THE EFFECT OF DEFICIT IRRIGATION ON WATER RELATIONS, GROWTH, AND FRUIT QUALITY OF'BRAEBURN' APPLES (*Malus domestica* BORKH.) GROWING IN LYSIMETERS

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"Om Sai Sri Sai Jaya Jaya Sai"

Dedicated to my grandpa

ABSTRACT

This project investigated the feasibility and practicality of using deficit irrigation (DI) at different times of the growing season on water relations, growth and fruit quality of 'Braeburn' apples grown in lysimeters. Five-year-old trees on MM. 106 rootstock were subjected to three irrigation treatments in a completely randomised design. The treatments were: Well-watered control (C), deficit irrigated for the entire season (ED), and deficit irrigated late in the season (LD) from 102 days after full bloom (DAFB) to harvest.

Both ED and LD trees developed a lower predawn and midday leaf water potential than C trees. For LD and ED trees towards the end of growing season, reduction occurred in the photosynthesis (Pn), stomatal conductance (g_s), and the rate of transpiration. The reduction in Pn was caused by stomatal and non-stomatal factors. Deficit irrigation caused an increase in canopy temperature (T_c) and canopy-air temperature difference (T_c-T_a) in ED and LD. Fruit growth was not affected by DI although shoot growth and increase in trunk circumference were significantly reduced under DI. Deficit irrigation also reduced mean fruit weight at harvest as well as return bloom.

Deficit irrigation increased the concentration of fruit soluble solids and volatiles, decreased that of N, and did not have any effects on the concentration of P, Ca^{2+}, Mg^{2+}, and K^{+}. The ED and LD treatments resulted in more advanced fruit maturity based on higher ethylene production and TSS concentration. Firmness was higher in LD and ED fruit than the C fruit after 12 weeks of storage at 1 ^{0}C.

This study showed that water deficit late in the season may be used in apple production with improved fruit quality in terms of increased TSS, firmness in storage, and higher volatiles without adversely affecting on fruit size.

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'Truth lives for ever so as God'

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ABBREVIATIONS

A	- Surface area of the fruit (m^2)
ABA	- Absicic acid
ACC	- 1-aminocycloprpane-1-carboxylic acid
ANOVA	- Analysis of variance
C	- Control
CD	- Crop density (grams of fruit per unit trunk cross sectional area)
C_a	- External CO_2 concentration ($\mu mol\ mol^{-1}$)
C_i	- Intercellular CO_2 concentration ($\mu mol\ mol^{-1}$)
CRD	- Complete randomised design
DAFB	- Days after full bloom
DI	- Deficit irrigation
ED	- Entire-season deficit irrigation
ET	- Evapotranspiration
GLC	- Gas liquid chromatography
GLM	- General linear models
g_s	- Stomatal conductance ($mol\ CO_2\ m^{-2}\ s^{-1}$)
H	- Hue angle (^{0})
HPLC	- High performance liquid chromatography
IR	- Infra red
L	- Lightness (%)
LD	- Late-season deficit irrigation
MPa	- Mega Pascal (1 MPa = 10 bars)
n	- Number of observations
Pn	- Rate of photosyntheasis ($\mu mol\ CO_2\ m^{-2}\ s^{-1}$)
SEM	- Standard error of the mean
T	- Rate of transpiration ($mmol\ m^{-2}\ s^{-1}$)

TA	- Titratable acidity (% malic acid)
Ta	- Air temperature (^{0}C)
Tc	- Canopy-air temperature (^{0}C)
T_c-T_a	- Canopy-air temperature difference (^{0}C)
TCA	- Trunk cross-sectional area
TDR	- Time domain reflectometry
TSS	- Total soluble solids
VPD	- Vapour pressure deficit
θ	- Soil volumetric water content ($m^3\ m^{-3}$)
Ψ	- Leaf water potential

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Figure 12. Carbon dioxide production for control (C), entire-season deficit (ED), and late-season deficit (LD) treatments. Vertical bars represent the pooled standard errors of means based on four replicates per treatment.

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CHAPTER ONE

INTRODUCTION

Water constitutes one of the most important constraints to increased food production worldwide. Apple (*Malus domestica* Borkh.) production is important in many regions of the world with varied climatic conditions. The apple has a diverse climatic adaptation which makes it the most widely planted fruit of the temperate zone, including Southeastern Europe, Southwestern Siberia, Central Asia and North America (Westwood, 1978, p.1). In some of the regions where the apple is grown, water is often limiting. Dwindling available water resources create problems not only for the general public and governmental agencies, but also for scientists working on water research and crop production (Csizinski, 1993). In some countries or states, there is powerful legislation to control water use and discharge, and growers will need to adhere to rules concerning levels of water use and contamination of ground and surface water with nitrates and pesticides (Kabashima, 1993).

One solution to the problem of dwindling water supply in the face of growing demand is the use of regulated deficit irrigation (RDI). Considerable work has been done on the effect of regulated deficit irrigation (RDI) on the yield and vegetative growth of fruit crops. The term RDI is normally used to denote DI of trees early in the season, before rapid fruit growth starts. Here I will refer to all deficit irrigation treatments as DI and specify the timing of the deficit irrigation in each instance. In a study on peaches, Chalmers et al. (1981) found that high tree density with low rate of water application increased yield. They concluded that water stress if induced at the right time, limited shoot growth but stimulated subsequent fruit growth. Thus vegetative vigour of the tree was suppressed in favour of fruit growth. In subsequent paper, Chalmers et al. (1986) suggested that the mechanism by which RDI reduces vegetative growth but increases fruit yield may be based on the differences in

sensitivity to water stress between different tissues and organs. In this case, shoot growth was more sensitive to water stress than fruit growth and therefore the former was suppressed without any adverse effect on the latter. Similar conclusions were reached by Irving and Drost (1987) and Proebsting et al. (1984). Thus an additional benefit of using DI is the control of vegetative growth and hence a reduction in pruning costs. In this thesis, responses of vegetative and reproductive growth of apple trees to withholding irrigation at different times of the growing season are discussed.

This thesis also explores the effect of reduced irrigation on photosynthesis, stomatal conductance, canopy temperature and canopy-air temperature difference. The use of canopy temperature and canopy-air temperature difference as a measure of plant water status has hitherto been largely confined to field crops with very little application in deciduous fruit crops (Jackson, 1982).

Although DI in some fruit crops has shown benefits in terms of reduced vegetative growth and improved fruit quality, there is limited information regarding its effects on fruit quality. If the practice is to be used successfully as a management tool, there is need to quantify its effects not only on fruit quality at harvest but also on the postharvest behaviour of the fruit. Some of the important postharvest quality attributes for which literature is nonexistent include the effect of preharvest irrigation amount on postharvest aroma and the flavour of the fruit.

Given the limited work done on the effects of reduced irrigation on fruit quality, this study was conducted to gain further understanding on the effect of DI at different times of season on the quality of 'Braeburn' apples.

This thesis evaluates the impact of late and entire-season water deficit on the water relations, growth and fruit quality of apples. The cultivar 'Braeburn' was selected due to its importance as a major export cultivar in the New Zealand export industry.

CHAPTER TWO

LITERATURE REVIEW

Water is one of the most valuable natural resources to increased food production worldwide. Minimising water use leads to a reduced level of nutrients and pesticides leaching into the ground water. Hence the demand for more efficient fruit production has encouraged the search for an improved understanding of plant water requirements. Deficit irrigation is complementary to other management techniques which control excessive vegetative growth and increase water use efficiency without negative effect on yield. This represents a major shift in approach because previously irrigation was used simply to alleviate all water stress throughout the growing period. The principle concept of the DI is to establish a controlled water deficit in the plant during the periods of rapid shoot and/or slow fruit growth. This water deficit is achieved by irrigating trees at a rate of water replacement lower than the actual water use. Water is then made readily available during subsequent periods of rapid fruit expansion (Chalmers et al., 1981; Mitchell et al., 1984).

2.1 PHYSIOLOGICAL BASIS OF DEFICIT IRRIGATION

2.1.1 Phenology of tree and crop growth

Phenological separation of periods during which tissues and organs are actively growing presented one opportunity to inhibit an organ, tissue or process without seriously affecting others. Most events in plant development are seasonal and periodic and would only be sensitive to reduced water potential during active periods. Times can therefore be identified during a season when low Ψ would affect one process but not others.

At the beginning of the growing season fruit development follows shoot development. In terms of assimilate demand there is a significant separation of the

early active periods of vegetative growth and fruit growth (Chalmers, 1989). Hence one can target vegetative growth with reduced Ψ without affecting fruit growth.

2.1.2 Differential sensitivity of tissues, organs and processes

As Ψ decreases with decreasing water availability, organs, tissues and processes will be affected differently depending upon the sensitivity of the process to Ψ. Water availability can therefore be manipulated in a variety of ways to beneficially modify developmental processes in fruit trees.

Cells expand following uptakes of solutes and/or increased cell wall elasticity when they absorb water to restore the balance between the turgor potential of the cell and the negative pressure exerted by the combined resistances to stretching of the cell wall (Serpe and Matthews, 1994). Therefore cell expansion of both vegetative and fruit growth are extremely sensitive to reduced Ψ. Fruit cells expand by a similar mechanism, but because they are strong solute sinks they can attract water more strongly (Chalmers, 1989). There is ample evidence of the above phenomenon in studies conducted on apple, DI reducing excessive vegetative growth without reducing fruit growth and yield (Ebel, et al., 1995) peaches and pears(Chalmers et al., 1981, 1986; Mitchell and Chalmers, 1982; Mitchell et al., 1984) . Similarly, Irving and Drost (1987) found that water stress, if applied after the cell division phase, reduced the terminal shoot growth but did not affect fruit growth. Thus vegetative growth is more sensitive to reduced Ψ than fruit growth.

2.1.3 Functional equilibrium between shoot and root growth

Restricted root growth will tend to restrict shoot growth, because vegetative growth of plant is usually limited by the growth potential of the root system (Chalmers, 1989). It has been demonstrated that shoot growth can be inhibited by limiting the root volume of plants (Richards and Rowe, 1977). This also results in an increased ratio of fruit growth to vegetative growth per unit tree size. DI may cause the roots in the dry soil to become physiologically inactive, effectively decreasing root

volume. Smaller root volumes produced with drip irrigation, resulted in less vigorous, more fruitful trees (Mitchell and Chalmers, 1983) than more vigorous trees produced under sprinkler or flood irrigation. This suggests it may have been having an impact by way of root restriction.

2.2 PHYSIOLOGICAL CONSEQUENCES OF WATER STRESS

2.2.1 Leaf/xylem water potential

For a long time leaf and/or xylem water potential has been accepted as a measure of plant water status. The xylem water potential of water stressed plants is generally lower than the plants receiving adequate water. Brun et al. (1985) subjected pear trees to wet, normal and dry treatments and found that the Ψ of the trees under dry treatment decreased steadily after full bloom and continued as soil water depleted whilst the values remained higher in the normal and wet treatments. In 'Braeburn' apples, differences of up to 0.7 MPa in Ψ between irrigated and nonirrigated trees were observed by Mills et al. (1994). In a study by Chalmers et al. (1986) withholding irrigation reduced the leaf water potential of 'Barlett' pear trees at dawn and midday by about 0.1 and 0.5 MPa, respectively, compared to control.

Leaf water potential is also affected by time of the day. Goode and Higgs (1973) studying water potential and its component, osmatic and pressure potentials, in apple leaves of irrigated and non-irrigated trees obtained a considerable diurnal change in Ψ (from between -0.1 and -0.2 MPa before sunrise to -1.5 and -2.5 MPa after midday) even when the soil moisture level was low. The diurnal pattern of leaf water potential obtained by Olssen and Milthorpe (1983) in peach trees reached the maximum daily values after sunrise, which did not change markedly during the drying cycle. Minimum values were obtained in the early afternoon and more closely reflected the decrease in soil water potential as drying progressed. Similarly, Chalmers et al. (1983) found that the diurnal maximum values in Ψ of peach trees paralleled those of soil water potential whilst the minimum values did not change greatly with the decrease in soil water potential during the drying cycle.

In this study, the measurement of predawn and midday Ψ were taken to determine plant water status. These were complemented by measurements of soil water content.

2.2.2 Physiological processes

Because plants are often exposed to water deficit during growth and development, it is important that they are equipped with mechanisms that allow adaptation to the prevailing conditions so as to avoid serious damage to the plant. When soil water is adequate, the movement of water in plants is controlled by transpiration which is regulated by both stomatal aperture and the evaporative demand. Water stress occurs when absorption lags behind transpiration (Kramer and Boyer, 1995, p.183), and is characterised by a decrease in water content, turgor, and total water potential resulting in wilting, partial stomatal closure and decrease in cell enlargement and plant growth. Severe water stress conditions may cause cessation of growth, decrease or cessation of photosynthesis, disturbance of many metabolic processes and even death of the plant. This section discusses some of the physiological processes that are affected by water stress in the plant system.

2.2.2.1 Stomatal conductance

Stomatal conductance plays an important role in the maintenance of plant water potential as transpiration accounts for the most of water loss through the plant. Initially it was believed that decreasing Ψ caused loss of turgor in cells and resulted in stomatal closure. In several plant species, stomatal closure has been observed in response to the dryness of the air regardless of the Ψ (e.g., Cohen and Cohen, 1983). However, now it appears that the mechanism behind the stomatal closure may be somewhat complex. The involvement of hormones such as abscisic acid (ABA) (Tallman,1992) and cytokinins (Gollan et al., 1986) that are modified under conditions of water deficit may also play a role. Stomata provide dominant short time control over transpiration (Jones et al. 1985) and hence plant water status.

In addition, stomata are also sensitive to relative humidity and vapour pressure deficit. Schulze et al. (1972) found that at high RH, stomata remained opened inspite of decrease in Ψ. Midday closure of stomata on hot days as observed in many plant species might be a response to high vapour pressure deficit (VPD) in order to prevent the development of critically low leaf water potential (Lakso, 1985). Stomatal conductance in conjuction with photosynthesis under DI is investigated in this thesis.

2.2.2.2 Transpiration

Transpiration may be defined as the loss of water from the living tissue of aerial parts of the plant in the form of water vapour. High rates of transpiration often cause injury, especially in limited soil water availability. Transpiration is unavoidable. As long as there is an ingress and egress of oxygen and carbondioxide, the condition to water loss will be unavoidable. Till the vapour pressure of the plant cell walls is greater than vapour pressure of atmosphere and the water permeable membranes exists in the plants, transpiration will occur. The closure of stomata under low water potential (both soil and leaf) is an adaptive mechanism by the plants to reduce water loss via transpiration (Mcdermitt, 1990).

According to Kozlowski et al. (1991), the rate of transpiration is controlled by the energy supply, vapour pressure gradient from leaves to air, the boundary layer resistance around the leaves or plant canopy, leaf/stomatal resistance, and the water supply to the roots. In another words, the rate of transpiration increases as the steepness of the vapour pressure gradient increases from leaves to air, which depends on temperature and humidity of the atmosphere. The extent to which stomatal movement affects canopy transpiration thus depends on several factors such as energy supply and the proportion of the boundary layer resistance to stomatal resistance (Hsiao, 1990).

