional in these later stages of ripening.

b) The greatest berry weight was maintained throughout the duration of ripening and at harvest with the increased irrigation (DI); thereby causing a significant increase in yield relative to the SD irrigation.

c) The combination of UN/DI had the highest berry weight.

d) Vine growth data indicated that increased irrigation increased pruning weights and mean cane weight only in 2007; additionally, there were no significant differences in the fruiting

zone light environment or leaf area index. The Y/P ratio was only significantly different due to crop load in 2007.

e) Degrees Brix development was consistently greater for the SD irrigation; in contrast sugar (g)/berry was consistently greater for the DI—particularly in the UN crop load. Degrees Brix at harvest was consistently greater for the SD and lowest for the DI; however, the difference was not substantial enough to consistently lower ETOH in subsequent wines.

f) Overall the DI had a consistently lower pH and higher TA relative to the SD. Crop load had different results between years suggesting that irrigation had the greater effect on pH and TA development within berries and juice at harvest. These differences in TA and pH were not relevant in practical terms.

g) Increased irrigation significantly lowered alcohol in subsequent wines in 2007, but was not significant in 2006.

h) Wine color was significantly affected by irrigation. The SD consistently had higher total phenols and color density relative to the DI. Crop load had no significant affect on wine color density.

i) Irrigation caused significant differences in expert wine scores however, irrigation had little impact on flavor intensity and few differences were detected between treatment wines in the difference testing. All wines, all years were designated for the same commercial wine program and price point.

In summary, there were few detrimental effects on wine quality due to high crop load and increased irrigation. A substantial yield was maintained by the unthinned and increased

irrigation in the later stages of ripening. Therefore, increased irrigation during the late stages of ripening may be a useful tool for mitigating the negative effects of extended ripening. However, careful monitoring is required.

Chapter 7: INTEGRATIVE DISCUSSION

7.1 Introduction to the Experiment

The effects of viticultural practices on wine quality are being constantly debated and investigated. The relationship between crop load and wine quality has been, and continues to be, a prominent issue in viticultural research and farming (Chapman *et al.* 2004, Keller *et al.* 2005) in order to optimize wine quality and economic sustainability. In addition, the definition of 'ripeness'—as it pertains to harvest date decisions—has changed considerably in the past decade. Few experiments have investigated the effects of crop load and extended ripening through to wine quality and sensory analysis—particularly on a commercial scale. The aim of this study was to investigate the interactions of crop load and extended ripening on yield, vine balance, wine and fruit composition, wine quality, sensory analysis and cost benefits. This comprehensive study tested the hypotheses that: (1) wine quality is improved by crop reduction; (2) extended berry ripening increases wine quality; and (3) vineyard economics are negatively affected by extended ripening and crop load adjustment.

The experiment was a 4 x 5 factorial investigating the factors, crop load and target °Brix at harvest. In 2005, 2006 and 2007 a commercial vineyard of clone 8 Cabernet Sauvignon located in Paso Robles, CA was adjusted to four crop levels post fruit set (E-L 31). Each crop level was harvested at five target °Brix levels from 22.5 to 28.5 °Brix and fermented into wine. Yield components, vine growth, wine and fruit composition and wine sensory were measured and assessed on all replicated treatments.

An additional experiment (Experiment 2) was conducted in 2006 and 2007 on the same site to investigate the effects of crop load and late season irrigation on extended ripening. This

experiment was a 2 x 2 factorial including thinned (TH) and unthinned (UN) vines irrigated at two different regimes, standard irrigation (SD) and double the standard (DI) from 22 °Brix to harvest at 27.0 target °Brix. Each treatment was replicated four times. Hypotheses tested were that: (1) increased irrigation in late season increases yield and (2) increased irrigation in late season reduces berry total soluble solids and wine alcohol. Leaf water potential, yield components, fruit and wine composition, and wine sensory data were collected and analyzed each year.

7.2 Effects of Crop Load

Crop thinning significantly reduced overall yield, in terms of both kg/vine and tonnes/ha, relative to the unthinned (control). On average over three years, crop thinning to 20 cl, 40 cl and 60 cl reduced tonnes/ha relative to the control (UN) by 56 %, 25 % and 9 % respectively. The average yield varied each season and was presumed to be associated with amount of winter rainfall.

Thinned grapevines exhibited yield component compensation in addition to shoot growth regulation. There was an inverse relationship between clusters/vine and mean cluster weight, berry weight and berries/cluster. These effects are illustrated in the significant negative linear relationship between clusters per vine and pruning weight (Figure 7.1), and clusters/vine and mean cluster weight (Figure 7.2). Although only data for 2007 are shown, similar relationships existed for all years.

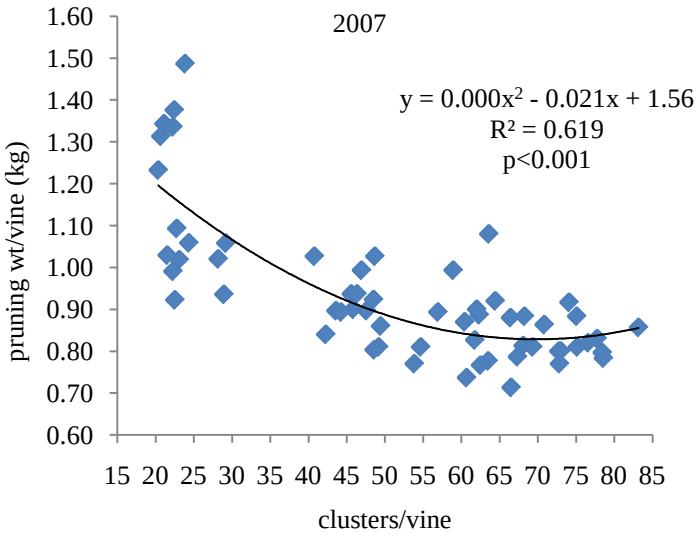


Figure 6.18: Common regression model for the negative linear relationship between clusters/vine and pruning weight/vine in 2007.

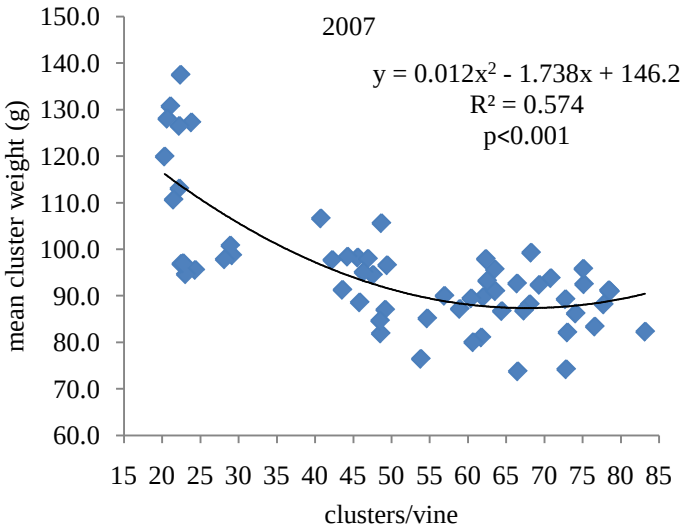


Figure 6.19: Common regression model for the negative linear relationship between clusters/vine and mean cluster weight in 2007.

Crop reduction had detrimental effects on both vine balance and wine quality. Canopy growth was regulated by crop load. Generally, as crop load decreased, canopy growth increased and consequently increased shading of the fruiting zone. The data suggest that both main shoot length and lateral shoot growth increased with crop thinning. Increased lateral shoot growth

most likely contributed to the decreased PAR in the fruiting zone as clusters per vine decreased. Additionally, LAI indicated that leaf area increased as crop load decreased. The negative effects of crop thinning on vine balance were further indicated by the Y/P ratio, which became less optimal as crop load decreased—relative to the UN which was the most optimal each year. Pruning weight differences between the lower crop loads and the UN became increasingly greater as the years progressed. This suggests not only increased partitioning of resources towards vegetative growth rather than reproductive growth due to crop thinning, but also a possible carryover effect each year. The carbohydrate testing did not indicate significant differences; however, a more in-depth study including roots and pre-budbreak sap may have shown differences in carbohydrates due to crop load. Previous studies have indicated that increased yield by way of increased bud number at pruning, significantly decreased leaf area available for producing photosynthates and consequently reduced carbohydrate storage (Miller *et al.* 1993, Edson *et al.* 1995b, Miller and Howell 1998, Weyand and Schultz 2006).

Yeast assimilable nitrogen in juice at harvest clearly demonstrated that the 20 cl consistently had the highest YAN relative to all other crop loads. Additionally, the lowest YAN was consistently for the 60 cl or UN. These findings are supported by Kliewer and Ough (1970) who reported that concentrations of the nitrogenous compounds—arginine, proline, total free amino acids and total nitrogen in berry juice—greatly increased with crop reduction. Furthermore, YAN was positively correlated with increased leaf area per vine ($p<0.01$). Most likely, increased vegetative growth in the lower crop loads consequently increased photosynthetic capacity of the vines and caused a greater translocation of amino acids and ammonium into the fruit. The consequences of reduced YAN due to extended ripening and/or crop load, has implications for

increased nutrient additions of di-ammonium phosphate (DAP) and Superfood™ to supply yeast with adequate nutrition to complete primary fermentation.