2.2.2.3 Canopy temperature and canopy-air temperature difference

As indicated earlier, plant water status integrates the effects of available soil water, evaporative demand and the hydraulic fluxes within the soil-plant- atmosphere continuum (SPAC). According to Jackson (1982), some of the effects of evaporative demand have been incorporated in the measurement of plant water status by the use of canopy temperature and canopy-air temperature difference as indicators of water stress. It has been shown through energy balance analysis that (T_c-T_a) is related to the leaf to air vapour pressure deficit and depends on the aerodynamic resistance to water flow and on the net radiation level for a constant T_a (Glenn et al., 1989). In general, the use of canopy temperatures to detect water stress in plants is based on the assumption that evaporation cools the leaves below the temperature of the surrounding air. As water becomes limiting, transpiration is reduced and the leaf temperature increases above air temperature because of absorbed radiation.

Measuring T_c and T_c-T_a is attractive because, it is nondestructive, and is relatively easy to use hence one can sample a large number of plants in relatively short time. It encompasses both plant and environmental factors in the determination of plant water status and may be used for whole plant measurements as well as single leaves or plant parts.

2.2.2.4 Photosynthesis

Photosynthesis is reduced in many plant species as water deficit is imposed, but the mechanism by which this reduction takes place is not fully resolved (Flore and Lakso, 1989). Other environmental factors affecting Pn include water, light, CO_2 and O_2 concentrations, mineral nutrients and leaf temperature (Lawlor, 1987). The process of Pn may be considered as diffusion of CO_2 from the gaseous phase in the atmosphere and leaf intercellular spaces into the liquid reaction site at the chloroplast, photochemical process and electron transfer, and biochemical stage, when CO_2 serves as a substrate of reduction processes to organic compounds. Inhibition of one or more of these characters limits the whole process. Plant water status may result in low

photosynthetic rates in leaves (Sritharan and Lenz, 1989). For example, water stress reduce the rate of photosynthesis in 'Golden Delicious' apples by as much as 50%. In Kiwifruit, water deficit reduced the rate of photosynthesis by 53% to 64% in relation to the well-watered control treatment (Chartzoulakis, 1993).

The process of photosynthesis requires an exchange of gases and this takes place through stomata. Therefore stomatal conductance is an important factor in photosynthesis. Stomatal closure is commonly reported in fruit crops under DI (Brun et al., 1985; Flore et al., 1985), restricting the uptake of CO_2 and hence the rate of Pn.

In some instances however, a low stomatal conductance does not closely correlate to decrease Pn. For example Mills et al. (1994) found a significant decrease in g_s in 'Braeburn' apples under DI, yet Pn was not affected. Hsiao (1993) reports that stomatal closure appears to have limited influence on leaf Pn as reduction in g_s under water deficit conditions does not necessarily results in reduction of leaf internal CO_2 concentration. Additionally, Such evidence has led to speculations that Pn may also be under non-stomatal inhibition such that the water stress has direct effect on the rate of Pn even without stomatal closure (Kozolowski et al., 1991). The inhibition of CO_2 assimilation by water stress is thought to be closely linked with to the extent of stomatal closure. The study by Janoudi et al. (1993) showed water stressed cucumber plants had lower g_s and CO_2 assimilation rates. The authors were able to show that some non-stomatal factors were contributing to decrease in the rate of Pn in the stressed plants. Non-stomatal factors include inhibition of the activity of photosynthetic enzymes e.g., fructose-1-6-biphosphate and/or ribulose biphosphate carboxylase oxygenase (Berkowitz and Gibbs, 1983; Vu and Yelenosky, 1988) and feedback inhibition of Pn due to photoassimilate accumulation (Janoudi et al., 1993).

2.2.3 Vegetative growth

The response of vegetative growth constitutes one of the underlying focuses of DI studies (Mitchell and Chalmers, 1982). The total tree growth components of leaf area, shoot growth and trunk growth were studied in this thesis.

The regulation of leaf area plays an important role in the adaptation of fruit crops to water deficit (Jones et al., 1985). Leaf area index (LAI) is a measure of the total leaf area of plant, or plants, divided by the land area covered (Westwood, 1988, p. 220). A reduction in LAI was observed in peach under early season water deficit (Boland et al., 1993). A decrease in LAI may result in reduction in the interception of photosynthetically active radiation (PAR). This in turn may reduce total carbon assimilation and dry matter production (Hsiao, 1993). Aside from reduced leaf area expansion, leaf area may also reduce due to the adaptive responses of the plant to reduced water status. Adaptation include leaf folding about the midrib in apple (Lakso, 1983), and, in extreme cases of water deficit, leaf abscission can occur (Hsiao, 1993).

Goode and Ingram (1971) studied the growth of apple trees under different irrigation systems and found that in deficit treatments trunk growth rate was not affected during the first season, but appeared affected in successive seasons. However, an effect on shoot growth occurred in the first season. In the long term, the most marked effect was on shoot number rather than the shoot elongation. Assaf et al. (1974, 1975) found low shoot and trunk growth in low irrigation treatments. In addition, Higgs and Jones (1991) demonstrated that water-stressed trees showed 40% to 50% reduction in the number of shoots and up to 62% reduction in weight of shoot removed by the pruning.

Different physiological reasons have been put forward to account for the reduction in vegetative growth in response to water stress. According to Kozlowski et al. (1991), reduced growth is that of reduced cell expansion and division. The quantity and quality of plant growth depend on the cell division, enlargement, and differentiation, and all the processes are affected by the water stress. Thus water stress reduces growth by reducing cell tugor and hence cell expansion. However, cell expansion is a complex process that requires simultaneous intake of water, extension of cell walls, and a sustained supply of solutes necessary to maintain tugor in the

expanding cell (Boyer, 1985). Hence it is often difficult to find a clear relationship between turgor and cell growth.

The possibility of irrigation strategies to control vigour of fruit crops by water stress and reduction of vegetative growth has been applied successfully in peaches and pears using deficit irrigation.

2.2.4 Reproductive growth

In the development of suitable DI strategies for fruit trees, it is important to understand the effects of water stress on the various processes involved in the reproductive phase of growth. Hanan (1972) found water stress caused serious effects on bud initiation, formation, and maturation, while Chalmers et al. (1981) reported that fruit set was reduced on peaches and pears by water stress, but increased per unit tree in DI treated trees. According to Faust (1989, p.159), severe water stress during the period of flower bud may decrease return bloom in the following year. Conflicting results were obtained by Mitchell et al. (1984) who reported that DI increased flower density in pears.

2.2.4.1 Fruit growth and yield

Fruit growth rate is linked to plant water status but little is known of the causes and affects linking water availability and fruit growth (Failla et al., 1992). Hilgeman et al. (1959) reported that water stress should be avoided during blossom and fruit set and also during the second stage of citrus fruit growth, since rates of fruit growth and final size may be diminished. Hilgeman (1972) reported that fruit from citrus trees irrigated both sides grew faster than trees irrigated on alternate sides.

The response of 'Cox's Orange Pippin' apple trees to different soil moisture conditions was studied by Goode and Ingram (1971). They found a higher yield in irrigated apple trees than in non-irrigated trees. In studies by Guelfat'Reich et al. (1974) and Looter et al.(1985) 'wet' treatments always yielded larger fruit and higher yield than 'dry' treatments.

Assaf et al. (1974) found a linear correlation between the percentage of apple fruit with diameter greater than 6.5 cm and number of days in which the 0-60 cm layer of soil was subjected to less than 30% of available water during the main period of fruit growth. They emphasised the importance of frequent irrigation to maintain the available water well above 30% in the top soil layer in order to obtain maximum fruit size.

Chalmers et al. (1981), using DI, obtained an increase in fruit size and yield of peaches. Beukes and Weber (1982) found that the optimum sequence of water levels necessary to optimise yield on apples was one which indicated a deficit during the first and fourth phenological phases and the highest water level in the third phenological phase.

In this study, I attempt to determine if applying DI during the late and entire season has any effects on the reproductive growth of 'Braeburn' apples.

2.3 FRUIT QUALITY

The production of high quality fruit is critical to maximise returns to the grower with premium prices being paid for export fruit. Any management strategy must therefore address the impact it may have on the quality of produce. Consumer's preference define fruit quality (Kingston, 1992). According to Krishnaprakash et al. (1983), the quality of apples is largely determined by the maturity of the fruit when harvested and on the rate of further maturation, ripening and subsequent behaviour in storage. In most fruits flavour, texture and appearance play importance roles. As fruit reaches maturation and ripening several changes takes place. These include changes in colour, flesh firmness, fruit respiration rate, acidity levels and total soluble solids concentration (Kingston, 1992: Westwood, 1993, p.301). Thus fruit quality changes are expected under DI. Although there is limited information concerning the effect of DI on the quality of the fruit, this is of paramount importance if DI is to be used as a management tool in the fruit production. The influence of late and entire-season DI is therefore addressed in this thesis.

2.3.1 Effects of water deficit on total soluble solids

Assaf and Bravdo (1975) evaluated the effect of six irrigation regimes on quality of apples. At harvest, fruit from dry treatments had the highest total soluble solids (TSS), whereas fruit from extreme wet treatments had the lowest TSS. Differences among the rest of the treatments were not significant. Guelfat'Reich et al. (1974) obtained the highest TSS at harvest and during storage, and the best shelf life in those treatments which were irrigated two weeks after soil water content reached permanent wilting point in apples. However in the study by Proebsting et al. (1984), the soluble solids concentration was higher in the stressed fruit well before maturity (108 days after full bloom). In Asian pears, Behboudian et al. (1994) found a higher TSS in fruit from stressed trees as early as 35 days after full bloom. The increase in TSS could be due to enhanced conversion of starch to sugars which occurs as a result of water stress (Kramer, 1983) or a dilution of solutes in well-watered treatments. It would be of interest to determine whether the seasonal timing of reduced irrigation has any effect on the concentration of TSS.

2.3.2 Effects of water deficit on titratable acidity

Malic acid is the predominant organic acid in apples (Kays, 1991, p.278). During maturation, ripening, and storage, the total organic acid content of the fruit gradually declines (Kays, 1991, p.278; Kingston, 1992). This loss of acidity is largely due to utilisation of these compounds as respiratory substrates (Biale and Young, 1981). A measure of titratable acidity (TA) is thus a useful maturity index in apples. The flavour of the fruit results from the combination of acids, sugars, and volatile within the fruit (Kingston, 1992).

Layne et al. (1981) reported no change in acidity of peach under DI, but information on TA in apple is conflicting. Guelfat'reich et al. (1974) reported that apples from trees subjected to water stress had lower TA levels than those from well-watered trees. However, Irving and Drost (1987) showed no change in TA between irrigated and DI treatments. Drake et al. (1981) also found that TA was

similar at harvest between fruit receiving adequate moisture and those receiving DI. After five months storage, however, the TA was much less in the stressed fruit. Therefore, it is not known whether the time at which water stress or reduced irrigation is applied has any consequence on the TA of the fruit at harvest and during storage.

2.3.3 Effects of water deficit on fruit colour

Fruit colour in apples is an important quality attribute and is the primary index to determine the degree of fruit maturity (Lancaster et al., 1994). Fruit colour is dependent on various pigments present in the skin and the type of radiation (Gorse and Creasy, 1977). According to Lancaster (1992), red colour is due to anthocyanins and flavonols and is stimulated by light and cool temperature.

The degree of red blush development of fruit may not be a good indicator of fruit maturity because it varies markedly between fruit position in the canopy. This is mainly due to effect of light on fruit colour (Kingston, 1992). Studies have shown that red colour development is higher in apples developing under high light intensities than in apple developing under shade (Krishnaprakash et al., 1983). Kingston (1992) therefore suggests the use of ground colour, i.e. the colour of the unblushed portion of the fruit, as a more reliable indicator of maturity. Indeed, in 'Braeburn' apples the yellow background is widely used as an indicator of fruit maturity (Kupferman, 1994).

Fruit nitrogen levels may also influence ground colour development with high fruit nitrogen levels being associated with poor fruit colour (Saure, 1990; Bramlage, 1993). High nitrogen levels enhance chlorophyll retention hence retarding yellow ground colour development (Magness et al., 1940). Deficit irrigation is known to reduce the level of N in pear fruit (Raese et al., 1982) and in apple (Goode and Ingram, 1971). However, direct experimental data are not available to substantiate this. Fruit colour development under DI has been minimally studied in apple. Mills et al. (1994) reported that late-season withholding of irrigation enhances the red colour of 'Braeburn' apples. Proebsting et al. (1984) found that DI applied throughout the season did not affect the red colour of 'Delicious' apples. Ebel et al. (1993) also

found no effect on fruit colour under early-season DI. There is a need to quantify the effects of water deficit on apple fruit colour and to determine the optimum time during the season for withholding irrigation for enhanced skin colour in apples.

2.3.4 Effects of water deficit on fruit firmness

Fruit firmness is one of the most important quality alterations strongly influenced by fruit maturity, with firmness decreasing in apple and pears as the fruit ripen (Kingston, 1992). Changes in texture of fruit during ripening results from the changes in structure and composition of their cell walls (Rhodes, 1980). Fruit firmness is also influenced by fruit size (Ebel et al., 1993; Amen et al., 1983), while small fruit exhibit the highest firmness than larger, due to higher cellular density. Ebel et al. (1993) found when the firmness was adjusted to remove the effect of size, there was no difference in firmness between treatments.

2.3.5 Effects of water deficit on fruit ethylene evolution

Fruit ethylene evolution has been associated with the respiratory rise of the climacteric. It is, therefore, a good indicator of optimum harvest time (Watkins et al., 1989). Fruit softening, colour changes, development of desirable taste and aromatic flavour are associated with the climacteric cycle (Biale and Young, 1981) and hence the fruit enters the ripe edible stage at or shortly after this peak. Therefore, the rate of ethylene evolution and/or internal ethylene concentration is a useful indicator of the fruit maturity and fruit potential storage life.

Narayana et al. (1991) reported that water deficit is one of the more commonly reported stresses to cause an over production in ethylene. Guelfat'reich et al. (1974) observed that in apple fruit exposed to water deficit during development, ethylene production was greater after harvest than it was in fruit from the well- watered treatments. Ebel et al. (1993) also found that internal ethylene concentrations were higher in DI apples than in the control apples during developmental period. Further, they indicated that water deficit encourages ethylene production of apple fruit and

thus influences the physiological maturity of the fruit. According to Ebel et al. (1993), the apples receiving DI early in the season entered an 'earlier-than-normal' climacteric rise.

2.3.6 Storage life and disorders

An important aspect of fruit quality is the occurrence of fruit disorders, both at harvest and after cold storage. Inferior keeping quality in storage of water dificit apple fruit was recorded following irrigation trials by Guelfat'reich et al. (1974). The occurrence of physiological and pathological disorders such as bitter pit and scald was lower in fruit from drier treatments than in wet treatments. In contrast Goode et al. (1975) observed increased cracking and rusetting in apple fruit grown under reduced irrigation. Irving and Drost (1987) found less incidence of bitter pit in fruit that was grown under water stress imposed six weeks after full bloom. Non physiological disorders such as the incidence of fungal rot may be also be modified by reduced irrigation as was reported for peach (Li et al., 1989). Proebsting et al. (1984) working on apple, suggested that irrigation has no effect on storage and that changes in fruit were the same independent of irrigation. Part of my research was therefore devoted to the study of storage effects on some fruit quality attributes for fruit grown under different DI treatments.

2.3.7 Effects of water deficit on fruit mineral concentration

The storability of fruit and development of disorders have been attributed to the concentration of minerals, (Faust, 1989, p. 55). For example, bitter pit is a particularly important Ca^{2+} related physiological disorder in 'Braeburn' apples (Kupferman, 1994). Cork spot a disorder of both apples (Miller, 1980) and pears (Raese et al., 1982), is also a Ca^{2+} related disorder.