Crop load significantly affected wine color density and anthocyanins, and the highest levels of both were found in wines made from unthinned vines. Furthermore, the lowest levels were with the 20 cl. Concomitantly, the greatest light interception in the cluster zone (PAR) was in the UN. Therefore, changes in the light environment within the fruiting zone most likely contributed to wine color. Light is known to be an important factor in anthocyanin synthesis (Mullins *et al.* 1992) and shading experiments on Cabernet Sauvignon have indicated light intensity affects anthocyanins and total phenols (Smart *et al.* 1985b, Morrison and Noble 1990, Dokoozlian and Kliewer 1995) and is necessary for color formation in grapes. However, more recent work by Mori *et al.* (2007) showed decreased anthocyanin accumulation in exposed bunches which reached high temperatures near or greater than 30 °C. Other studies have shown that high bunch exposure reduced anthocyanins (Mahbrouk and Sinoquet 1998, Haselgrove *et al.* 2000, Bergqvist *et al.* 2001, Spayd *et al.* 2002). Furthermore, light and temperature appear to have synergistic effects on anthocyanins (Tarara *et al.* 2008). In my experiment, higher crop loads had lower shoot vigor and consequently increased cluster exposure of the UN— which most likely increased wine color and anthocyanins relative to the other treatments. Cortell *et al.* (2007) reported increased anthocyanins with lower vigor sites of Pinot Noir and concluded that it was primarily due to shading differences in the fruiting zone microclimate. Moreover, both studies highlight the important relationship between shoot vigor and clusters per vine. An important consideration is that overexposure can be detrimental to wine color, particularly in the warm areas of Paso Robles if excessive berry temperature is reached. Therefore, the balance between

fruit exposure and berry temperature is critical to optimize final wine color in the Paso Robles region.

Crop thinning increased the rate of sugar accumulation; however, all crop loads achieved maturity greater than 26 °Brix. Although the rate of sugar accumulation and maximum sugar levels varied somewhat between seasons, overall the photosynthetic capacity of all crop loads was adequate for ripening and demonstrates that crop reduction is not always necessary to promote ripening to sugar maturities near or beyond 26.0 °Brix.

In this project (specifically Experiment 1), wines made from unthinned grapevines, which averaged 16.8 t/ha over three years, had the highest color density and wine quality scores by the expert panel.

In addition, the higher crop loads adequately achieved the desirable flavor profile—based on descriptive analysis (DA)—and, in some years, at moderate ripening levels (i.e. 25.5 °Brix target). For example, treatment 20 cl/25.5 °Brix consistently correlated with the non desirable flavor attributes: vegetative and astringency. This was in contrast to the higher crop loads (particularly 60 cl and UN) which were more correlated with the attributes berry, body, duration and color depth, at the 25.5 °Brix target. Initial levels of isobutylmethoxypyrazine (IBMP) increased as crop load decreased. Furthermore, the intensity of the 'vegetative' attribute was a main factor determining wine sensory and quality—as indicated by the expert wine scores and DA. Presumably, the lower levels of IBMP and vegetative score for the higher crop loads positively influenced wine quality scores compared with 20 cl and 40 cl. Interestingly, total phenols and tannins were generally lowest in the UN and highest in the 20 cl. The collective results of wine composition and sensory suggest that, although tannins and total phenols were

higher for the 20 cl, it did not necessarily enhance wine quality. It is likely that the higher total phenols for the 20 cl was driven by increased tannins or other non-color related phenolic compounds, given that color density was consistently highest for the UN. Descriptive analysis results demonstrated that astringency was rated highest overall for the 20 cl. Astringency and bitterness in wines are generally associated with polyphenols and tannins (Noble 1999). My study suggests that the relationship between high shoot vigor with reduced clusters per vine may have influenced trends in tannins and total phenolics, and is recommended for investigation in future research.

Concomitantly, the results from this study emphasize that yield *per se* is not a good indicator of wine quality. Rather, it is the environment and management that affects yield and thus influences wine quality (P.R. Dry 2008, personal communication). The collective data clearly showed that the most optimal wines came from higher crop loads and in contrast the least optimal wines came from lowest crop loads. Furthermore, these results indicate that crop reduction in this experiment was not justified for the improvement of wine quality. In addition, the losses due to crop thinning were far more substantial than those due to extended ripening, even though both were significant.

Therefore, the hypothesis that wine quality is improved by crop reduction must be rejected.

7.3 Effects of Extended Ripening

Extended ripening caused a significant yield loss due to berry weight reduction—presumably due to berry dehydration. On average for all years, extended ripening to the 28.5 °Brix target reduced yield (kg/vine) by 14 %. As expected, °Brix increased as days after veraison increased. Sugar (g)/berry results suggest that, in some cases, increased °Brix was more a consequence of a

concentration effect within the berry due to dehydration rather than transport of sugar to berries. Evidence of a concentration effect was more apparent for the 20 cl than the UN, and suggests differences in the hydraulic connection between berries and shoots for the different crop loads. This may have been due to changes in the rate of ripening in response to crop load. Previous research supports this notion (Bondada *et al.* 2005, Keller *et al.* 2006, Tillbrook and Tyerman 2008, Mendez-Costabel 2007).

Titratable acidity decreased and pH increased as °Brix at harvest increased. In addition, wine ethanol was positively correlated with °Brix at harvest—as °Brix at harvest increased, ethanol in subsequent wines increased. There were no 'stuck' fermentations. Both yeast assimilable nitrogen (YAN) and isobutylmethoxypyrazines (IBMP) decreased significantly as °Brix at harvest increased. In addition, LAI decreased and PAR in the fruiting zone increased with extended ripening. Most likely, the reduction of IBMP as °Brix at harvest increased was related to increased 'time on the vine' for the degradation of IBMP—IBMP is known to breakdown in the presence of light (Hashizume and Samuta 1999).

All sensory testing indicated that extended ripening improved wine quality. Generally, the descriptive analysis indicated that treatments harvested at higher target °Brix (i.e. 25.5 or greater) had contrasting attributes to those from lower °Brix targets (i.e. 22.5-24.0 °Brix). For example, the higher °Brix target wines were more correlated to a 'desirable' flavor profile, e.g. in terms of berry aroma and flavor, body and duration, than the lower target °Brix wines which correlated with less desirable attributes such as vegetative aroma and flavor, astringency and acid. Furthermore, the best wine scores from the expert panel were for wines harvested at target °Brix between 27.0 and 28.5. Additionally, wine quality was positively correlated with the attributes

flavor intensity and body, and negatively correlated with vegetative intensity. This outcome was also consistent with results from both the descriptive analysis and the scaled attribute ratings. It is likely that the increased 'body' attribute in treatments harvested at higher °Brix targets was due to higher ethanol in those wines.

Although negative consequences of extended ripening such as yield loss and high alcohol wines were evident in this experiment, generally wine quality was improved with extended ripening. Therefore, the hypothesis that extended berry ripening increases wine quality can be accepted.

7.4 The Interaction of Crop Load and Extended Ripening

The practice of extended ripening improved wine quality overall, particularly for the lowest crop load which required the most extended ripening (in terms of °Brix at harvest) in order to achieve a desirable flavor profile and sufficient wine score. In contrast, the highest crop load (UN) had the most optimal vine balance, color density, wine score and flavor profile and the lowest % berry weight loss while requiring the most days after veraison to reach °Brix targets. Overall, this study demonstrated that extended ripening is a good remediation tool for unbalanced vines; however, the quality potential will always be greater for vines with optimal balance. Extended ripening lends to a certain flavor profile and style of Cabernet Sauvignon. However, this style may not be the most preferred by non-American consumers. Results from this experiment emphasize that optimal vine balance can be achieved with unthinned vines—in the Paso Robles environment—and highlights the importance of vineyard site characteristics and the grapevine's ability to self-regulate.

Experiment 2, on the effects of increased irrigation late season on crop load and extended ripening, demonstrated that irrigation is a useful tool to mitigate some of the negative effects of

extended ripening; however, careful monitoring is required. Certainly, the double irrigation maintained the greatest berry weight and thus increased yield overall relative to the standard irrigation. However, differences in °Brix at harvest were not significant. The double irrigation had more sugar per berry even though °Brix was reduced. This suggests that °Brix is more determined by a concentration effect than by sugar transport and should be considered if using °Brix, exclusively, as the ripeness measurement for harvest decisions. Although wine quality was improved with extended ripening to the 27.0-28.5 °Brix targets, acceptable wine quality may be achieved near 26.0 °Brix—depending on overall vine balance and seasonal effects.

7.5 Economic Evaluation

The data from previous chapters showed significant yield losses as a result of both crop thinning and extended ripening, which results in financial losses for growers paid on weight. In 7.5.1 and 7.5.2, two models are presented which show potential gross margin based on tonnes/ha. The grower perspective (7.5.1) addresses differences based on both crop load and extended ripening. The winery perspective (7.5.2) evaluates the differences in number of bottles produced and hence gross margin at each crop load, and is specific to a winery which grows its own grapes. Vineyard and winery input costs and price/tonne of Cabernet Sauvignon are based on the average production costs and grape pricing at J. Lohr Vineyards and Wines for 2005-2007—and specific to the designated wine program. Note that although a fixed vineyard input cost is used here; generally, labor cost would be higher with increased cluster thinning per vine.

7.5.1 Grower Perspective Model

Two grower perspective models are presented in Tables 7.1 and 7.2. Gross margin calculations were developed using the following assumptions and calculation: the average price/tonne (t) in 2005-2007 was $1,375.00/t, vineyard input costs were $6,200.00/hectare (ha), dollars ($) represent US dollars. Gross Margin ($) = (t/ha) x ($1,375.00/t) – (6,200.00 $/ha).

Table 7.1: Economic analysis of crop load if grower harvested at peak berry weight (between 24.0-25.5 °Brix targets)

Crop load	Yield (t/ha)	Income ($/ha)*	Variable costs ($/ha)	Gross margin ($/ha)**
20	8.0	11,000.00	-6,200.00	**$4,800.00**
40	13.0	17,875.00	-6,200.00	**$11,675.00**
60	17.0	23,375.00	-6,200.00	**$17,175.00**
UN	18.0	24,750.00	-6,200.00	**$18,550.00**

*assumes grape price = $1375.00/t; ** assumes variable costs = $6200.00/ha

Table 7.2: Economic analysis of crop load with extended ripening to 28.5 °Brix target.

Crop load	Yield t/ha	Income ($/ha)*	Variable costs ($/ha)	Gross margin ($/ha)**
20	7.0	9,625.00	-6,200.00	**$3,425.00**
40	12.0	16,500.00	-6,200.00	**$10,300.00**
60	14.0	19,250.00	-6,200.00	**$13,050.00**
UN	16.0	22,000.00	-6,200.00	**$15,800.00**

*assumes grape price = $1375.00/t; ** assumes variable costs = $6200.00/ha

7.5.2 Winery Perspective Model

The winery perspective model is presented in Tables 7.3 and 7.4. The following calculations and assumptions were used to obtain the gross margin for the winery perspective model.

Assumptions**:** 716 Liters (L)/tonne (t), production cost/L = $3.20/L x 0.75 L/bottle = $2.40/ bottle, wholesale bottle price = $12.00/bottle.

Bottles/ha = (t/ha) x (716 L/t) = (L/ha) ÷ (0.75 L/bottle)

Production cost = ($2.40/bottle) x (bottles/ha)

Gross margin = [(bottles/ha) x ($12.00/bottle)] – (production cost)

Note: t/ha for each crop load is the overall average yield (t/ha) for the individual crop loads at all °Brix targets.

Table 7.3: Economic analysis of crop load and extended ripening treatments from a winery perspective.

Crop load	Yield (t/ha)	bottles/ha*	$/bottle	production cost ($)**	Gross margin ($/ha)
20	8.0	7,637.00	12.00	-18,329.00	**$73,315.00**
40	12.0	11,456.00	12.00	-27,494.00	**$109,978.00**
60	14.0	13,365.00	12.00	-32,076.00	**$128,304.00**
UN	16.0	15,275.00	12.00	-36,660.00	**$146,640.00**

* assumes 716 L/t = 7,637 bottles/ha, **assumed production cost $3.20/L = $2.40/bottle

Table 7.4: Economic analysis of crop load and extended ripening treatments from a winery perspective if grapes were harvested at peak berry weight.

Crop load	Yield (t/ha)	bottles/ha*	$/bottle	production cost ($)**	Gross margin ($/ha)
20	8.0	7637	12.00	-18,329.00	**$73,315.00**
40	13.0	12411	12.00	-29,786.00	**$119,146.00**
60	17.0	16229	12.00	-38,950.00	**$155,798.00**
UN	18.0	17184	12.00	-41,242.00	**$164,966.00**

* assumes 716 L/t = 7,637 bottles/ha, **assumed production cost $3.20/L = $2.40/bottle

Both the grower and winery models demonstrate that significant loss occurs with both crop thinning and extended ripening. Grower gross margin was reduced by $13,750.00/ha with crop reduction to 20 cl/vine relative to the UN when fruit was harvested at peak berry weight.

Extended ripening reduced tonnes/ha for all crop loads; however, the gross margin differences solely due to extended ripening were not as great.

When analyzed as gross margin based on bottles/ha produced, the 20 cl had substantially lower gross margins relative to all other crop loads. Based on the sensory data—particularly the expert scores—all crop loads at 27.0-28.5 target °Brix were designated for the same wine program and price point. Moreover, crop thinning did not improve wine quality enough to increase bottle price or price per tonne. Therefore, the gross margin reduction due to crop thinning was not offset by any significant gain in quality.

While extended ripening to the 28.5 °Brix target improved quality, yield was reduced. Furthermore, the quality gained was not sufficient enough to elevate any treatment wines into the ultra premium wine program which would command a higher bottle price or price per tonne.

The hypothesis that vineyard economics are negatively affected by both extended ripening and crop load adjustment can be accepted.

7.5.3 Economic Analysis: Experiment 2

Results from experiment 2 showed that the increased irrigation treatment (DI) significantly increased yield. The following analysis indicates that this irrigation strategy was cost-beneficial. Pumping cost—including diesel fuel and pump maintenance—was assumed at $8.50/irrigation hour, and the average pumping costs are presented in Table 7.5. The 600 gallon per minute (gpm) pump supplies 20.2 ha @ 1 gallon/hour/vine (equivalent to 2271.2 L per minute @ 3.79 L/hour/vine). All dollar values are in US dollar ($). The pumping cost/ha was calculated as: pumping cost ($/ha) = total irrigated hours x 8.50/hour ÷ 20.2 ha. Figure 7.6 shows the gross

margin for the UN at both irrigation treatments. Note: the TH/SD and TH/DI averaged the same t/ha for 2006 and 2007, and therefore was not included in the economic analysis.

Table 7.5: Experiment 2 pumping costs for two different irrigation treatments in 2006 and 2007.

Irrigation treatment	Total irrigated hours (22.0 °Brix to harvest)	Average pumping cost
SD (2006)	115	48.40
SD (2007)	94	39.60
SD Average		**$44.00**
DI (2006)	230	96.80
DI (2007)	200	84.20
DI Average		**$90.50 ($91.00)**

*Assumes pumping cost 8.50/ irrigated hour/20.2 ha

Table 7.6: Gross margin of the unthinned crop load at two different irrigation treatments.

Treatment	Yield (t/ha)	Income ($/t)	Variable input costs ($/ha)	Pumping costs ($/ha)	Gross Margin ($/ha)
UN/SD	11.9	16,362.50	-6,200.00	-44.00	**$10,119.00**
UN/DI	13.8	18,975.00	-6,200.00	-91.00	**$12,684.00**

The sensory tests on Experiment 2 indicated that all treatments were designated for the same wine program and price point due to no or few significant differences in the expert scores and difference testing. Hence, there was no wine quality gain due to crop thinning. In addition, the UN/DI had a profit gain of $2,565.00/ha relative to UN/SD and establishes that the UN/DI was the most economical treatment. Degrees Brix was lower with increased irrigation, although the reduction in wine alcohol was significant only in 2007. Additionally, crop load and season interacted with the magnitude of irrigation effects on wine alcohol and must be considered. In conclusion, the hypotheses that (1) increased irrigation in late season increases yield and (2)

increased irrigation in late season reduces berry total soluble solids and wine alcohol can be accepted.

7.6 Recommendations to the Industry and Further Research

Results from this study aid in the advancement of industry knowledge on both crop load and ripeness. A replicated experiment on crop load and extended ripening to levels greater than 24.0 °Brix had not been previously conducted—to my knowledge. Therefore, the results reported here are novel. Future research on both crop load and ripening will undoubtedly follow and could ultimately add to the improvement of viticultural effects on wine quality. This research is unique in its scale and may be the only scientific experiment where vineyard field replications were fermented at this volume, replicated, and utilized to assess multiple facets of wine quality. Recommendations to the wine industry and future research are as follows.

Crop Load Management: Results from this study indicate that yield alone is a poor indication of wine quality, and highlights that yield reduction is neither necessary nor beneficial for all vineyard sites. Furthermore, vineyard terroir and seasonal characteristics are paramount in affecting overall wine quality; however, vineyard management practices provide a tool set for altering wine quality, both positively or negatively.

Canopy Management: Managing vine vigor in Cabernet Sauvignon by obtaining the most optimal crop load for the site should be a major focus for wine grape growers in order to improve the balance between vegetative and reproductive growth. In addition, optimizing the light environment in the fruiting zone is recommended. Annual yield/pruning ratio assessment and other canopy assessments are recommended to evaluate and refine management practices. Improved measurements of vine balance should be pursued in future research. This may be

achieved through refinement of the optimal Y/P range for key varieties and specific to certain site characteristics. For example, Cabernet Sauvignon grown in the Paso Robles region may require more shading of the fruit to protect against high berry temperature than in cooler regions such as Bordeaux, France. Therefore, the optimal Y/P value may differ—in favor of slightly more leaf area, hence increased pruning weight. The use of additional tools to assess canopy microclimate in the fruit zone such as LAI, PAR, cluster temperature, leaf temperature is also recommended to improve the definition of vine balance. Finally, studies which link vine balance parameters and wine quality would greatly add to the overall definition and indices of vine balance.