The concentration of other major minerals in the fruit also influence storage behaviour of the fruit. For example, Mg^{2+} as well as K^{+} are considered antagonistic to Ca^{2+}. High K^{+} and Mg^{2+} generally aggravate problems caused by lack of Ca^{2+}

(Bangerth, 1979). While low P increases the risk of low temperature breakdown and is also associated with reduced firmness (Johnson et al., 1987). Too much N leads to decreased fruit firmness, increased chlorophyll and therefore less red and yellow skin colour and may also increase fruit susceptibility to rotting (Kingston, 1992).

Several experiments have been undertaken to investigate the effect of water stress on fruit mineral concentrations. However, results from these studies are conflicting. Goode and Ingram (1971) found that water stress led to decrease in Ca^{2+} concentration and an increase in K^+ concentrations. In contrast, Mills et al. (1994) found no effect of water stress on K^+ concentrations. Whereas Guelfat'reich et al . (1974) and Looter et al. (1985) found a lower K^+ concentrations in water-stressed fruit at harvest. Irving and Drost (1987) reported that water stress had no effect on the concentrations of N, P, K^+, Mg^{2+}, and Ca^{2+} . These conflicting results from the different authors may have been due to water stress/DI being imposed at different times of the growing season. Therefore further investigation is needed.

2.3.8 Effects of water deficit on volatile compounds

The importance of flavour and aroma to the consumer cannot be overemphasised. Yet there is no literature report on the effect of water stress on the aroma and the flavour of apples. These attributes are as a result of the many components of volatile compounds found in the fruit (Panasiuk et al., 1980). These compounds are synthesised from the basic nutrients such as carbohydrates, Proteins, fats, vitamins and minerals (Salunkhe and Do, 1976). Because of the changes in the chemical composition of the fruit as a result of water stress, one can expect that the composition and quantity of volatile compounds within the fruit is also affected. As indicated earlier there is no information available as to the effect of reduced irrigation on the composition and quantity of volatile compounds of apples. This aspect was therefore studied in this research.

2.3.9 IRRIGATION TIMING

The discussion so far indicates that the impact of DI is strongly dependent on the timing of the DI, since most events in plant development are seasonal and/or periodic. Importance of proper timing of irrigation applications has been well researched (Rawitz, 1969; Acevedo et al., 1971; Hillel and Guron, 1973; Garnier and Berger, 1987). Timing of irrigation has to be adjusted to soil water depletion according to crop requirements which vary with evaporative demand, root depth, size of the soil particles and the water holding capacity of the soil (Clothier et al., 1987). The trees enter dormancy over the winter months so that plant water is minimal, although root growth will continue in winter (Kramer and Boyer, 1995, p. 125). Implementation of DI during different phenological stages constitute the primary focus on this thesis.

2.3.9.1 Early-season deficit irrigation

Water deficit during flowering is likely to inhibit fertilization (Hsiao, 1993; Powell, 1974). However, early-season DI following the completion of flowering and fruit set could result in the same yield as full irrigation with considerable saving in water (Ebel et al., 1995; Li et al., 1989). Early-season DI decrease some fruit disorders such as cork spot in pear (Brun et al, 1985). But in some cases it can increase the disorders such as flesh spot decay in 'Nijisseiki' Asian pear (Behboudian and Lawes, 1994) and also reduce cell number (Hsiao, 1973) and thus the final fruit size. Nevertheless, early-season DI can be used as an effective management tool.

2.3.9.2 Entire-season deficit irrigation

Hsiao (1993) reported that entire-season water deficit would reduce economic crop yield and total biomass production as assimilate accumulation is reduced under water stress conditions. Kilili et al. (1996) showed reduced fruit weight, advanced fruit maturity in apple encouraging an earlier climacteric rise in ethylene production,

yellower background colour and increase in TSS from apple trees exposed to an entire-season DI. A entire-season DI on apple is also studied in this thesis.

2.3.9.3 Late-season deficit irrigation

This refers to a deficit that is imposed during the later stages of fruit growth prior to harvest. In 'Hosui' Asian pears, a late-season deficit did not reduce shoot growth, or total leaf area (Caspari, 1994). According to Lotter et al. (1985) late-season deficit may reduce fruit yield. According to Li et al. (1989) and Irving and Drost (1987), Fruit composition and some fruit quality attributes are modified under late-season DI. Desirable fruit quality changes may make late-season DI advantageous in certain situations. However, more information is needed on particular species and varieties before it can be widely recommended. A late-season DI on apple is also focused in this thesis.

2.3.9.4 Postharvest deficit irrigation

In most fruit crops, especially early maturing varieties, a significant amount of tree growth occurs during autumn. Johnson et al. (1992) showed that water deficit at this time reduce the pruning requirements of peach, while increasing flower density and in blackcurrents it has shown a reduction in flower buds and fruit set in the following season. Unfortunately it also increased the occurrence of double fruits. Postharvest deficit reduces radial trunk growth more than shoot growth, as shoot growth is predominant during spring. A postharvest deficit may be of benefit to some species and varieties, and it may even help control tree vigour and make tree winter hardy by reducing late season growth. However, limited information on this technique makes any conclusion difficult. It is not expected that a postharvest deficit will have a significant influence on vegetative growth of 'Braeburn' apples. Because fruits were harvested late in the season with leaf fall occurring shortly after final harvest. Therefore, this was not studied in my research.

CHAPTER THREE

MATERIALS AND METHODS

The experiment was carried out at the Massey University's lysimeter facility, Palmerston North, New Zealand (Lat. 40.2^0 S, Long. 175.4^0 E) during the 1995-1996 growing season. All laboratory analysis of fruit attributes was carried out using the laboratory facilities of the Plant Science Department, Massey University.

3.1 EXPERIMENTAL SETUP

3.1.1 Lysimeter facility

The facility consists of 12 drainage lysimeters, each being constructed from a steel cylinder 1.2 m deep and 1 m in diameter. Each steel cylinder is surrounded by a concrete sleeve which gives a 1.2-m tree spacing within the row. The tops of the cylinders are about ground level. The bottom 0.15 m of steel containers is filled with sand, while the bottom 0.05 m of the cylinder is conical in shape to provide drainage water to a subterranean facility. Eight of the cylinders were packed with Manawatu fine sandy loam excavated from the B horizon of the surrounding orchard. The remaining four cylinders were packed with 0.4 m gravelly coarse sand excavated from the C horizon of the surrounding orchard. The B horizon was then placed on top. Trees were trained as central leaders with central support wires installed to give additional support. Irrigation and fertilizer was applied to each tree via a closed nutrient-irrigation system fed from two 9100-liter tanks. Nutrient concentration of the solution was (in mg liter^{-1}): N, 105; P, 31; K, 68.4; Ca, 125; Mg, 48.6; S, 98; Fe, 3.0; Cl, 26.6; B, 0.5; Mn, 0.5; Zn, 0.05; Cu, 0.02; and Mo, 0.05. The pH of the solution was adjusted to 6.5 by adding sulphuric acid. The solution was pumped, filtered, and divided into three lines. Each line was controlled by a solenoid (model M886N24D, Bermad, Israel) which was independently operated and supplied nutrient solution to

four individual lysimeters each containing one tree. The nutrient solution was applied to the surface of the soil in each lysimeter via four pressure-compensating trickle emitters (Netafim, Israel). Each emitter was rated at 2 liters hr^{-1}. However, calibration of individual emitters indicated some variability. The emitters were placed in a circle approximately 0.3 m from the trunk of the tree in an attempt to provide a uniform water distribution across the rootzone. Each individual lysimeter was covered with white reflective plastic covers to exclude rainfall from the rootzone. Each lysimeter drained through a polyethylene pipe to a tipping-bucket gauge (Rain-O-Matic, Pronamic, Them, Denmark) located within a subterranean control room. A multi-tasking controller/datalogger (Wormald 1830, Wormald Vigilant Ltd., Christchurch, New Zealand) maintained operations of the lysimeter facility automatically. The controller/datalogger stored digital pulses from flow meters and drainage gauges, and controlled the pump and solenoids.

3.2 PLANT MATERIAL

Five-Year-old 'Braeburn' trees grown on MM.106 rootstock, trained as central leader, were used in the experiment. A total of 12 trees were assigned in a completely randomised design with tree treatments. The treatments were: Control (C), comprising well watered trees; entire deficit (ED), where DI was applied from full bloom (20 October) until final harvest i.e. throughout the season; late deficit (LD), where DI was applied from 102 days after full bloom (DAFB) up to final harvest on 3 April 1996 (196 DAFB). The DI treatment was irrigated with 2 liters/day during their stress period and 4 liters/day for C. Covers were installed under all trees to exclude rainfall.

3.3 SOIL MOISTURE

Weekly measurements (two per tree) were made of the soil volumetric water content (θ, $m^3\ m^{-3}$) to a 1-m depth, using time domain reflectometry (TDR) equipment

(model 1502C; Tektronix, Redmond, Ore.) (Topp and Davies, 1985). Measurements commenced at 63 DAFB.

3.4 LEAF WATER POTENTIAL (Ψ)

Midday leaf water potential measurements were made weekly between 1200 and 1300 HR, using a Scholander pressure chamber (Soilmoisture Equipment Corp., Santa Barbara, Calif.). Measurements commenced at 50 DAFB. Two fully expanded mature leaves per tree were used from exposed shoots in the middle tree canopy. The leaves were immediately enclosed in small plastic bags to avoid water loss due to evaporation then the leaves were placed in the pressure chamber which was humidified with moist tissue paper. Nitrogen gas was used to apply pressure until leaf sap appeared at the cut cross-section of the vascular tissue. The pressure applied was taken as an estimate of bulk leaf water potential. Predawn Ψ measurements were taken weekly between 0500 and 0600 HR.

3.5 PHOTOSYNTHESIS AND STOMATAL CONDUCTANCE

Photosynthesis and stomatal conductance were taken on two leaves per tree using a portable gas exchange measurement system (model 6200, LI-COR, Lincoln, Neb.). For leaf measurements, the youngest mature leaves were selected from the current season's growth and were in full sun at the collection time. The other parameters recorded at the time of measurement using the same equipment were: photosynthetically active radiation (PAR in μmol quanta m^{-2} s^{-1}), vapour pressure deficit (VPD in KPa), and the rate of transpiration (mmol m^{-2} s^{-1}). Plant measurements were taken between 1200 and 1400 HR, commencing at 42 DAFB.

3.6 CANOPY TEMPERATURE AND CANOPY-AIR TEMPERATURE DIFFERENCE

Canopy temperature (T_c) and canopy air temperature difference (T_c-T_a) were determined weekly using an Infrared (IR) thermometer (Everest Inter science Inc.,

Tustin Ca.). The instrument measures the radiation emitted from the target and relates this radiation (R) to the surface temperature (T_s) by the Stefan-Boltzmann law (Jackson, 1982):

$$\mathbf{R} = \varepsilon\delta\mathbf{T_s^4}$$

where ε = Emissivity of the surface which is set at 0.98 for leaves

δ = Stefan-Boltzmann constant ($5.674*10^{-8}$ Wm^{-2} K^{-4})

T_s = Surface temperature R = Radiation (Wm^{-2})

In this experiment T_c and T_c-T_a measurements were made weekly at 1300 HR. Values were recorded from the display window within 5 seconds after reading had stabilized.

3.7 VEGETATIVE GROWTH

In order to determine the effect of treatment on vegetative growth of the trees, the trunk circumference of all experimental trees was measured at 300 mm above the ground level using a metric tape. This was done at the beginning of the experiment and at time of final harvest. The data were used to calculate the change in trunk cross-sectional area (TCA) over the season, leaf area was estimated by removing every tenth leaf from each tree after fruit harvest. The leaf area of this fraction was then measured with area meter (model LI 3100; LI-COR), and the total leaf area per tree subsequently inferred.

3.7.1 Shoot growth

For the measurement of the shoot extension, four current season's shoots from each tree were selected at the beginning of the experiment. The shoots were tagged and their lengths measured at weekly intervals starting 45 DAFB until growth ceased. Growth was assumed to have ceased at 138 DAFB because there was no evidence of further shoot growth.

3.8 REPRODUCTIVE GROWTH

Four fruit of uniform size were tagged and labelled on each tree at 45 DAFB. Fruit diameter was measured once a week across the widest part of the fruit using a digital calliper (Mitutoyo Corp. Japan).

3.9 FRUIT QUALITY UNDER DEFICIT IRRIGATION

At final harvest (196 DAFB), four fruits were sampled from each of the top and bottom canopy positions from each tree for quality determination. The sampled fruit were used for the determination of total soluble solids concentration, titratable acidity, fruit colour, flesh firmness, fruit mineral composition (K^+, Mg^{2+}, Ca^{2+}, N and P), and concentration of volatile compounds.

3.9.1 Total Soluble Solids (TSS)

The concentration of total soluble solids was determined using an Atago refractometer (0-20% Brix; ATC-1; Atago, Tokyo) with automic temperature compensation. Total soluble solids concentration was determined on juice squeezed from approximately 5 mm thick apple slices taken from across the equator on two opposite sides of each fruit. The prism surface and the sunlight plate were thoroughly washed and dried with soft tissue paper after each reading.

3.9.2 Titratable Acidity

For the determination of titratable acidity (TA), fruit were peeled and the flesh cut into small pieces. The samples from each tree were then homogenised into one sample per tree. Once, homogenised, 1 g of fruit pulp was mixed with 39 ml of double-distilled water and titrated with 0.1 N NaOH using an automic titrator (model DL 21; Mettler, Griefensee, Switzerland) to an end point of pH of 7.1. The values were expressed as percent malic acid.

3.9.3 Fruit Colour

After harvest, 16 fruits per treatment were selected randomly and placed randomly on trays. The trays were then kept at room temperature (approximately 20 ^{0}C). The skin colour was determined using a chromameter (CR-200; Minolta. Osaka, Japan). The values were obtained as the mean of the two measurements.

3.9.4 Flesh Firmness

Approximately 1-mm thick peel section was removed from the equatorial part of two opposite sides of each fruit using a sharp blade. Flesh firmness was determined using an Effegi penetrometer (model FT 327; R. Bryce, Alfosine, Italy) with an 11.1 mm plunger and mounted on a drill stand with a lever to apply a constant and even pressure to the fruit. Flesh firmness was obtained as the mean of two measurements and converted to neutons (N) by multiplying by 9.807. The penetrometer was zeroed then washed with distilled water after each reading.

3.9.5 Fruit ethylene evolution and CO_2 production

To collect the gas, the fruits were placed in the plastic jar with an air tight lid. After 1/2 hour sampling was done by taking 1 ml from each jar using 1 ml gas-tight glass syringes (Hamilton Co., Nev.). The needle was inserted through the septum into the chamber and flushed before the sample withdrawal.

Ethylene concentration (µl litre^{-1}) in 1 ml gas samples was determined within approximately 30 seconds of sample withdrawal using a PYE Unicom gas-liquid chromatography (series 104) fitted with a flame ionisation detector (FID) and with a stainless steel activated alumina column (80/100 mesh, 6' long and 1/8 " diameter), and GC-8A Shimadzu gas liquid chromatography for CO_2 production, as an estimte of respiration. The temperature of column injector, and detector were 100 ^{0}C, 100 ^{0}C, and 130 ^{0}C respectively. The carrier gas was nitrogen with a flow rate of 30 ml min^{-1}. Hydrogen and air were used for the detector with a flow rates of 30 ml min^{-1} and 300

ml min^{-1} respectively. The response to sample injection was measured as peak height using an integrator (Hewlett Packard 3393A).