Extended Ripening: Extended ripening is a useful tool for increasing wine quality for specific styles of Cabernet Sauvignon and proved to be a good remediation tool for out-of-balance grapevines in this case. However, the best wine quality will always be achieved from vines which have the most optimal vine balance regardless of extended ripening. Extended ripening significantly improved wine color; however, the exact mechanism remains unknown. It is speculated that increased ethanol increased extractability of color and flavor compounds and/or an additional stage of ripening exists. My work in addition to Rogiers *et al.* (2006) and Kennedy (2007) postulates the idea of an additional stage of berry development in which concentration of flavor compounds and metabolites occurs. Continued research on the late stages of ripening—i.e. >24.0 °Brix or after peak berry weight is achieved—is recommended to investigate the mechanism by which the changes in wine color and flavor compounds occur, and to better define the later stages of berry development.

Defining Ripeness: The definition of ‘ripeness’, including ‘flavor ripeness’, merits continued attention and clarification. The decision of when fruit is ‘enologically ripe’ and ready for harvest

will vary based on site, stylistic goals and consumer expectations of the product. This study provides a scientific basis for extended ripening; however, defining when a vineyard reaches flavor ripeness without objective indices continues to be a challenge to the industry. Therefore, a time efficient yet objective measure of flavor ripeness is needed for industry use. Research which utilizes the development of rapid analytical tools such as the electronic nose (portable GCMS) or rapid spectral methods—which define and customize flavor profiles for key varieties—will aid in closing the gap on subjective indicators of flavor ripeness. My study provides limited data on the complex yet important relationship between tannins and ripening. Therefore, future research on this relationship and its role in consumer acceptability should be investigated.

Water Relations and Ripening: A significant interaction between crop load and ripening exists and has prompted several areas for further investigation. Crop load affected the rate of ripening and the point at which peak berry weight was achieved, and the amount of berry weight loss thereafter. It is hypothesized that the water relations between the berry and shoot, and perhaps soil moisture depletion, affected this outcome. Continued investigation on this interaction and its effect on fruit and wine composition is recommended.

Increased irrigation during late ripening can mitigate some of the negative effects of extended ripening. However, it is speculated that 'too much' irrigation could negatively influence wine color and flavor intensity; therefore, the practice requires careful monitoring. The threshold of actual irrigation amounts will vary with season. As the trend for extended ripening continues, investigation of both practical and cost-beneficial methods for mitigating the negative effects of extended ripening should be pursued. My results in addition to those previously reported by Bondada (2005), Sanchez *et al.* (2006), Keller *et al.* (2006), Mendez-Costabel (2007), Tillbrook

and Tyerman (2008) greatly add to the body of knowledge on irrigation effects and water relations within the grapevine during late ripening. However, continued research on this topic—including detailed vine physiology measurements—will improve the understanding of hydraulic relations between the berry and vine throughout ripening.

Management of Methoxypyrazines: Vegetative flavors, primarily due to methoxypyrazines, have a significant effect on the flavor profile and quality perceptions of Cabernet Sauvignon. Based on previous research and results from this study, the management of methoxypyrazines is tightly linked with vine vigor, including the role of the light environment within the fruiting zone—affecting both the accumulation and breakdown of methoxypyrazines. Previous research by Roujou de Boubee *et al.* (2002) and Ryona *et al.* (2008), support that pre-veraison canopy management is more influential than post-veraison on methoxypyrazine accumulation. However, my research showed that extended ripening will aid in IBMP reduction due to more time on the vine for IMBP degradation. Moreover, adequate management of vine vigor with tools such as crop load, irrigation and harvest date is crucial to final wine quality.

Fruit Exposure: This study in addition to published research suggests that optimal light and temperature levels are very specific to site characteristics and variety. Fruit exposure including the relationship between light and temperature on cluster microclimate warrants continued research in order to better understand its influence on both wine color and flavor development. A similar experiment should be conducted on a site with contrasting site characteristics such as temperature and soil water holding capacity.

Sensory Analysis and Wine Quality: Sensory analysis proved to be an effective way of analyzing wine quality, particularly when compared with instrumental data on wine color and

flavor compounds (e.g. IBMP). It is highly recommend that vineyard research should follow experiments through to sensory analysis of wines to assess wine quality. Continued studies on the sensory attributes of primary importance to consumers and how to achieve the desired sensory profiles through viticultural practices and harvest decisions should be pursued. There were dissimilarities between sensory data from an exclusively American panel and a mainly Australian panel in my research. Therefore, a study which investigates consumer acceptability utilizing panelists from different countries may reveal the effects of product familiarity and drivers of overall wine preference.

Economic Considerations: Although extended ripening reduced yield and thereby gross margin, wine quality was improved. Therefore, economic losses due to extended ripening may be justified for some growers and wineries. A suggested compromise is a quality bonus based on color for growers to ease frustration about yield/financial losses. Conversely, the decision to implement crop thinning requires careful consideration. Crop thinning did not improve wine quality and hence incurred a large economic loss from both the grower and winery perspective. I recommend that this research should be conducted on a site with lower maximum daily temperatures during the growing season, lower vine vigor, lower autumn temperatures—and specific to the variety Cabernet Sauvignon—to investigate whether wine quality is improved with extended ripening, crop thinning or both. Lastly, cultural practices such as crop thinning and/or extended ripening are only a secondary means to alter wine quality—therefore growers and winemakers must be mindful of the ultimate quality potential of a site and assurance of economic sustainability.

APPENDIX 1: BERRY COLLECTION PROTOCOL USED FOR ALL BERRY SAMPLES WITHIN BOTH THE CROP LOAD X EXTENDED RIPENING AND CROP LOAD X IRRIGATION EXPERIMENTS.

General guidelines:

- Double check to make sure label on bag matches label on map and field tags.
- Begin sampling on vine designated by the specific vine frequency per crop load treatment (stated below). Alternate at each stop between the north and south side of the vine. i.e. if you start on the south side, the next sample should be taken off the north side. Alternate this pattern until end of row.
- Never sample an end vine.
- Do not sample from a damaged or abnormal vine or cluster.
- Pluck 15-20 berries alternating between top, middle and bottom areas of different clusters within vines in close vicinity to that stop.
- Be sure to sample berries from all sides of the cluster with no bias.
- Place completed bag in cooler and move on.
- Goal: approximately 200 grams of berries/sample, 60 samples/experiment

***Specific treatment guidelines*:**

- **20 cl/vine** treatments (152 vines/ row)
  Frequency = Stop every 10 vines to collect berries.

- **40 cl/vine** treatments (75 vines/row)
  Frequency = Stop every 6 vines.

- **60 cl/vine and UN** treatments (50 vines/row)
  Frequency = Stop every 4 vines.

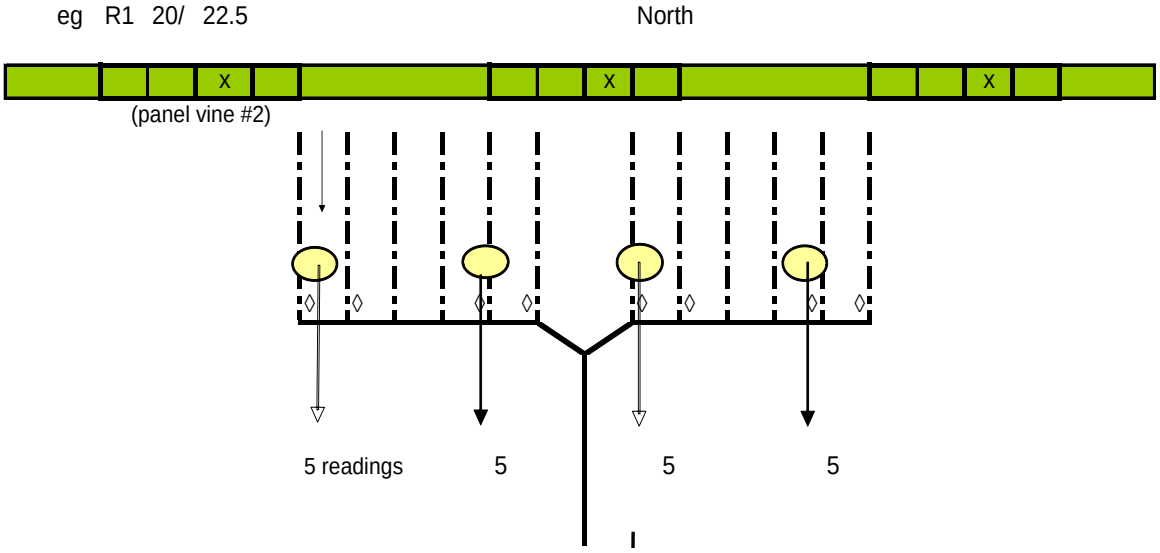
APPENDIX 2: CEPTOMETER PROTOCOL FOR LAI AND PAR MEASUREMENTS

Measurement hours: 11:30am-1:30pm

Phenological stages: Veraison (identified by 100 % veraison) and harvest (taken 1 day before actual harvest for logistical simplicity)

Sampling sites: "Panel vines"

- 3 panel sites per rep of CL x °Brix treatment
- Measure Vine #2 (#1 is the most easterly vine, *select another vine if abnormal*)
- Measure 4 positions on vine # 2 (between spur 1, 2 and between end spur and second to end spur)
- Take 5 readings in full sun (above canopy light) always keep level by using level on light bar
- Place light bar just above fruiting zone (to intercept light that would hit fruit below)
- Take 5 measurements at each of the four positions/vine
- Move on to next panel set and repeat.