3.9.6 Fruit Mineral Composition

Approximately 5 grams of cortical tissue was taken from each fruit and placed in a labelled glass vial to make one composite sample from each tree. The fruit samples were dried at 70 ^{0}C for 14 days and then stored in a dry environment at room temperature prior to analysis. At the time of analysis, fruit samples were ground into powder using a coffee grinder and kept in an oven at 70 ^{0}C for about 12 hours to drive away any moisture. Two separate extractions were carried out in different sets of digestion tubes. For each series of analysis, 0.1 g of dried sample was weighed into labelled digestion tube.

All glassware used for K^+, Mg^{2+} and Ca^{2+} analysis were washed in 2M HCL . Samples were digested by adding 4 ml concentrated nitric acid (70%) to approximately 0.1 g of each sample or standard in a digestion tube. The digestion tubes were set in a heating block with a small funnel in the top to cause refluxing, and heated at 150 ^{0}C until the solution became clear. The funnels were then removed and the temperature raised to 250 ^{0}C. This reduced the extract to dryness after approximately four hours. Fifty ml of 0.2 M HCL was added to each digestion tube and well mixed using a Vertex mixer. Sufficient volume (30 ml) was poured into labelled glass vials and K^+, Mg^{2+} and Ca^{2+} were analysed using an atomic absorption spectrophotometer (model GBC 904AA; GBC Scientific Equipment, Dandenong, Victoria, Australia). Dilution was performed as required to ensure that the results were within the range of a standard curve which was computed at the same time.

The amounts of N and P were determined by colorimetric autoanalysis (Technicon Instrument Corp., NY, USA) following Kjeldahl digestion (Twine and Williams, 1971). Kjeldahl solution was made in a fume cupboard by heating a mixture of 250 g K_2SO_4, 2.5 g selenium powder and 2.5 l concentrated H_2SO_4. Four ml of Kjeldahl solution was added to digestion tubes containing preweighed 0.1 g

ground fruit sample or standard then heated at 350 ^{0}C for 4-5 hours until the solution became clear. The volume was made up to 50 ml using distilled water and mixed using a Vertex mixer. Sufficient volume (30 ml) was poured into labelled glass vials and the concentrations of N and P were determined within the range of standard curve.

3.9.7 Volatile compounds

Apples were cut into eight pieces first longitudinally giving two halves. Each half was then cut into four pieces which were then ground in a domestic fruit juicer (Kenwood brand model JE 500). The juice was collected and left ambient for at least 30 minutes before solvent extraction. Aroma volatiles were extracted using a solvent mixture of diethyl ether and pentane at a ratio of two to one (2:1 v/v) (Larsen and Poll, 1990). One 10 ml sample of juice was placed into each of two 20 ml Wheaton disposable scintillation vials. An internal standard consisting of 4 ml N-Ocyl aectate was added to the juice followed by the solvent mixture. The juice/solvent mixture was thoroughly mixed using a Vertex stirrer. Juice/solvent samples were placed at -18 ^{0}C for at least 3 days before the solvent was decanted off and combined to give a 20 ml solvent extract. The remaining juice was discarded.

The diethyl ether/pentane solvent extract was dried from 20 ml to about 200 µl and placed into a glass 200 µl flat bottom insert (Sun International Training cat. no.200 232) in 1.5 ml glass screw top auto sampler vials (Sun International Training cat. no.200 252) suitable for a Hewlett Packard 5890 series II plus gas chromatograph auto sampler (Hewlett Packard 7673 Controller and Injector and Model 185968 100 Sample carousel). The vials were sealed with a plastic septum (Sun International Training cat. no. 200 368) before placing in the carousel. One ml samples were injected into the gas chromatograph and the syringe was washed 8 times with clean diethyl ether/pentane between each injection.

One ml of solvent extract was measured by capillary gas chromatography using a Hewlett Packard 5890 Series II Plus gas chromatography connected to an IBM compatible personal computer equipped with Hewlett Packard Chemstation software

(Version B.02.04). The capillary column was a J&W 30m x 0.32 mm (i.d) fused silica, DBWAX, 0.5 mm film thickness (Alltech cat. no 93526). Injector and detector temperatures were 150 ^{0}C and 250 ^{0}C, respectively. The temperature programme was according to the following: oven temperature was held at 40 ^{0}C for 5 minutes, then programmed to 120 ^{0}C at 50 ^{0}C/min then to 190 ^{0}C at 20 ^{0}C/min with no holding time making a total run time of 24.5 mins. The split injection mode was used with a split flow rate of 100 ml/min. Septum purge flow rate was 5-6 ml/min. Air and hydrogen flow rates to the detector were 400 and 30 ml/min respectively.

Identification of aroma compounds was by comparison with the retention times of authentic compounds. Concentration of aroma compounds was estimated by comparison to known amounts of authentic compounds and correction for sample losses estimated from reduction in the internal standard.

The Intact harvested fruit were stored for 12 weeks at 0 ^{0}C for inspection of any possible disorders. After 12 weeks the fruits were taken out and all the above post harvest measurements were done on sixteen fruits per treatment.

3.10 STATISTICAL ANALYSIS

Statistical Procedures included analysis of variance performed using SAS software (SAS Institute, Cary, N.C.). Data were analysed as a complete randomised design (CRD) for four replications and three treatments. Mean comparisons were carried out using the Duncan's multiple range test.

CHAPTER FOUR

RESULTS AND DISCUSSION

This section attempted to establish if and to what extent water stress developed in 'Braeburn' apples by application of DI at different times of the growing season and to relate this to various physiological responses.

4.1.1 Soil volumetric water content

Results for soil volumetric water content (θ) are presented as the means of four measurements per treatment (Fig. 1). From start of the measurements 63 DAFB, θ was significantly lower in ED than in C until harvest. As the late deficit irrigation started (at 102 DAFB), θ of LD plants declined to 0.11 $m^{-3}m^{-3}$.

4.1.2 Leaf water potential

Predawn Ψ for C remained high for most of the season whereas ED was always lower than C (Fig. 2). The largest difference of 0.4 MPa was recorded between the trees not receiving irrigation (ED and LD) and the C at 140 DAFB (Fig. 2). Midday Ψ followed a similar trend as the predawn Ψ (Fig. 3A and B). Most of the time Ψ for C remained high. A significant reduction in midday Ψ was observed in ED as early as 49 DAFB ($P \leq 0.05$). However, differences between the C and ED at this time were generally small being less than 0.3 MPa in most cases. A lower midday Ψ in ED in relation to C was maintained until harvest. For LD the midday Ψ gradually declined to the same level as ED.

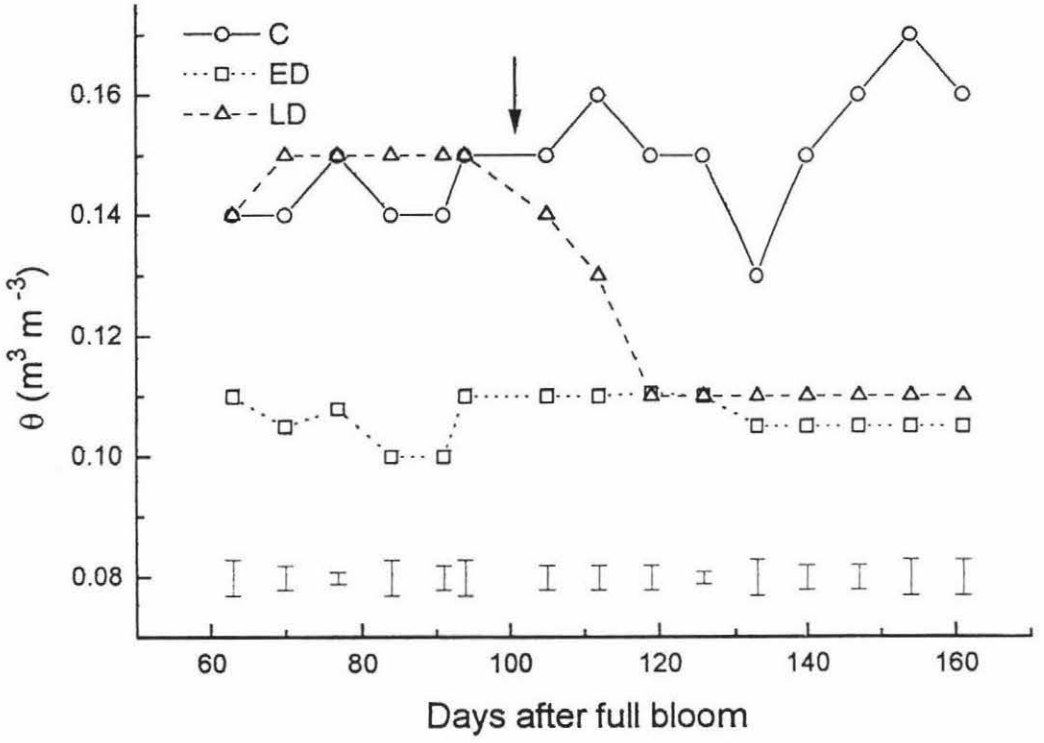

Figure 1. Changes in soil volumetric water content (θ) during the season for control (C), entire-season deficit (ED), and late-season deficit (LD) treatments. Arrow indicates the start of LD. Vertical bars represent the pooled standard errors of means based on four replicates per treatment.

Midday Ψ is a sensitive measure of plant water status in apple (Erf and Proctor, 1987). In this study, experimental trees did not experience severe water stress at any stage. During the early season (before 102 DAFB), Ψ was between -1.4 MPa and -1.8 Mpa. During the late season, the lowest Ψ was -2.0 MPa for LD. Mills et al. (1994) reported differences of about 0.7 MPa between irrigated and nonirrigated trees and this was effective in causing differences in various fruit and plant characteristics. The result from the current study are similar to those of Irving and Drost (1987) who found Ψ values were in the range of -1.3 MPa to -1.8 MPa for C and -1.7 MPa to -2.3 MPa for stress treatments.

Predawn Ψ was also a clear plant-based indicator of the treatment effects, and the data indicated that the DI treatments had significant effects on the plant water status. Previous studies have shown that predawn Ψ is closely correlated with soil moisture content since it closely represents soil water availability after equilibration of soil-plant potential at the end of the night (Xiloyannis et al., 1980). The lowest predawn Ψ was -0.8 MPa for DI trees compared to -0.4 MPa for C which indicates that the trees developed an internal water deficit. Seasonal predawn Ψ differed between treatments in a pattern similar to mean θ and there was clear relationship between Ψ and θ during the course of experiment. Linear regression analysis indicated a significant ($P \leq 0.0001$) relationship between predawn Ψ and θ and between midday Ψ and θ as follows: $\theta = -0.8 + 3.6 \times \Psi$, $n = 38$, $r^2 = 53.90\%$, and $\theta = -2.4 + 7.6 \times \Psi$, $n = 32$, $r^2 = 50.6\%$, respectively. A reduction in θ therefore does accompany a reduction in Ψ. Although ED plants were subjected to the longest period of DI, their Ψ was similar to that of LD which only had a short period without irrigation in late season. This might be due to Ψ adaptation under prolonged stress in ED and/or the effect of humid climactic conditions and the limited degree of soil drying. The soil in the experimental block is deep and has a good water retention capacity (Clothier et al., 1977).

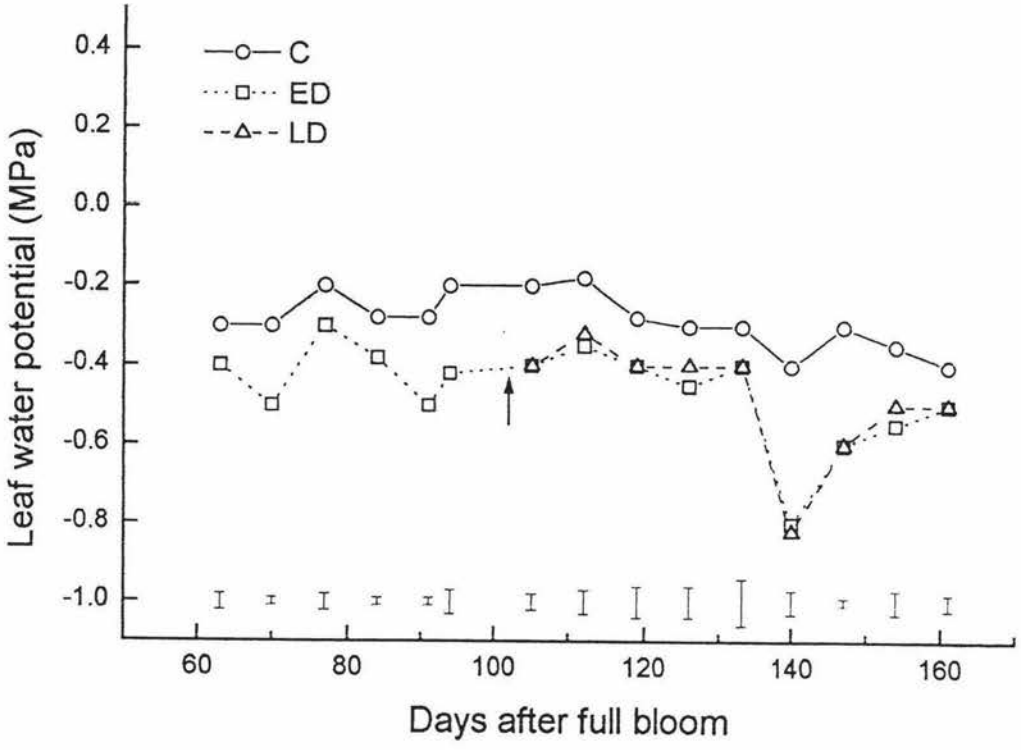

Figure 2. Changes in predawn leaf water potential during the season for control (C), entire-season deficit (ED), and late-season deficit (LD) treatments. Vertical bars represent pooled standard errors of means based on four replicates except for C which was based on eight replicates before LD started. Arrow indicates the start of LD.