For Veraison sampling, begin with Rep 1 until all CL x °Brix treatments are complete, and then proceed to Rep 2 and so on.

Explanation of light meter calculation and theory:

Leaf Area Index is an algorithm calculated from: above and below canopy PAR, zenith angle, fractional beam measurement value and leaf area distribution parameter. In simple terms this algorithm factors in cloud cover, angle to the sun, soil or leaf reflective light and leaf area distribution of certain crops (respectively).

Theory for the LAI algorithm is based on the Norman Jarvis Model, Norman and Jarvis (1975) and Goudriaan (1988) and is discussed in the AccuPar LP-80 operator's manual (section PAR and LAI Theory) or these related published papers (Norman 1979, Campbell 1986).

APPENDIX 3: GRAPEVINE PETIOLE AND BLADE NUTRITION COLLECTED AT BLOOM 2008

BLADES

Treatment	Nitrate Nitrogen ppm	% Total Nitrogen	% Total Phosphorus	% Potassium	% Calcium	% Magnesium	% Sodium	Boron ppm	Zinc ppm	Manganese ppm	Iron ppm	Copper ppm
20/22.5	22	2.87	0.76	0.96	2.40	0.67	0.03	131	52	203	181	246
20/28.5	26	1.15	0.75	0.97	2.25	0.68	0.04	117	55	193	109	317
40/22.5	18	3.64	0.65	0.94	2.11	0.63	0.03	108	57	200	135	252
40/28.5	19	3.53	0.60	0.96	2.17	0.69	0.03	120	54	162	146	267
60/22.5	23	3.36	0.66	1.15	2.58	0.80	0.02	142	91	179	405	304
60/28.5	22	3.42	0.59	0.89	2.14	0.63	0.02	113	50	153	311	191
UN/22.5	20	2.99	0.54	0.86	1.99	0.65	0.03	101	56	143	112	274
UN/28.5	24	2.25	0.65	1.04	2.37	0.72	0.03	126	54	169	158	333

PETIOLES

Treatment	Nitrate Nitrogen ppm	% Total Nitrogen	% Total Phosphorus	% Potassium	% Calcium	% Magnesium	% Sodium	Boron ppm	Zinc ppm	Manganese ppm	Iron ppm	Copper ppm
20/22.5	261	0.92	1.05	2.46	1.91	1.26	0.02	54	24	76	88	31
20/28.5	203	0.89	1.11	2.30	1.95	1.43	0.04	57	32	78	71	39
40/22.5	378	0.92	1.08	2.46	1.64	1.27	0.02	51	65	85	77	45
40/28.5	212	0.91	0.97	2.37	1.77	1.37	0.05	54	23	66	76	36
60/22.5	261	0.91	1.00	2.64	1.84	1.36	0.30	56	23	68	77	38
60/28.5	253	0.90	0.89	2.29	1.69	1.31	0.03	48	21	60	96	27
UN/22.5	277	0.89	0.93	2.36	1.75	1.37	0.02	51	21	60	88	31

APPENDIX 4: NUTRIENT ADDITION SCHEDULE USED FOR ALL EXPERIMENT FERMENTATIONS

Fermentation stages:

Stage 1 (day 1): At yeast inoculation

Stage 2 (day 2): Fermentation is fully active and °Brix has dropped 2 or 3 degrees

Stage 3 (day 3): Before reaching 10 °Brix

Addition amounts given in pounds/1000 gallons: 1 lb/1000 gal= 120 ppm = .12 g/L = 12 g/hL

Nutrient additions based off 1000 lbs (454 kg) of fruit i.e (322 L)

YAN <100	Cerevate (g)	Superfood (g)	DAP (g)
Stage 1	3.2	77	77
Stage 2		39	77

YAN 100-150	Cerevate (g)	Superfood (g)	DAP (g)
Stage 1	1.6	77	39
Stage 2		39	39

YAN 150-200	Cerevate (g)	Superfood (g)	DAP (g)
Stage 1	1.6	77	0
Stage 2		39	39

YAN 200-250	Cerevate (g)	Superfood (g)	DAP (g)
Stage 1		39	19
Stage 2		39	19

YAN 250-300	Cerevate (g)	Superfood (g)	DAP (g)
Stage 1		39	0
Stage 2		19	0

Superfood™ is made by Fermentis, a Division of S.I. Lesaffre group. It is a complete yeast nutrient used to stimulate fermentation, improve yeast survival and reduce stuck fermentations. Ingredients include: yeast cell walls (hulls), diammonium phosphate, primary yeast grown extract, minerals, thiamine and other vitamins.
DAP is diammonium phosphate
Cerevate vitamix is a powdered mix added to promote yeast survival during fermentation. Cerevate generally contains important vitamins for yeast such as thiamine and minerals.

APPENDIX 5: PROCEDURE FOR TITRATABLE ACIDITY OF MUST

Standardize NaOH once prior to beginning TA procedure and use that standardization for the final calculation pertinent to data for that day with the following procedure:

1. Add 10 mL of 0.1N HCL to a 50 mL beaker. Add several drops of phenolphthalein indicator.
2. Titrate with 0.01N NaOH until faint pink endpoint is achieved. Record the volume of 0.1N NaOH used in the titration. Calculate the normality of NaOH using the following formula:

$$\text{N NaOH} = \frac{(10\text{ mL})(0.1\text{N HCL})}{\text{mL NaOH used}}$$

3. Note: the first acceptable titration flask was set aside and used as a visual reference for acceptable titration endpoint based on light pink color for the following titrations that day.
4. Add several drops of phenolphthalein indicator to 100 mL of DI water in beaker.
5. Pipet 5.0 mL of clean, filtered must sample to beaker.
6. Titrate with 0.1N NaOH until faint pink endpoint is achieved. Record the volume of 0.1N NaOH used in the titration.
7. Repeat three times per sample and average the three measurements for the final TA calculation.
8. Calculate TA using the following formula:

$$\text{TA (g/L)} = \frac{(\text{mL NaOH})(\text{N NaOH})(75)}{\text{mL of sample (i.e. 5.0 mL)}}$$

APPENDIX 6: WINEMAKING PROTOCOL

1. **Harvesting**
   - Fruit was hand harvested—at consistent temperatures ideally (65°F) to keep consistent beginning fermentation temperature. Harvest start time with crews was scheduled at the best time to optimize arrival temperature. Generally harvests became later in the day as Fall/Autumn progressed and morning temperatures became cooler.
   - All 'material other than grapes (MOG) were removed
   - All bins of fruit were balanced to equal weight in order to assure the same volume/mass for fermentation.
   - Fruit was to arrive at 70 ºF ± 5 ºF (i.e. 21.3 ºC)
   - Fruit was delivered to outside F2 for processing
   - If fruit was above 70 ºF (21.3 ºC) bins were to be placed inside F2 cellar to cool. If fruit was below 21.3 ºC it was processed outside when temperature warmed up. Adjusting the harvest time to the ideal temperature of the day to deliver desired fruit temperature generally alleviated this scenario.
   - The plastic Macro Bins used weighed 41.7 kg (92 pounds) and at full capacity could hold just over a half ton of grapes (i.e between 900-950 kg).

2. **Crush/De-stem**
   - Bins were tipped into crusher
   - Crusher was manually fed for efficient throughput
   - Receiving bin was placed under the crusher-destemmer and the corresponding label with the treatment ID was taped on to all four sides.
   - Colorpro enzyme addition of 33 mL/tonne was added at the crusher
   - SO_2 was added at 30 ppm

3. **Analysis**
   - A 500 mL juice sample was taken from each bin
   - pH, TA, Temperature, °Brix and YAN were measured on each bin's juice sample. Specifically, there was a separate analysis for each replication R1, R2, R3 of each treatment e.g. 20/22.5
   - pH, TA were re-measured the day after the acid addition on each individual bin.

4. **Additions (after crush/destem)**
   - Acid was adjusted to 7 g/L a few hours after crushing and after TA analysis.
   - 30 ppm SO_2 were added and mixed into to each bin fermentation
   - Yeast Lallemand ICV-D254 was used @ 300 ppm. This was added to the bin fermentation aproximately 3-4 hours after crushing. The yeast addition followed a standard hydrating procedure. The yeast was then added to the bins by pouring evenly across the top and lightly mixed (plunged) into must with a punch down tool.

5. **Daily Pump overs**
   - Pump overs were conducted twice daily using an air pump and funnel shaped mesh screen. Two minutes was the amount of time needed to adequately pump the

underlying juice/wine onto the cap and submerge the cap. This was strictly done for 2 minutes twice a day except for day 7 (pressing) day to keep consistency in the amount of color extraction due to cap management among all treatments. Furthermore, a pump over rather than punch down was conducted because it is the commercial standard for Cabernet Sauvignon at J. Lohr Winery and it was the most consistent method for equal cap management among treatments. No punch downs were conducted due to the inconsistency in force and pressure between persons doing the punch down—even if for a given time limit.

Daily Schedule consisted of the following: Note: a strict 7 day interval was kept for each fermentation to maintain the same amount of 'time-on-the-skins' or extraction time. Thereby, all treatments were pressed on day 7 even if they were not completely dry.

Day 1 (First day that grapes are crushed – i.e day of harvest)

- SO_2 and enzyme (color pro) added at crusher into receiving bin.
- Measure temperature and °Brix, using densiometer and thermometer. Plunge strainer and beaker into underlying juice. Fill beaker completely and conduct measurement immediately.
- Acid addition, to be mixed in with plunger for one minute maximum. All additions mixed in 10 times its volume of juice.
- Yeast to be rehydrated using standard commercial protocol. Yeast mixture to be spread across top of bin (after additions have been mixed in) and lightly plunged.
- Record daily temperatures and °Brix on all bins
- Place lids on bins.

Day 2 Measure temperature and °Brix at 1 pm

- Conduct morning (am) pump over using screen and air pump for 2 minutes.
- Re-check pH and TA
- Add nutrients according to nutrient schedule
- Further acid addition made if necessary
- Afternoon (pm) pump over using screen and air pump for 2 minutes

Day 3 Measure temperature and °Brix

- am pump over for 2 minutes
- Addition of nutrients according to nutrient schedule
- pm pump over for 2 minutes

Day 4 Measure temperature and °Brix 1 pm

- am pump over for 2 minutes
- pm pump over for 2 minutes

Day 5 Measure temperature and °Brix 1 pm

- am pump over for 2 minutes
- pm pump over for 2 minutes

Day 6 Measure temperature and °Brix 1 pm

- am pump over for 2 minutes
- pm pump over for 2 minutes

Day 7 Drain and Press (to 2 bars) also see press procedure in chapter 4.

- 200 mL free run for final chemistry (include 2, 50 mL tubes for phenolic analysis, label appropriately)
- Press until sufficient wine to fill 1 barrel and 1, 18.90 L (5 gallon) carboy
- Barrel to be neutral oak (identical cooperage of World Cooperage Medium toast, no toasted heads)
- Add Malolactic (ML) culture *Hansen viniflora*

Post Primary Fermentation

- ML monitoring using paper chromatography and enzymatics
- Note: ML was generally complete in 8-10 weeks

Post Malolactic Fermentation

- Barrels were racked to another barrel (same cooperage, year, toast, etc.)
- 25 ppm SO_2 was added.
- SO_2 was monitored/adjusted to 30 ppm free
- Barrels were topped using corresponding 18.90 L carboy (5 gallon) that were set aside at pressing.

Racking and Storage

- Barrels were topped after 1 week and SO_2 adjusted
- Identical wine was used for topping (amount was measured)
- Wine was topped and SO_2 adjusted again after 2 months.

Additions Notes: Enzyme and SO_2 added at crusher

- Acid always adjusted to 7 g/L
- Super Food 77 g, Cerevate 1.6 g, DAP 39 g *

* Amount calculated for 1000 lbs (454 kg) of fruit based on nutrient schedule.

APPENDIX 7: SPECTROPHOTOMETER SAMPLE PREPARATION AND PROCEDURE FOR TOTAL PHENOLS AND COLOR DENSITY

General Notes

- Samples were collected and set aside at pressing and post malolactic fermentation—as stated in the other related procedures. A 'sample' consisted of 50 mL of wine made from the 60 treatment replications e.g R1 20/22.5, R2 20/22.5, R3 20/22.5 and so on. Each sample was run in quadruplicate on the spectrophotometer, (i.e. Absorbance was measured on four tubes per sample) and these four spectrophotometer readings were averaged to make one data point per treatment replication.
- Shimadzu UV-1700 pharma spec was used for all samples in all years.
- Pathlength is 5 so each sample's Absorbance result was multiplied by 2 to meet the standard 10 pathlength.

Total Phenols Absorbance (280 nm) and Color Density Absorbance (420 nm + 520 nm):

- Absorbance (280 nm) measurements on the spectrophotometer consisted of 250 µL into 5 mL of deionized (DI) water (H_2O); degrees of freedom (DF) = 20; 19:1 (i.e. 4.75 mL or 4750 µL of DI H_2O, and 0.25 mL or 250 µL sample).
  Calculation= spectrophotometer reading of Absorbance (280 nm) x 20 x 2 = Total Phenols for sample
- Absorbance (420 nm) measurements on the spectrophotometer consisted of 500 µL in 5 mL of DI H_2O; DF = 10; 9:1 (4.5 mL or 4500 µL DI H_2O, 0.5 mL or 500 µL sample).
  Calculation= spectrophotometer reading of Absorbance (420 nm) x 10 x 2 = Absorbance at (420 nm) for the wine sample.
- Absorbance (520 nm) measurements on the spectrophotometer consisted of 500 µL in 5 mL of DI H_2O; DF = 10; 9:1 (4.5 mL or 4500 µL DI H_2O, 0.5 mL or 500 µL sample).
  Calculation= spectrophotometer reading of Absorbance (420 nm) x 10 x 2 = Absorbance at (520 nm) for the wine sample.
- Calculation for Color Density was: Absorbance (420 nm) + Absorbance (520 nm) = Color Density

Note: *only* Press samples were centrifuged for 10 minutes, speed 8 (2500-3000 revolutions per minute) on Beckman TJ6 Centrifuge

APPENDIX 8: NOPA AND AMMONIUM PROCEDURE TO ACQUIRE YAN

NOPA procedure:

1. Set spectrophotometer to Absorbance (335.0 nm)
2. Set up cuvettes: 2 blanks, OPA/NO-OPA, 5 isoleucine standards OPA, (# samples x 2) for A_1 NO-OPA and A_2 OPA.
3. Calculate and make OPA/NO-OPA buffers.
4. Centrifuge juice samples in 1.5 mL eppendorf tubes for 3 minutes @ maximum revolutions per minute
5. Read absorbance for NO-OPA samples A_1
6. Zero spec with NO-OPA blank = 3 mL NO-OPA Buffer + 50 µL DI H_2O
7. Record A_1 NO-OPA = 3 mL NO-OPA buffer + 25 µL DI H_2O + 25 µL juice samples.
8. Read ABS for isoleucine standards and OPA samples A_2.
9. Zero spec with OPA blank = 3 mL OPA buffer + 50 µL DI H_2O
10. Make isoleucine standards and OPA samples
11. Have 10 minutes total when OPA mixes with isoleucine or juice. Start with standards:
    i. 10 µL DI H_2O e.g. abs = 0.68
    ii. 20 µL DI H_2O e.g. abs = 0.922
    iii. 30 µL DI H_2O e.g. abs = 1.114
    iv. 40 µL DI H_2O e.g. abs = 1.297
    v. 50 µL DI H_2O e.g. abs = 1.528
12. Prepare A2 samples.
13. 25 µL DI H_2O + 25 µL juice
14. Start timer for 10 minutes and add 3 mL if OPA Buffer to standards and A_2
15. Read ABS for A_2 at 10 minutes
16. Enter ABS results into excel
17. Run Ammonia Ion test on samples and A-E isoleucine standards.
18. Enter then combine with NOPA to get YAN results. i.e. NOPA + Ammonia Ion = YAN

APPENDIX 9: PREPARATION OF SAMPLES FOR FOSS WINE SCAN™ MODE

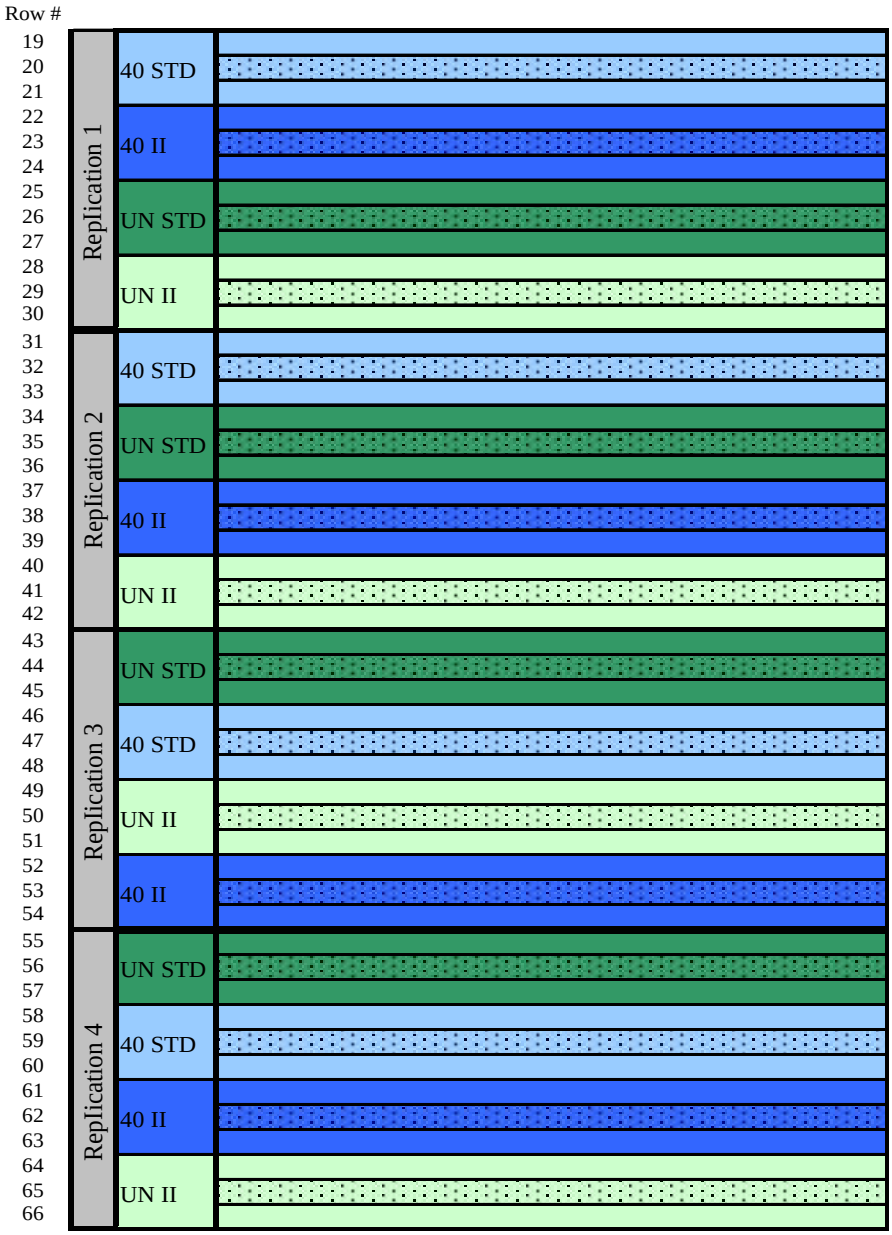
The FOSS Wine Scan™ was used for general wine and juice analysis such as titratable acidity of wine free and total SO_2 and ethanol. The FOSS Wine Scan™ uses near infrared spectroscopy to measure chemical properties of wine and yields a rapid analytical solution. Calibrations are included in the FOSS Wine Scan™ to assure accuracy. The sample preparation procedure for all FOSS Wine Scan™ analysis was as follows.