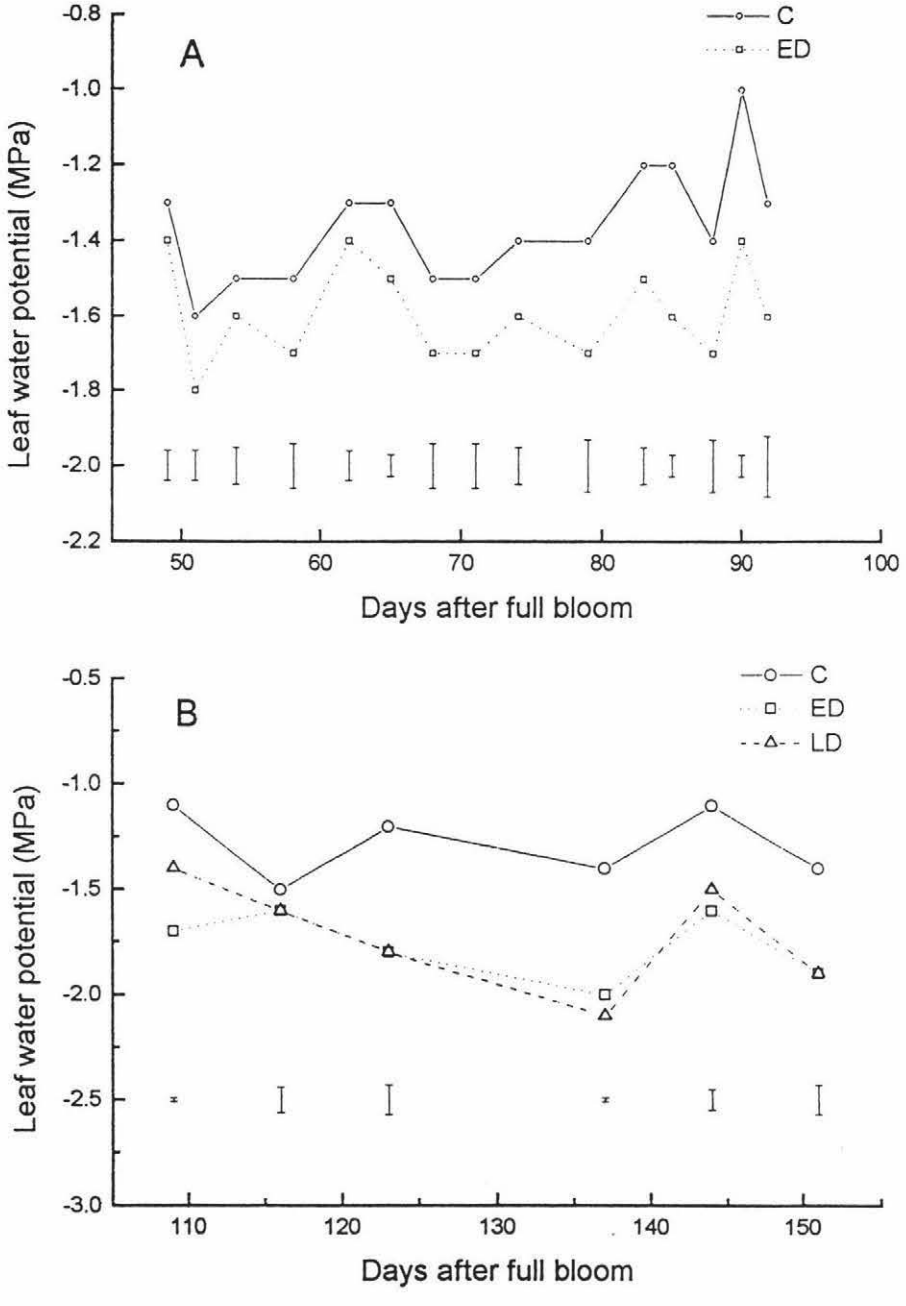

Figure 3. Changes in noon leaf water potential (Ψ) during the early season (A) and late-season (B) for control (C), entire-season deficit (ED), and late-season deficit (LD) treatments. Vertical bars represent pooled standard errors of means based on four replicates except for C which was based on eight replicates before LD started (A).

4.1.3 Photosynthesis and stomatal conductance

During the early season (before 102 DAFB), the rate of photosynthesis was not affected by the treatments (Fig. 4A). A reduction in Pn was observed in ED treatments compared to C, at 115 DAFB. At the last date of the measurement Pn of the ED was not significantly different from the LD but differed from C (Fig. 4A). Stomatal conductance remained unaffected by the treatments until 140 DAFB (Fig. 4B). Thereafter g_s was always lower in ED and LD compared to C.

4.1.4 Rate of transpiration

The rate of transpiration was closely followed with changes in g_s. There was no significant difference between the treatments for most of the growing season. Earliest difference between the treatments was observed at 138 DAFB (Fig. 5) when transpiration in C was higher than the stressed trees. These differences were maintained until harvest.

As seen from the Ψ and θ measurements, DI trees were stressed significantly compared to C trees. It is also clear that limited water availability caused significant decrease in g_s and Pn which was observed in the late season towards harvest. Stomata are sensitive to lowering of Ψ (Jones et al., 1985). In this study, reduction in Ψ and θ did not affect g_s until 140 DAFB. These results are similar to those obtained by Mills et al. (1994) who observed a reduction in g_s from 140 DAFB in nonirrigated trees. Stomatal resistance in apple leaves increased dramatically when Ψ fell below -1.9 MPa (West and Gaff, 1976). In the current study, Ψ declined to less than -2.0 MPa for stressed trees observed at 140 DAFB when a reduction in g_s was first observed.

4.1.5 Canopy Temperature and Canopy-air Temperature Difference

The treatments had no effect on T_c during the early season (Fig. 6A). At 140 DAFB, ED had a higher T_c than ED and C by 1.5 ^{0}C. There was no significant difference between treatments although T_c in ED and LD tended to be higher than C.

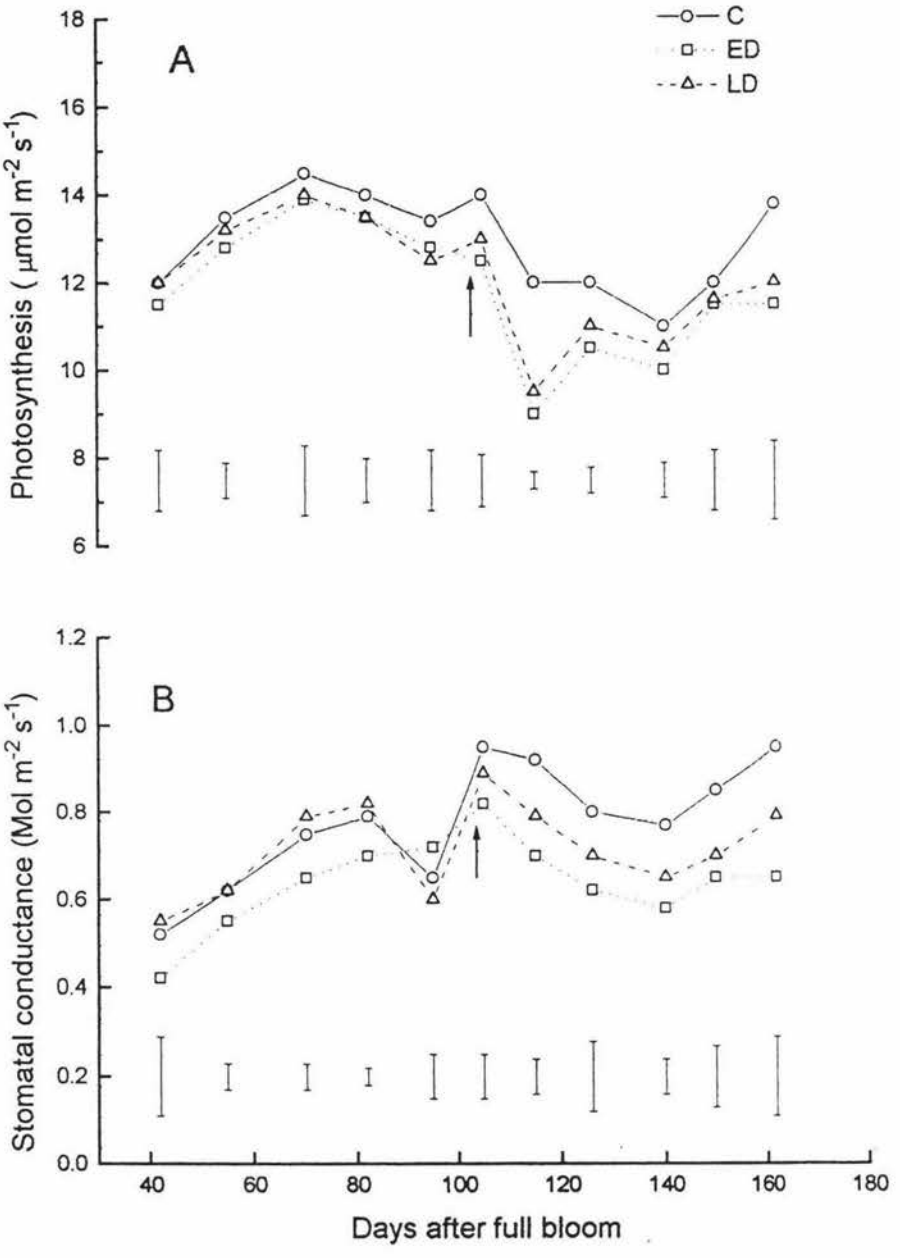

Figure 4. Changes in A) rate of photosynthesis and B) stomatal conductance during the season for control (C), entire-season deficit (ED), and late-season deficit (LD) treatments. Arrow indicates the start of LD. Vertical bars represent the pooled standard errors of means based on four replicates per treatment.

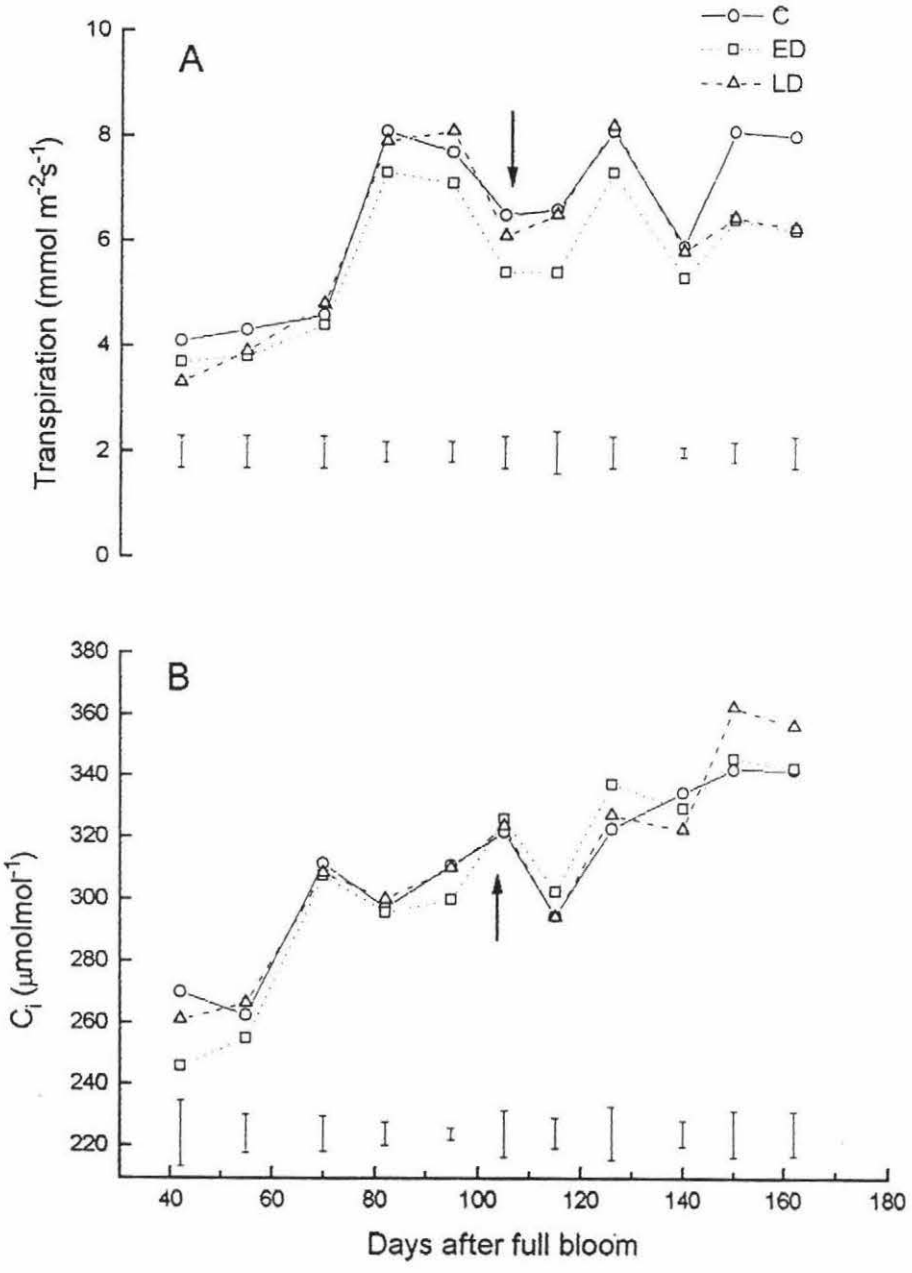

Figure 5. Changes in A) rate of transpiration and B) leaf internal CO_2 concentration(C_i) during the season for control (C), entire-season deficit (ED), and late-season deficit (LD) treatments. Arrow indicates the start of LD. Vertical bars represent the pooled standard errors of means based on four replicates per treatment.

Differences in T_c-T_a were observed earlier (before 102 DAFB) below than those of T_c (Fig. 6B). At 116 DAFB, higher T_c-T_a was observed in LD and ED than C . Furthermore, the values for ED and LD were positive while in C showed negative values. Positive values mean that the canopy is warmer than the surrounding air. Positive values for ED and LD were maintained up to the last day of measurement, 179 DAFB, except on 150 DAFB when values for ED and LD fell below 0. However, there was still a significant difference in T_c-T_a between treatments.

Seasonal canopy temperature increased in the stressed plants from about 140 DAFB (Fig. 6A). This closely corresponds to the decrease in g_s and the subsequent decrease in the rate of transpiration for the water stress plants. Canopy-air temperature differences values were generally higher in stressed plants than those of well-watered plants. This means well-watered plants were cooler than stressed plants.

These results are consistent with the idea that a well-watered plant transpires at its maximum potential rate resulting in leaf temperatures lower than the air temperature and that as water deficit increases (Ψ decreases), transpiration declines and the leaf temperature rises relative to the air temperature (Jackson, 1982). Regression analysis of Ψ on both T_c ($y = 19.55 + 5.55\ x + 3.39x^2$, $r^2 = 38.28$ %, $n = 48$) and T_c-T_a ($y = -2.56 + 1.64\ x + 0.69\ x^2$, $r^2 = 39.25$ %, $n = 48$) indicated a significant ($P \leq 0.01$) quadratic relationship between canopy temperature data and Ψ. As water stress increased, both the T_c and T_c-T_a increased in a quadratic manner.

4.1.6 Vegetative growth

Leaf area in apple develops mostly during the first two months of the season (Palmer, 1988). Once the canopy is developed, water stress will have minimal influence on leaf area, unless the water deficit is severe enough to cause leaf abscission (Castel and Fereres, 1982). Table 1 shows no reduction in leaf area occurred when water deficit was induced in the late season. The only decreasing trend was for leaf area in the ED trees which were stressed throughout the season.

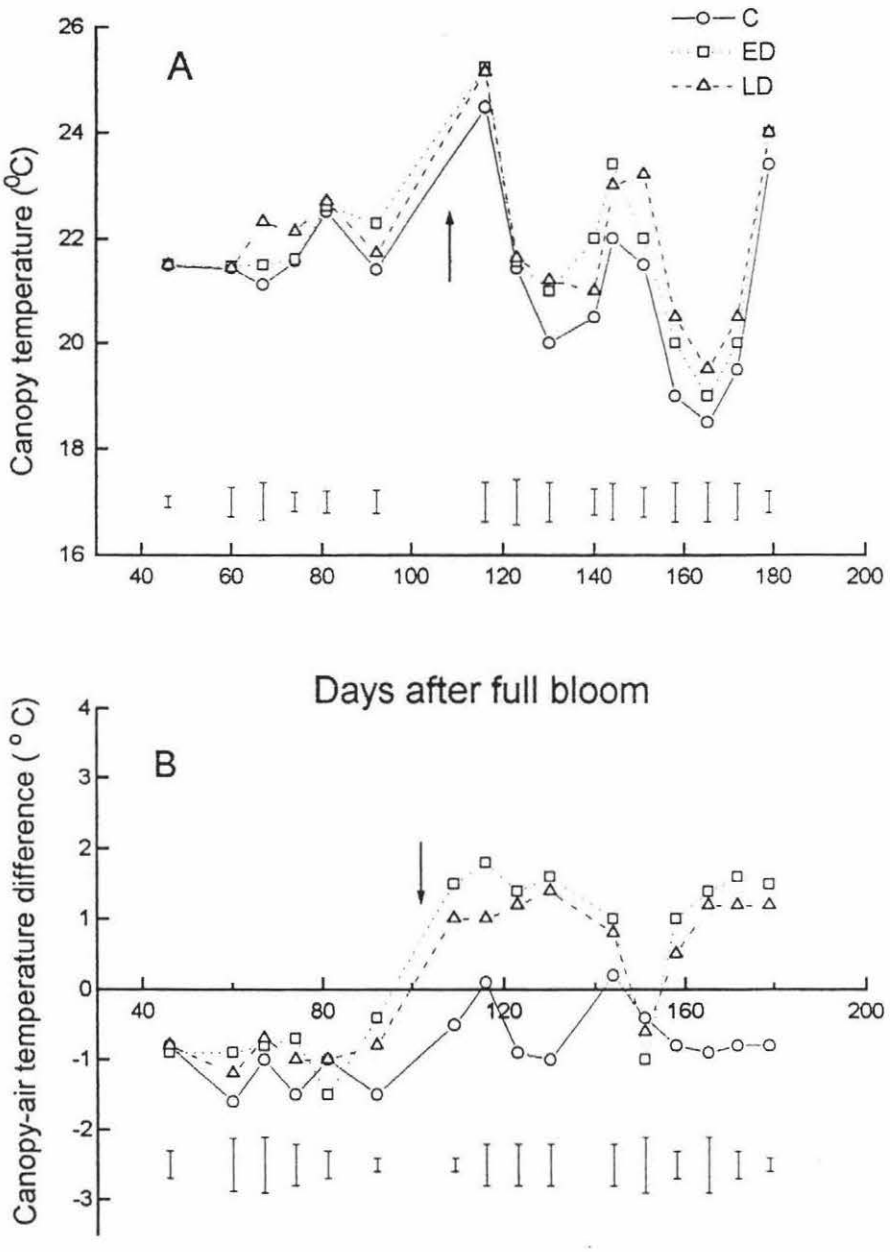

Figure 6. Changes in A) canopy temperature and B) canopy-air temperature difference during the season for control (C), entire-season deficit (ED), and late-season deficit (LD) treatments. Arrows indicate the start of LD. Vertical bars represent the pooled standard errors of means based on four replicates per treatment.

The largest increase in trunk circumference was observed in C while the lowest was in ED (Table 1). Deficit irrigation during the late season had no effect on increase in trunk circumference whereas ED decreased it. Increase in trunk circumference was decreased by 25% in ED compare to C. Trunk growth is related to the above ground weight of the tree (Westwood and Roberts, 1970) and trunk circumference area (TCA) increases consequently with tree growth (Higgs and Jones, 1991). Overall, in this study, vegetative growth of the trees was significantly reduced in ED but not in LD.

Generally, shoot growth was rapid up to 100 DAFB, thereafter, it was slower (Fig. 7). However, there was a higher cumulative shoot growth in LD than in C, and ED. Shoot growth was increased following DI, this increase was consistent with less negative Ψ of these DI trees at the time growth was taking place (Chalmers et al., 1986). Clear difference in shoot growth between treatments were observed from the first day of the measurements 45 DAFB. On average, total shoot length was reduced by about 25% between treatments. Reduction in vegetative growth as a result of reduced plant water status has been reported previously in apples (Ebel et al., 1995; Failla et al., 1990 ; Higgs and Jones, 1991). The results obtained in the current study agree with those of Failla et al. (1990) who found that water deficit in 'Granny Smith' apples affected shoot growth only in the early season, during which the shoots were still actively growing.

Although the reduction in growth by water stress has been well documented, different physiological processes have been put forward to account for this reduction in the different species. Chartzoulaksi et al. (1993) found a reduction in plant height of 78% to 84% by severe water stress in kiwifruit. These authors attributed the decrease to a reduction in photosynthesis, leaf area development, and photosynthate partitioning. In this study, it is unlikely that reduced vegetative growth in ED was a consequence of reduced photosynthesis because Pn was not reduced until much later in the season (152 DAFB) by which time shoot growth had ceased. Indeed Pn was not

affected at all by ED, yet a reduced shoot, leaf area and trunk growth was recorded in this treatment.

Different physiological reasons have been put forward to account for the reduction in vegetative growth in response to water stress. According to Kozlowski et al. (1991), reduced growth is that of reduced cell expansion and division. Kriedemann (1986) indicated that cell division could be inhibited by low water potential as a result of reduced supply of photoassimilates while cell enlargement is hampered by low osmotic potentials. An alternative explanation is that plant growth regulators such as auxins, ABA, and cytokinins or the ratio between these are influenced by low moisture content in the soil (Cleland, 1986). The growth regulators may be involved in growth either directly or through their influence on the hydraulic conductivity of cell membranes and cell wall loosening (Turner, 1986).

Table 1 Growth and return bloom for control (C), entire-season deficit (ED), and late-season deficit (LD) of 'Braeburn' apple trees. Column values followed by the same letter are not significantly different at 5% level.

Treatment	Leaf area (m^2)	Trunk growth (mm)	Return Bloom (no. of flowers per tree)
C	2.49±0.6a	14.5±0.41a	268±15.2a
ED	2.45±0.6a	11.9±0.41b	201±15.2b
LD	2.48±0.6a	13.7±0.41a	228±15.2a

Mean±SE (four replicates per treatment)

4.1.7 Fruit growth

The results in figure 8 show the cumulative fruit growth determined from 45 DAFB. The fruit diameter at this date was approximately 35 mm for all treatments. There was no difference in fruit growth in any of the treatments until final harvest. The mean fruit diameter across the treatments was 82 mm at the time of harvesting. After 102 DAFB, ED fruit diameter was slightly lower than the other two treatments, though the difference among them was not significant.

Final fruit size of apple is influenced by fruit cell number (Denne, 1961). Fruit cell number is determined by cell division which may be reduced under water deficit (Hsiao et al., 1976). Therefore water deficit during the cell division phase of fruit growth may reduce cell number and result in smaller fruit. In apples, cell division occurs from full bloom up to about 40 DAFB (Westwood, 1993, p.258). This period may be important in determining final fruit size. For this reason water deficit was not imposed in this study until the cell division phase of fruit growth was completed.

Although reduced plant water status, severe enough to reduce photosynthesis, was imposed on LD, no reduction in fruit growth was recorded. This is in agreement with data on Asian pears (Behboudian et al., 1994) and apple (Irving and Drost, 1987) which showed no reduction in fruit size when water stress was imposed late in the growing season. In the current study LD started at 102 DAFB by which time the fruit were past cell division phase.

Return bloom was significantly reduced in ED but not in LD relative to C (Table 1). Final trunk circumference was used as a covariate in the analysis of return bloom. In apples, floral initiation for next year's crop occurs in early summer (Westwood, 1993, p.258). This corresponds to late December and early January in the southern hemisphere. During this time, ED trees were not receiving irrigation and water stress may have affected floral bud development and hence return bloom. According to Faust (1989), flower bud differentiation is one of the processes adversely affected by water stress. In this study the reduction in return bloom was not

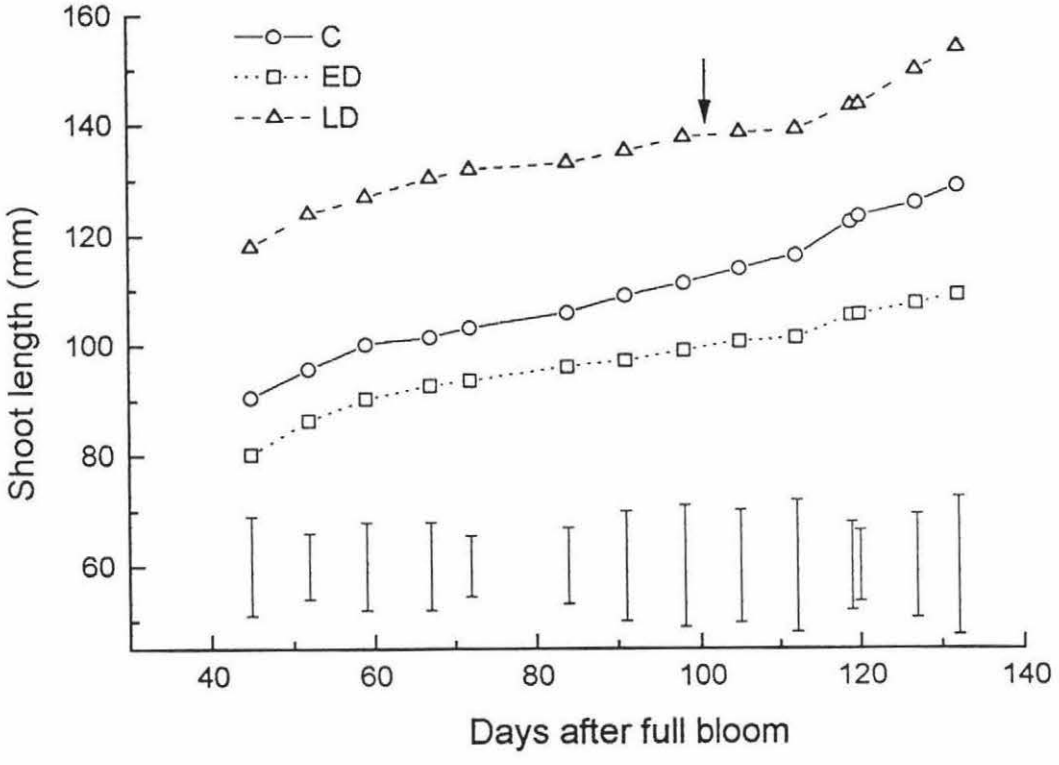

Figure 7. Changes in shoot length during the season for control (C), entire-season deficit (ED), and late-season deficit (LD) treatments. Arrow indicates the start of LD. Vertical bars represent the pooled standard errors of means based on four replicates per treatment.

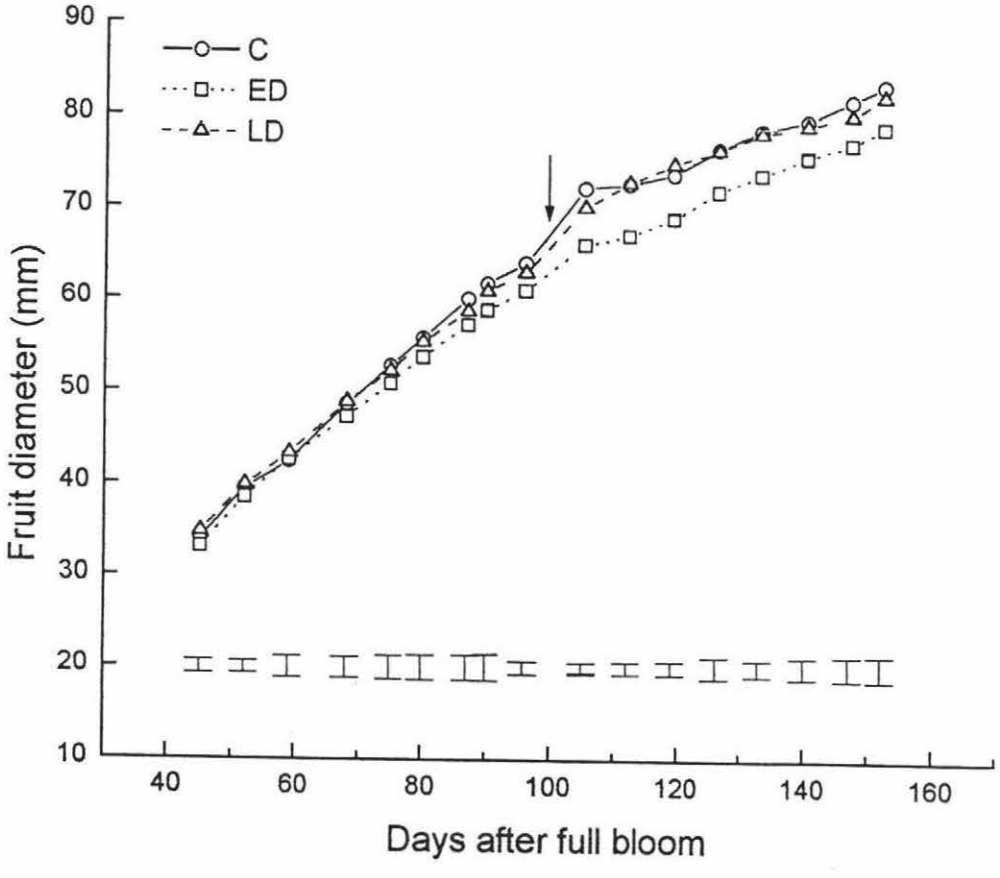

Figure 8. Cumulative fruit growth during the season for control (C), entire-season deficit (ED), and late-season deficit (LD) treatments. Arrow indicates the start of LD. Vertical bars represent the pooled standard errors of means based on four replicates per treatment.

so severe because further thinning was needed to attain commercially desirable crop loads. Therefore the reduction in return bloom may be potentially beneficial in terms of reduced costs of hand and chemical thinning.

4.1.8 Shoot vs Fruit growth

This study shows shoot extension is clearly more sensitive to water stress than the fruit growth in 'Braeburn' apples. According to Forshey and Elfving (1989), fruit growth is less sensitive to water stress than the other above ground portion of the tree. This phenomenon is the basis of the benefits obtained by DI. Chalmers et al. (1986) reported that DI reduces vegetative growth with increase in fruit yield. The mechanism is based on the phenomenon that different tissues and organs have different sensitivities to reduced plant water potential. For example, peach and pear fruit growth was not inhibited by the water deficit induced by DI, although vegetative growth occurring at the same time was reduced 52% (Mitchell and Chalmers, 1982). According to Hsiao (1973), photosynthesis and translocation are also less sensitive to Ψ than cell expansion in vegetative tissue. Thus fruit may continue accumulating solutes although plant Ψ may be too low to allow cell expansion.

This study has shown that DI can be successfully applied to 'Braeburn' apple as a mean of vigour control without reducing fruit growth. Benefit in terms of reduced vegetative growth was only obtained when irrigation was withheld in the early season (Before 102 DAFB), rather than later in the season. The implications of the treatments on the fruit quality are discussed in the following section (4.2).

The following is a summary of the main findings and conclusions. The use of under-tree covers combined with a DI was effective in reducing the soil moisture content. The DI trees developed a lower predawn and midday leaf water potential relative to the well-watered control. Water stress affected various physiological processes such as stomatal conductance, photosynthesis, and the rate of transpiration. The effect of water stress on the above mentioned processes generally occurred late in the season. Although there was a strong relationship between g_s and Pn, there was

evidence that the decrease in the g_s was not the sole factor responsible for decreased Pn. Leaf internal CO2 during the season was not affected by the treatments at any stage suggesting that there could have been an impairment of the carbon fixation mechanism hence the observed reduction in the rate of photosynthesis regardless of stomatal conductance.

Fruit growth was not inhibited by DI. But shoot growth and trunk circumference were greatly reduced by ED which also caused reduction in fruit weight at harvest (Table 3) and a significant decrease in return bloom (Table 1).

4.2 FRUIT QUALITY

Deficit irrigation can influence fruit quality (Ben Arie and Lurie, 1986). It is the potential for improvement in fruit quality, together with a saving on irrigation costs, which makes DI an effective management method.

4.2.1 Flesh firmness

Firmness at harvest was similar regardless of irrigation treatment. Differences in fruit firmness were observed between treatments only after storage (Table 2). There was a general decrease in firmness after storage. Differences in firmness of approximately 5 N were observed between ED and C after storage. Fruit softening occures with increased maturity and has been used as an index of maturity (Kingston, 1992), this study found a higher firmness in LD and ED fruit than C fruit. To compare with other results have indicated that LD and ED fruit were more mature.