Note: All samples to be filtered and centrifuged before run on the FOSS in order to avoid internally damaging the analyzer.

1. Rinse side arm flask with the sample before filtering.
2. Centrifuge for 10 minutes at speed 8, 3000 revolutions per minute (rpm) and filter.
3. Set up filter apparatus with pre-filters on bottom and 47 mm filter on top. Secure with clamp.
4. Attach tube from vacuum pump to the side arm of flask.
5. Pour sample in and power in the pump.
6. After sample has filtered through, fill FOSS cup to top and place rubber lid on. Arrange cups accordingly on white FOSS sample rack. Sample is ready for analysis.
7. Read and record corresponding analysis from printed report.

APPENDIX 10: SCHEMATIC MAP OF THE RESEARCH SITE FOR THE CROP LOAD AND LATE SEASON IRRIGATION EXPERIMENT (EXPERIMENT 2)

The patterned rows (in middle of each treatment replication) represent actual rows for data collection.



APPENDIX 11: IRRIGATION SCHEDULE FOR THE CROP LOAD AND INCREASED IRRIGATION DURING LATE STAGES OF RIPENING EXPERIMENT.

Irrigation schedule for crop load x increased irrigation late season experiment for the 2006 and 2007 season. All ET and Kc values are based on VSP 8' spacing and 7' x 8' vine by row spacing.

Month	Week of	degree days	Kc	ETo (in)	ETc (in)	ETc (ga/vn)	UN SD	UN DI	40 SD	40 DI
	14-20	**2417**	0.59	**1.51**	0.89	31	8	16	8	16
	21-27	**2540**	0.60	**1.56**	0.94	33	16	32	16	32
Sep-06	28-3	**2677**	0.60	**1.56**	0.94	33	14	28	14	28
	4-10	**2799**	0.60	**1.41**	0.85	30	9	18	9	18
	11-17	**2915**	0.60	**1.36**	0.82	28	12	24	12	24
	18-24	**3009**	0.60	**1.24**	0.74	26	6	12	6	12
Oct-06	25-1	**3088**	0.60	**0.92**	0.55	19	6	12	6	12
	2-8	**3153**	0.60	**0.87**	0.52	18	10	20	10	20
	9-15	**3199**	0.60	**0.51**	0.31	11	8	16	8	16
	16-22	**3269**	0.60	**0.85**	0.51	18	10	20	10	20
	23-29	**3339**	0.60	**0.75**	0.45	16	8	16	8	16
Nov-06	30-5	**3408**	0.60	**0.66**	0.40	14	8	16	8	16
Total				**13**	**7.9**	**277.0**	115	230	115	230
% Etc							**43%**	**86%**	**43%**	**86%**

Month	Week of	degree days	Kc	ETo (In)	ETc (in)	ETc (ga/vn)	UN SD	UN DI	40 SD	40 DI
	27-2	**2690**	0.61	**1.66**	1.01	35	10	20	10	20
Sep-07	3-9	**2822**	0.61	**1.38**	0.84	29	20	40	20	40
	10-16	**2932**	0.61	**1.24**	0.76	26	6	12	6	12
	17-23	**3002**	0.61	**0.94**	0.57	20	6	12	6	12
	24-30	**3094**	0.61	**1.16**	0.71	25	12	24	12	24
Oct-07	1-7	**3169**	0.61	**1.06**	0.65	23	6	12	6	12
	8-14	**3230**	0.61	**0.78**	0.48	17	12	24	12	24
	15-21	**3297**	0.61	**0.84**	0.51	18	0	0	0	0
	22-28	**3379**	0.61	**0.84**	0.51	18	10	20	10	20
Nov-07	29-4	**3447**	0.61	**0.66**	0.40	14	6	36	6	36
	5-10	**3490**	0.61	**0.51**	0.31	11	6	36	6	36
Total					**3.9**	**236.0**	94	236	94	236
% Etc							**40%**	**80%**	**40%**	**80%**

Note: The vast majority of annual rainfall in the Paso Robles area is during the winter and early spring months (November-March)—therefore the soil moisture content in this experiment—was only affected by irrigation during late ripening (i.e. September-October). The irrigation schedule was based on weekly ETo and ETc which generally became less each week as fall/autumn approached. Additionally the SD irrigation regime was consistent with the standard practice of the grower based on the grower's historical records and was thereby considered an adequate control.

APPENDIX 12: SAMPLE TASTING SHEET FOR DIFFERENCE TESTING (TRIANGLE TEST)

J. Lohr Vineyards&Wines

Triangle Tasting

07PR 12 RH -01

Panelist:_____

2 samples are the same. One is different.
Place a mark in the square that corresponds to the sample that you think is different.

flight 1-1:

121	130	254
665	213	204
365	102	541
339	854	474

flight 1-2:

121	130	254
474	339	854
541	102	365
665	213	204

flight 1-3:

204	339	213
474	854	254
541	130	121
102	365	665

APPENDIX 13: SCORING SHEET FOR DIFFERENCE SCREENING DONE PRIOR TO BLENDING IN 2007

Sensory Difference Screening

Date: Treatment: / lot: E- stage: post ML

COLOR		no difference				extremely different
	R1	1	2	3	4	5
	R2	1	2	3	4	5
	R3	1	2	3	4	5
	comment:					
AROMA		no difference				extremely different
	R1	1	2	3	4	5
	R2	1	2	3	4	5
	R3	1	2	3	4	5
	comment:					
OFF or FAULT?		yes	no	comment:		
FLAVOR Profile		no difference				extremely different
	R1	1	2	3	4	5
	R2	1	2	3	4	5
	R3	1	2	3	4	5
	comment:					
FLAVOR Intenstiy		no difference				extremely different
	R1	1	2	3	4	5
	R2	1	2	3	4	5
	R3	1	2	3	4	5
	comment:					
	BLEND:	YES	NO	other:		

APPENDIX 14: ASSAY FOR STARCH AND TOTAL SOLUBLE SOLIDS USED BY THE GU LAB

Plant material was collected in the field and immediately shipped to the Gu laboratory at Fresno State University, Fresno, Ca

Sample preparation:

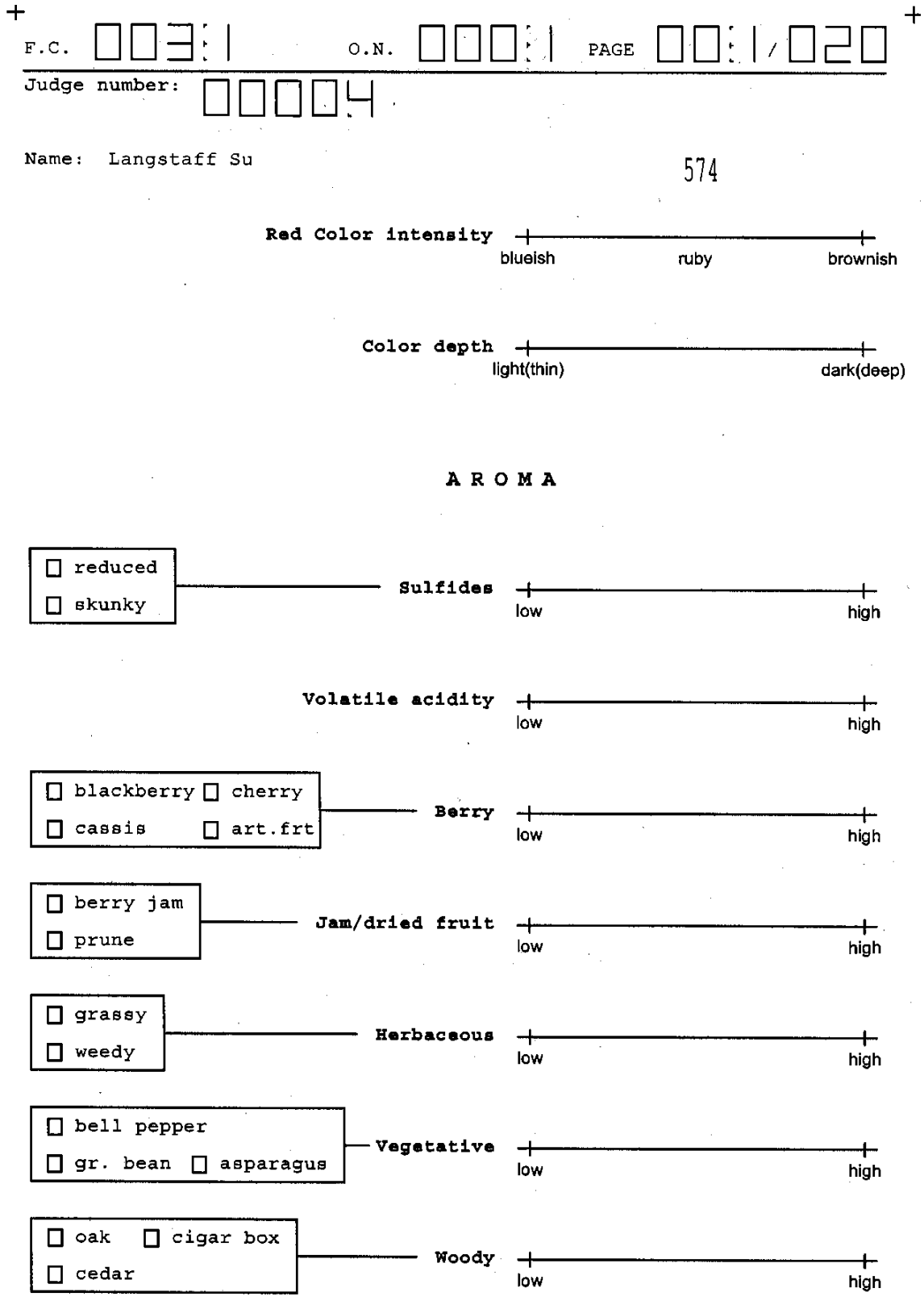
Place plant material in -65 °C freezer until frozen solid (~2d), remove from freezer, place in oven at ~70 °C until dry (~48 hrs), remove node pieces from the dried material and cut the remaining internode section into small discs.

Using a sample mill with a 40-mesh screen grind samples. Once all samples were ground they were placed into small labeled envelopes and placed into a drying oven at 100 °C for 12+ hours to ensure dryness.

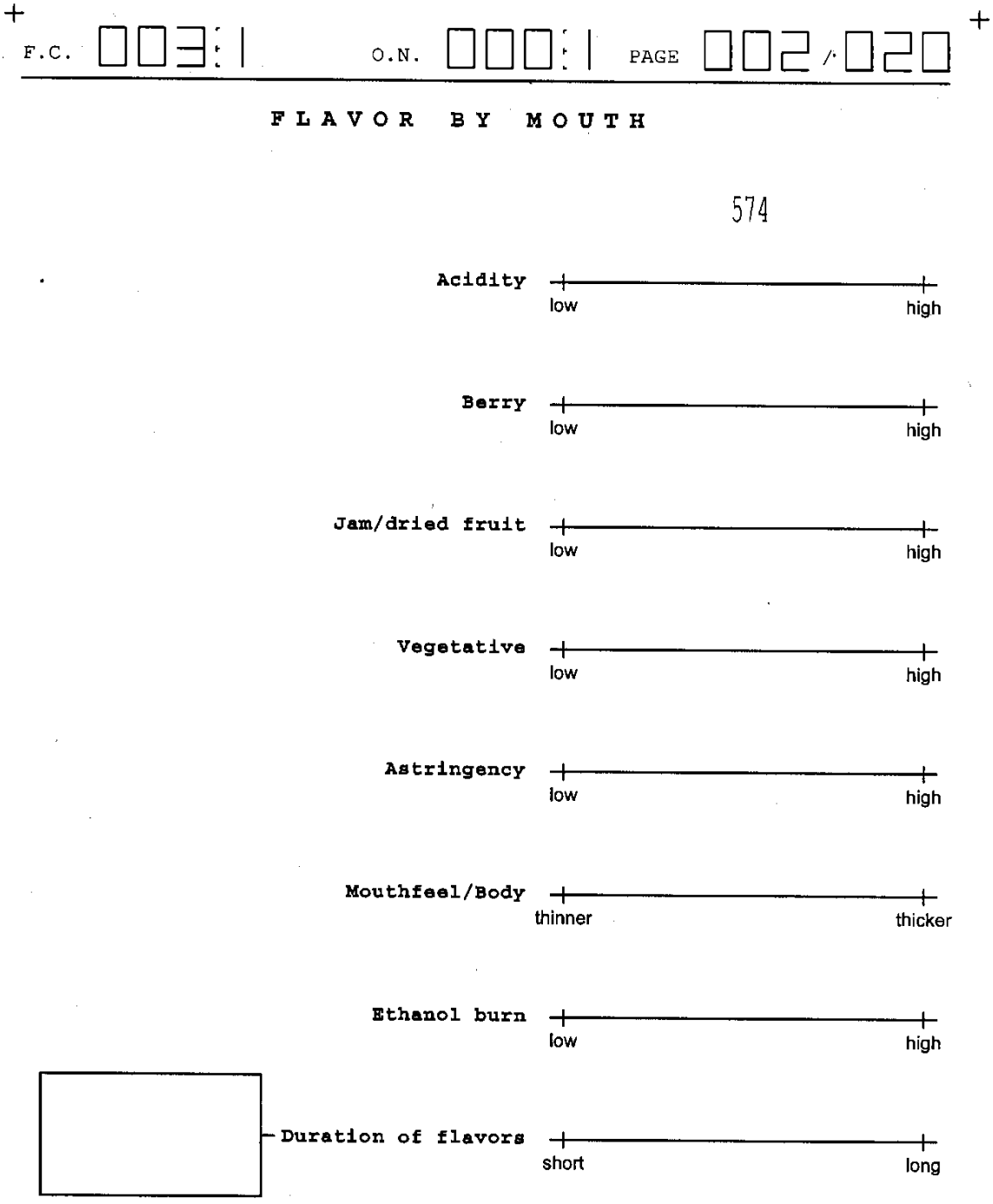
Samples were individually removed from the oven and immediately weighed out to 0.5 g and placed in a sample pouch made from No.1 Whatman.

The remainder of this procedure followed the procedure detailed in Rose *et al.* (1991) p. 8-9 (Perchloric Acid Method 1 Immersion).

APPENDIX 15: SENSORY SCORE SHEET FOR DESCRIPTIVE ANALYSIS AROMAS



APPENDIX 16: SENSORY SCORE SHEET FOR DESCRIPTIVE ANALYSIS FLAVOR BY MOUTH



APPENDIX 17: SENSORY AROMA DESCRIPTORS AND REFERENCE MATERIALS

Sensory aroma descriptors and reference materials

Descriptor	Reference
Sulfides-reduced	none
Sulfides-skunky	none
Volatile acidity	Acetic acid and ethyl acetate
Blackberry	Crushed blackberry
Cassis	Ribena (no wine)
Cherry	Capri Sun wild cherry drink
Artificial fruit	Kool Aid "Hawaiian tropic" mix
Berry jam	Blackberry, plum and boysenberry preserves (no wine)
Prune	Torn pieces of prune (no wine)
Grassy	Torn blades of green grass
Weedy	none
Bell pepper	Small pieces of bell pepper
Green bean	Brine from canned green beans
Asparagus	Brine from canned asparagus
Oak	Piece of toasted oak
Cedar	Piece of cedar wood
Cigar box	Pieces of cigar tobacco + cedar

*most sensory aroma descriptors were submerged in Carlo Rossi Cabernet Sauvignon wine

APPENDIX 18: HARVEST DATES FOR EACH TREATMENT 2005-2007

	Harvest Dates		
Treatment	2005	2006	2007
20/22.5	9/2/05	9/7/06	8/23/2007
20/24.0	9/19/05	9/11/06	8/31/2007
20/25.5	10/1/05	9/20/06	9/5/2007
20/27.0	10/10/05	10/12/06	9/26/2007
20/28.5	10/31/05	10/26/06	10/19/2007
40/22.5	9/14/05	9/8/06	8/29/2007
40/24.0	9/24/05	9/18/06	9/5/2007
40/25.5	10/6/05	9/30/06	9/17/2007
40/27.0	10/14/05	10/20/06	10/8/2007
40/28.5	11/15/05	11/1/06	10/29/2007
60/22.5	9/29/05	9/13/06	8/31/2007
60/24.0	10/7/05	9/22/06	9/10/2007
60/25.5	10/14/05	10/9/06	9/27/2007
60/27.0	11/1/05	10/27/06	10/22/2007
60/28.5	11/16/05	11/6/06	11/5/2007
UN/22.5	10/3/05	9/15/06	9/3/2007
UN/24.0	10/13/05	9/29/06	9/13/2007
UN/25.5	10/21/05	10/16/06	10/1/2007
UN/27.0	11/15/05	11/1/06	10/25/2007
UN/28.5	11/17/05	11/8/06	11/8/2007

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