However, the data was insufficient to conclude the effect of water deficit on fruit firmness. Westwood (1993, p.304) concludes that firmness is not a good maturity index for apples because it does not relate to well to other changes that signify maturity.

Table 2 Firmness for control (C), entire-season deficit (ED), and late-season deficit (LD) of 'Braeburn' apples. Column values followed by the same letter are not significantly different at 5% level.

	Firmness (N)	
Treatment	At harvest	After storage
C	96.16±5.13a	83.18±2.1a
ED	94.70±5.13a	88.18±2.1b
LD	95.47±5.13a	89.42±2.1b

Mean±SE (four replicates per treatment)

4.2.2 Total soluble solids

Soluble solids percentage was higher in the ED and LD treatments than C at harvest, and after storage (Table 3). A difference of about 1.6% was observed between treatments at harvest. An increase in TSS of apple fruit has been reported under water deficit (Irving and Drost, 1987; Ebel et al., 1993). Guelfat'Reich et al.(1974) suggested that fruit from water stressed trees may have been more mature at harvest than those from trees receiving ample water hence differences in TSS. Proebsting et al. (1984) reported increased TSS in deficit irrigated 'Delicious' and 'Golden Delicious' apples to advanced maturity of the fruit relative to the well-watered control. In the current study, there is evidence of more advanced maturity in LD and ED than in C in terms of changes in ethylene concentration and the skin colour. However, there are other more likely reasons. At harvest, there was increased dry matter concentration in ED and LD than C. The dry matter concentration (%± SEM) for C, ED, and LD fruit was 14.11± 0.09, 15.8± 1.07, 15.21± 1.13, respectively. Hence dilution effects could have led to increased TSS values in ED and LD at harvest.

4.2.3 Titratable acidity

Titratable acidity (% malic acid) declined with time during storage regardless of pre-harvest treatment (Fig. 9), but level of TA was not affected by treatments. Titratable acidity declines as the fruit matures due to the utilization of organic acids as carbon skeletons for the synthesis of new compounds during ripening (Biale and Young, 1981). Our results are similar to those obtained by other workers (Drake et al., 1981; Irving and Drost, 1987) who found that reduced irrigation did not affect TA.

4.2.4 Mineral composition

The changes in fruit mineral composition at harvest and after storage are shown in Table 4. Fruit mineral concentration affects the quality and storability of apple fruit as it influences the development of disorders both at harvest and during storage. Calcium concentration were the same between treatments at harvest as well as after storage. This was expected since Ca^{2+} uptake into the fruit predominantly occurs in the early stage of fruit expansion (Ferguson and Watkins, 1989). This is prior to the development of water deficit in the ED treatment.

A reduction in N content of the ED fruit, compared to the other two treatments was observed at harvest as well as after storage. Nitrogen levels are important to the storage quality of apples (Bramlage, 1993). Higher N associated with larger fruit will be associated with poorer colour development and more predisposition to the development of cork spot, bitter pit, internal breakdown, scald, and soggy breakdown. Richardson (1986) observed that an increased N concentration in apple fruit resulted in larger fruit which were softer and had a greener background colour. Bramlage (1993) also pointed out that many of these changes either relate to increased fruit N concentration, and/or an accompanying reduction in fruit Ca^{2+} concentration associated with increased tree growth under high nitrogen. Additionally, an increase in fruit size under High N conditions effectively dilutes fruit Ca^{2+} concentration. No differences were observed between treatments for the concentration of Mg^{2+}, P, and K^{+}.

Table 3 Influence of irrigation treatment on some fruit attributes for control (C), entire-season deficit (ED), and late-season deficit (LD) of 'Braeburn' apples. Column values followed by the same letter are not significantly different at 5% level.

TRT	Fruit weight	Fruit diameter	Total soluble solids	Colour	
				Lightness	Hue angle
	(g)	(mm)	(%)		(⁰)
At harvest					
C	283.72±9.1a	85.08±1.04a	13.67±0.29a	45.60±1.08a	47.17±0.7a
ED	248.01±9.1b	82.62±1.04b	14.66±0.29b	47.35±1.08b	51.42±0.7b
LD	263.83±9.1a	84.07±1.04a	14.75±0.29b	48.55±1.08b	55.68±0.7b
After storage					
C	275.25±6.2a	83.07±1.6a	14.61±0.27a	46.15±1.23a	45.9±3.1a
ED	236.60±6.2b	80.66±1.6b	15.53±0.27b	47.28±1.23b	49.5±3.1b
LD	257.12±6.2a	82.06±1.6a	15.71±0.27b	47.89±1.23b	51.64±3.1b

Mean±SE (four replicates per treatment)

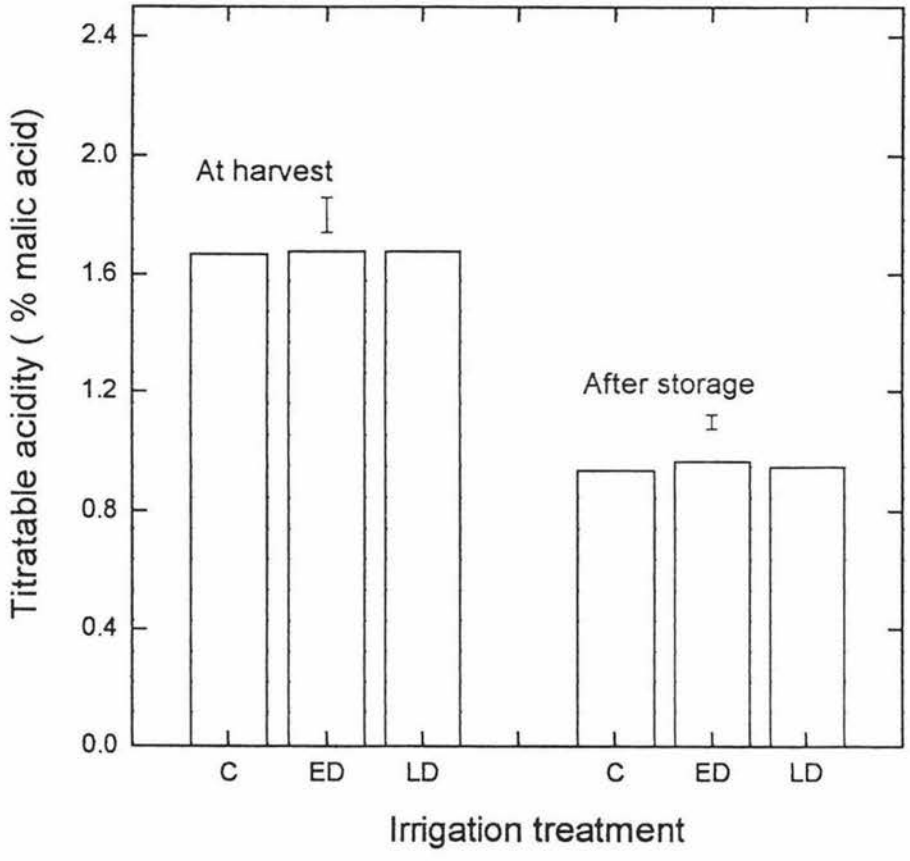

Figure 9. Titratable acidity for control (C), entire-season deficit (ED), and late-season deficit (LD) treatments. Vertical bars represent the pooled standard errors of means based on four replicates per treatment.

Table 4. Changes in the concentration of fruit minerals (mg g^{-1} dry wt) for control (C), entire-season deficit (ED), and late-season deficit (LD) of 'Braeburn' apples. Column values followed by the same letter are not significantly different at 5% level

Treatment	Ca^{2+}	Mg^{2+}	K^{+}	N	P
At harvest					
C	0.02±0.01a	0.02±0.1a	1.20±0.6a	0.27±0.09a	0.12±0.04a
ED	0.02±0.01a	0.02±0.1a	1.20±0.6a	0.17±0.09b	0.12±0.04a
LD	0.02±0.01a	0.02±0.1a	1.20±0.6a	0.24±0.09a	0.12±0.04a
After storage					
C	0.02±0.01a	0.03±0.01a	1.65±0.1a	0.13±0.1a	0.14±0.04a
ED	0.02±0.01a	0.03±0.01a	1.65±0.1a	0.08±0.1b	0.14±0.04a
LD	0.02±0.01a	0.03±0.01a	1.65±0.1a	0.10±0.1a	0.14±0.04a

Mean±SE (four replicates per treatment)

4.2.5 Skin colour

At the time of harvest and after storage lightness (L) and hue angle (H) values were lowest in C and higher in ED and LD (Table 3). Te colour of the apples is the result of a blend of various amounts of different pigments. The red colour on the blushed side of some apple strains is produced by anthocyanins and flavonols which are presented in vacuole (Lancaster, 1992). Lower values of H and L indicate a redder and darker skin and these correspond with a higher anthocyanin concentration (Singha et al., 1991). Thus the results obtained in the current study indicate that fruit from the water deficit had a lower anthocyanin content than those from the control. Previous discussion in the literature review (2.3.3) indicated that N may play a role in green colour of apple fruit. In this study, even though N concentration was affected by treatment but no correlation between green colouration and N concentration was observed.

4.2.6 Ethylene evolution and CO_2 production

Ethylene evolution is reported as a good indicator of apple fruit maturity (Watkins et al., 1989). Data presented by Watkins et al. (1989) indicate that ethylene evolution in 'Braeburn' apple is relatively low, when compared with cultivars such as 'Cox's Orange Pippin', 'Royal Gala', and Red Delicious'. The changes in fruit ethylene concentration and CO_2 production at harvest and after storage are shown in Figure 10 and 12, respectively. Fruit from LD show higher levels of ethylene concentration compare to C at harvest. Ethylene evolution is much in after storage compare to at harvest. Rate of CO2 production accompanied by rise in rate of ethylene. Respiration was higher at harvest in all treatments compared to after storage, accompanied by softening, development of aroma and a pattern of ethylene production (Fig. 12). Pratt and Reid (1974) found that the ethylene-induced respiratory response of harvested with decreased with increasing storage time. Apple fruits show greatly differing rates of ethylene production at different stages of their postharvest life increasing to about 1000 fold as fruit ripen (Biale and Young, 1981;

Knee, 1993). Results from the ethylene concentration measurements indicate that the fruit from ED and LD treatments enter the climacteric phase of ethylene production earlier than C. In general, stressed plant tissue produce higher ethylene (Abeles et al., 1992, p.266). Previous studies have shown that water stress at different stress period of fruit growth not only increases the rate of ethylene production, but also result in the fruit reaching an earlier climacteric phase. Ebel et al. (1993) found that 'Delicious' apples grown under DI showed a significant increase in ethylene evolution when compared to control fruit. In a study by Guelfat'reich et al. (1974) it was observed that in apple fruit exposed to water deficit during development, ethylene production was greater after harvest than it was in fruit from the well- watered treatments. These results indicate that water stress may advance fruit maturity as determined by the time required to reach the ethylene climacteric phase.

4.2.7 Volatile compounds

Concentration of volatlile compounds in the fruit juice of 'Braeburn' apples showed a great variation between treatments (Fig. 11). The concentration of alcohols (ethanol, methanol, butanol, pentanol, proponol, hexanol, 2*3 methyl butanol), aldehydes (trans-2-hexanal, hexanal), ethyl esters (ethyl acetate, ethyl propionate, ethyl butyrate, ethyl-2-methyl butyrate, ethyl 3 hydroxy butonoate, ethyl 3 hydroxy hexanoate, ethyl pentonoate, ethyl pentonoate), and non-ethyl esters (propyl acetate, butly acetate, hexyl acetate, amyl acetate, 2 methyl butyl acetate, propyl butyrate, hexyl hexanoate) were higher in water stress trees compared to well-waterd trees at harvest as well as storage. Ethyl 2-methylbutyrate, hexanal, and 2 hexanal are essential to the apple aroma (Flath et al., 1969). Concentration of the volatiles appear to be affected. Brackmann et al. (1993) indicated that high CO_2 concentrations supress the production of aroma compounds. In this study, CO_2 production was observed lower in the stressed treatments than control. Inhibition of ethylene inhibits repiration and the production of aroma volatiles (Halder and Doll, 1987).

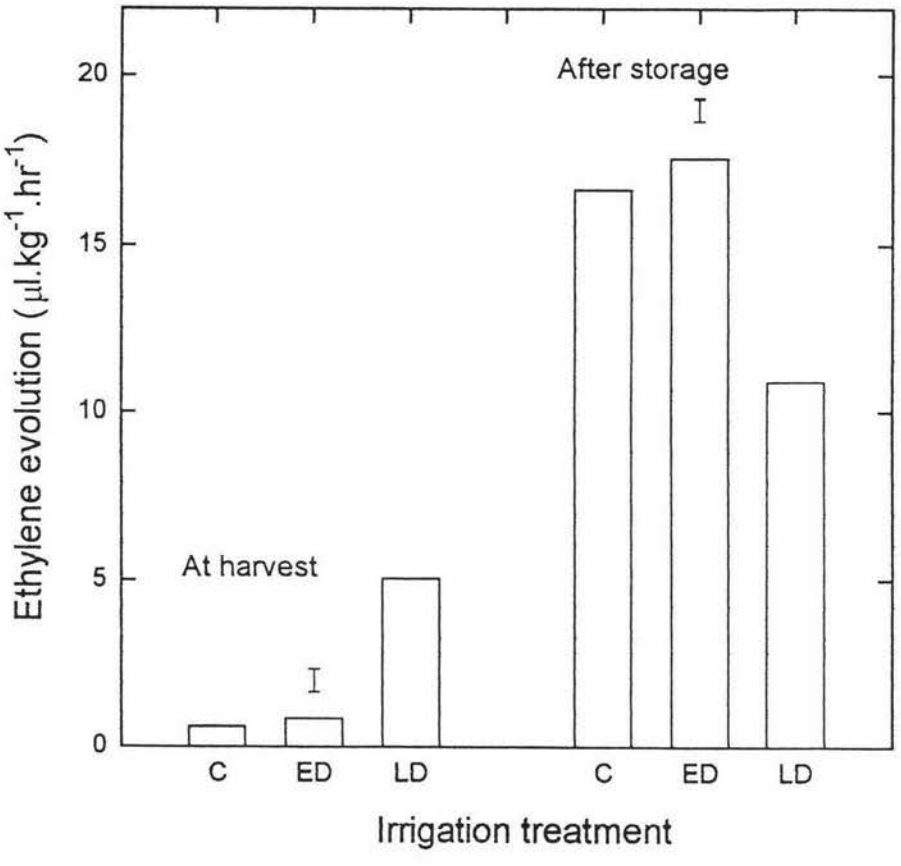

Figure 10. Ethylene evolution for control (C), entire-season deficit (ED), and late-season deficit (LD) treatments. Vertical bars represent the pooled standard errors of means based on four replicates per treatment.

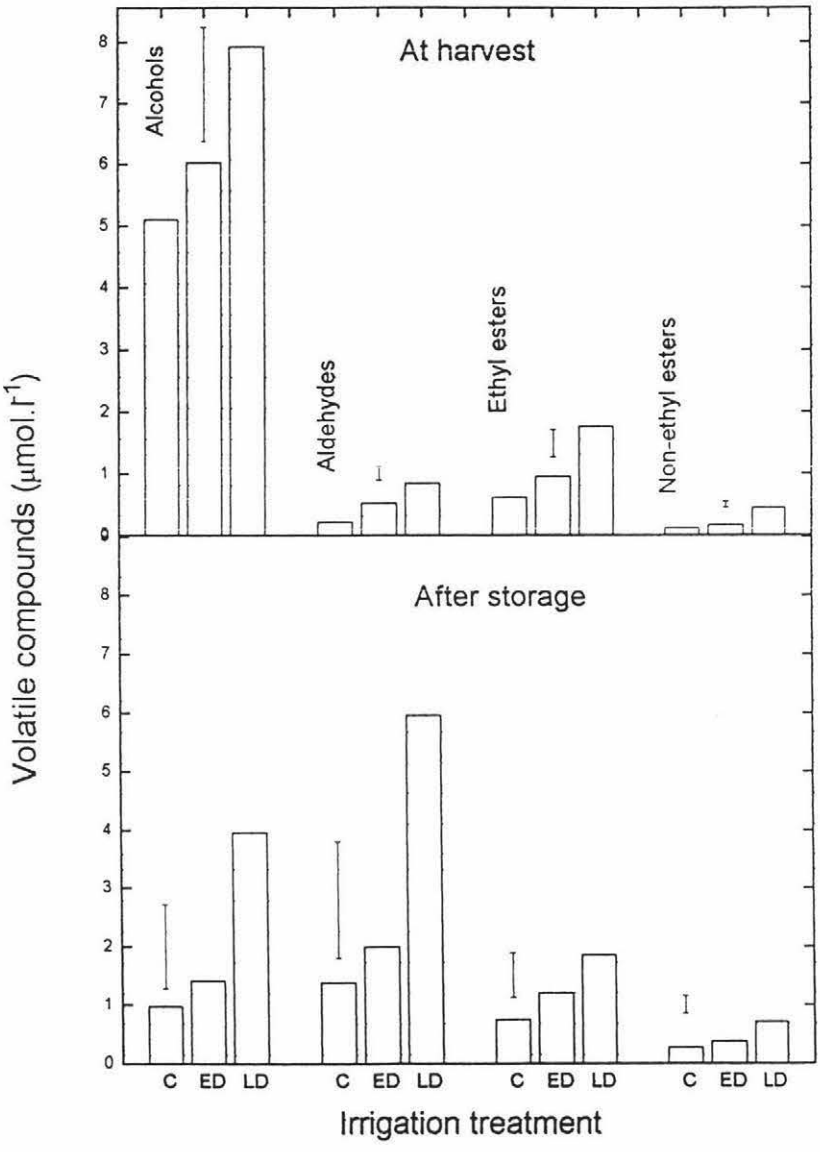

Figure 11. Concentration of volatile compounds in the fruit juice for control (C), entire-season deficit (ED), and late-season deficit (LD) treatments. Vertical bars represent the pooled standard errors of means based on four replicates per treatment.

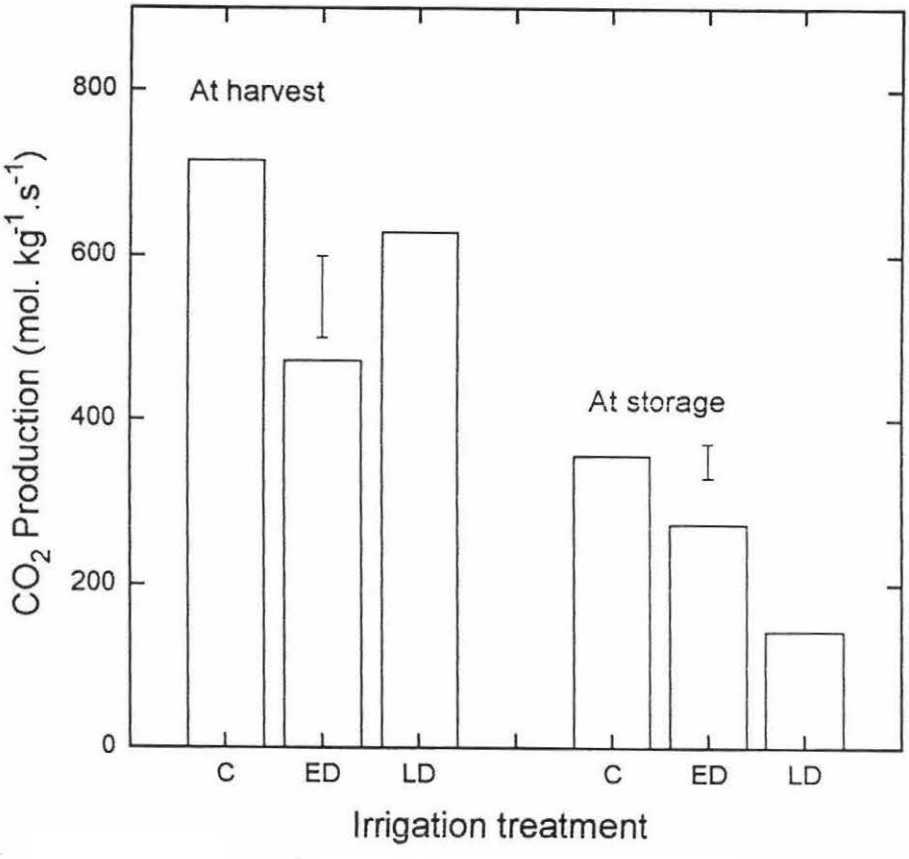

Figure 12. Carbon dioxide production for control (C), entire-season deficit (ED), and late-season deficit (LD) treatments. Vertical bars represent the pooled standard errors of means based on four replicates per treatment.

Harris et al. (1972) suggested that the fruit may be undergoing a slow ripening process during storage. If aroma compounds were released during this slow ripening process they would be lost and this could be explain the lower levels of aroma compounds during stored fruit.

However, this data is insufficient to conclude the effect of water deficit on fruit volatile concentration. There is a need for further studies to investigate the effects of irrigation timing and frequency on the flavour and aroma of the fruit.

The following is a summary of the main findings and conclusions, from this part. Fruit composition and quality attributes were evaluated at harvest and after storage. The time at which irrigation is withheld has important implications on the fruit quality attributes in 'Braeburn' apples. Deficit irrigation during late season and entire growing season led to increase in TSS, but did not affect TA. Flesh firmness was increased in LD and ED after storage than in C. Increased firmness is due to difference in fruit size and also probably due to a reduction in cellular hydration. Further research is needed in this area especially to determine the water potential of the fruit and its relationship with firmness. The concentration of Mg^{2+}, Ca^{2+}, K^{+}, and P was not affected by the treatments, but concentration of N was lower in the ED fruit. Considering the importance of the fruit mineral concentration especially Ca^{2+} on the keeping quality of the fruit, it is likely that under reduced irrigation treatments the storage life of the fruit is not affected. Withholding irrigation during the late and entire growing season caused advanced maturity and earlier ripening of 'Braeburn' apples as indicated by changes in ethylene concentration, TSS. Volatile concentration. were higher in LD and ED fruit than C fruit. These results may have implications on the ideal harvest time since harvest before climacteric is considered best for apples destined for long-term storage (Abeles et al., 1992, p. 266).

CHAPTER FIVE

GENERAL DISCUSSION AND CONCLUSION

Application of DI has been successful in peaches, pears and grapes in climates which experience insufficient rainfall during the growing season (Chalmers et al., 1981; Jerie et al., 1989). DI has resulted in a reduction in vegetative growth early in the growing season with no lasting effect on fruit growth. Resumption of irrigation later in the season allowed fruit growth to proceed normally with overall yields not being detrimentally affected by these stress treatments (Chalmers et al., 1984).

To achieve soil moisture deficit early in growing season in major fruit growing areas in New Zealand, it will be necessary to develop some method to prevent water entering the soil profile or else to create some system whereby the water in the soil is removed quickly in spring and early summer. New Zealand has a humid climate, the heavy rains that fall during the winter being sufficient to bring the orchard soil to field capacity. In the study by Durand (1990), DI was supplemented with lucerne cover to increase the rate of moisture depletion. In the current study, the approach was to install under-tree covers to prevent rainfall from reaching the soil and to ensure a water deficit. This approach was effective as evidenced by lowering of θ and Ψ as early as 45 DAFB. As the season progressed, there was a gradual soil moisture depletion in ED such that the midday Ψ levels as low as -2.0 MPa were recorded some days before 102 DAFB. LD was applied in early February 1996 (102 DAFB). Moisture depletion and decrease in Ψ for LD was observed to be more rapid than that of ED. At the end of growing season, differences of 0.7 MPa were observed between non-irrigated and irrigated trees. Previous studies on DI have obtained similar differences. For example, Mills et al. (1994) recorded differences of 0.7 MPa between nonirrigated and irrigated trees during the late season. These differences were sufficient to cause differences in fruit quality and trunk growth.

Reduced plant water status affects various physiological processes and organs (Kozlowski et al., 1991). However, different plant tissues, organs and processes have different sensitivities to water stress (Chalmers,1989). In the current study, fruit growth was not affected by reduced irrigation from 45 DAFB up to harvest, however, there was a large reduction (as much as 50%) in shoot growth of 'Braeburn' apples under ED. Ebel et al (1995) observed a reduction in vegetative growth due to water stress without reduction in yield or fruit size in 'Delicious' apples. The same observation was made for peaches by Chalmers et al. (1981) and for pears by Mitchell et al. (1986). This is one of the physiological bases for the beneficial effects of DI (Chalmers, 1989). Shoot growth is more sensitive to reduced Ψ than that fruit growth and therefore irrigation can be reduced to a level of Ψ that affects shoot growth without adversely affecting fruit growth. This may have an advantage in cases where assimilates no longer being used by shoot growth are diverted to fruit growth leading to increased yield and fruit size (Chalmers et al., 1986). Taking advantage of this phenomenon, however, needs proper management of DI.

Studies on kiwifruit have indicated that reductions in the rate of photosynthesis and photoassimilate partitioning are the main reasons of reduced growth under reduced irrigation (Chartzoulaski et al., 1993). This is unlikely in the current study because a reduction in the rate of photosynthesis occur much later in the season by which time shoot growth had ceased. A more likely reason for shoot growth elucidated by Chalmers (1989) is that cell expansion, being very sensitive to reduced Ψ, is adversely affected. Fruit cells expand by a similar mechanism as shoot cells, however, fruits are stronger assimilate sinks even under reduced Ψ.

A reduction in g_s was observed in ED and LD from 140 DAFB. Therefore g_s does not respond rapidly to reduction in Ψ. Similar results were obtained by Mills et al. (1994) who found a reduction in g_s from 140 DAFB.

A higher transpiration in the irrigated plants had 'cooling' effect on the canopy as shown by the infrared thermometry data. The regression between Ψ and canopy temperature data indicates that there is a good potential for the use of infrared

thermometry to assess water status in deciduous fruit crops. It offers advantages such as being easy to use and its non-destructive. However, there is a need for further investigation in relation to irrigation scheduling of deciduous fruit crops, especially in a humid climate.

Although it is widely accepted that water stress causes a reduction in the rate of photosynthesis, the actual mechanism involved is not clear. A reduction in Pn has been attributed to stomatal closure (Farquhar and Sharkey, 1982), reduced enzymatic activity (Vu and Yelenosky, 1988), or accumulation of photoassimilate in the leaves (Janoudi et al, 1993). In this study, internal CO_2 concentration did not decrease in DI leaves and although the regression analysis of Pn and g_s showed g_s was reduced in stressed plants, an impairment of the photosynthetic machinery might have occured. Similar conclusions were reached by Behboudian et al. (1994) who found increased C_i/C_a ratio in water stressed Asian pears and attributed this effect on impairment in carbon fixation at the chloroplast level.

Water stress adversely affects flower bud initiation and differentiation (Faust, 1989, p.159). This was manifested in the current study by a reduction in return bloom in ED trees. water stress in these treatments occurred at the time at which flower initiation and differentiation occurs i.e. late December and early January. This affected flower initiation and hence return bloom in the following season. A reduction in return bloom may, to some extent, be horticulturally desirable as it reduces thinning costs.

The second part of the experiment evaluated the fruit quality of 'Braeburn' apples under different irrigation regimes. In general fruit from reduced irrigation treatments developed increased TSS and flesh firmness. Ebel et al. (1993) attributed increased firmness in DI fruit to their smaller size hence increased cellular density. Other factors may have also been involved because fruit from ED were generally smaller than C while those of LD were not comparatively smaller than C and yet they were firmer. This suggests that fruit size was not a contributing factor to increase in the firmness in this case. It is therefore speculated that decreased cellular hydration as

a result of reduced irrigation may have caused the increased firmness. In this study, firmness was not a good measure of maturity as it did not relate with other maturity indices such as ethylene concentration and background skin colour.

At harvest ED and LD fruit had less yellow colour than C. Withholding irrigation in the late and entire season led to an earlier ethylene climacteric. These treatments had shown less yellow background after storage, coupled with increase TSS. Other workers have also reported that water stress leads to early fruit maturity (Ebel et al., 1993; Guelfat'Reich et al., 1974).

Many physiological disorders which develop during the storage of apples have been attributed to a low Ca^{2+} concentration in the fruit (Shear, 1975). In 'Braeburn' apples one of most important of these diorders is bitter pit (Kupferman, 1994). In this study, no differences in concentration of Ca^{2+} among treatments were observed. This implies that there may be no risk of decreased Ca^{2+} related disorders in fruit due to reduced irrigation. Indeed, during 12 weeks of storage, no calcium related disorders were observed in the fruit from any treatment. The trees were exposed to commercial Ca sprays during the season. This might have masked any possible negative effects of DI on Ca content.

In conclusion, the experiment presented in this thesis reduced plant water status in a humid and high rainfall environment. This was done by reduced irrigation and covering of the soil. The results present some dilemma as far as recommendations to apple growers are concerned. Shoot extension and trunk growth results suggest that for vigour control, the response is best if irrigation is withheld during the early season.

On the other hand, for improved fruit quality in terms of increased TSS, skin colour, firmness, and increased volatiles the results suggests that withholding irrigation late in the season is more appropriate. Furthermore, these quality attributes are improved without loss of yield or fruit size. The use of LD also depends on the postharvest storage requirements. LD tends to cause advanced maturation and hence

earlier ripening. This may mean that the storage life of the fruit is essentially reduced since the aim of postharvest technology is to delay ripening and senescence as much as possible. LD also affects the optimum time of harvest. Apples for long term storage are best harvested before climacteric (Abeles et al., 1992, p. 266). Thus LD may advance the harvest dates of the fruit. This may be desirable if fruit are available early in the season and command premium prices. This finding also provides evidence that the potential exist for the growers to manipulate the probable harvest time for apples by manipulation of the water status of the plant through a properly managed irrigation strategy. This would be beneficial in terms of controlling the fruit volumes in the market.

Differences in skin colour, firmness, flavour and aroma of the fruit were observed in this experiment, and it opens some new channels to further research. One is the possible use of infrared thermometry in irrigation scheduling for deciduous crops. There is need for studies aimed at a more precise recommendation of the best time of the withholding irrigation and the threshold levels of plant water status that is beneficial in terms of both improved fruit quality and the control of vigour without effects on yield or fruit size. Withholding of irrigation for a long period of time may be detrimental in some environments, and there is a need for comparative studies in areas with low humidity and rainfall conditions. Further, research is needed to find the response of flavour and aroma of the fruit by use of chromatography techniques for the analysis of flavour volatiles and sensory evaluation to determine the acceptability of the fruit produced under reduced irrigation in the market.

Overall, this study has demonstrated that reduced irrigation strategy can be used on apples successfully as an effective management tool.

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