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**Rural Industries Research and
Development Corporation**

Olive Harvest

**Harvest timing for optimal olive
oil quality**

**A report for the Rural Industries Research and
Development Corporation**

by Rodney Mailer, Damian Conlan and Jamie Ayton

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Foreword

Olive oil quality is known to show varietal differences and also to be influenced by environmental conditions from site to site and from year to year. However, the greatest differences occur during the maturation period, from young green olives to the final dark coloured fruit. During this period the oil quality will gradually change and this change will be greater than the variation between cultivars, sites or environments. The variation during this period therefore can be influenced by grower management.

This is the first Australian study of environment, cultivar and seasons to develop an understanding of the changes that take place during olive fruit maturity and the timing of those events. These results will allow the grower to monitor specific crop characteristics in order to optimise harvest timing for production of desired outcomes such as increased yield or specific oil quality characteristics as well as improving oil stability for better shelf life.

In this study we have utilised a grove in southern NSW and three olive cultivars over three years to observe changes in the major quality parameters that influence final oil quality and yield. The protocols for monitoring yield and quality development in ripening fruit that we have used here can be applied to any other grove in Australia where a technical approach to harvest management is being adopted.

This report details findings through the stages of fruit maturity from soft stone to black olives. It describes the range of factors individually, as well as the relationship with other parameters, which are important for the product to be considered high quality oil. Indicators are provided to identify the time to harvest fruit to optimise particular traits such as oil yield, oil quality, sensory characteristics and good shelf life stability.

This project was funded from industry revenue which is matched by funds provided by the Australian Government.

This report, an addition to RIRDC's diverse range of over 1,200 research publications, forms part of our New Plant Products R&D program, which aims to facilitate the development of new industries based on plant and plant products that have commercial potential for Australia

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Abbreviations & Description

AOCS	American Oil Chemists' Society
IUPAC	International Union of Pure and Applied Chemistry
IOOC	International Olive Oil Council
EVOO	Extra virgin olive oil
Pomace	residue after oil has been extracted
Must	oil/water mix from olive extract
GC	gas chromatography
HPLC	high performance liquid chromatography
G	Full irrigation
Y	Moderate irrigation
R	Deficit irrigation

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Executive Summary

The rapidly developing Australian olive industry and the large number of trees currently planted will result in an oversupply of olive oil for the Australian market in the near future. If producers are not aware of the factors that influence quality aspects of their oil, they will be limited to competing with highly subsidised, cheap imports with quality equivalent to current supermarket retail standards. It is necessary for Australian producers to concentrate on producing very high quality oil to gain a share of domestic and export olive oil markets.

Considerable variation exists between olive oils due to varieties, environments and growing areas but perhaps the greatest variation comes from the maturity of the olive at the time of harvest. As olives mature the oil passes through a range of quality levels in which many of the components are changing. At the same time the oil content is increasing but in many cases the ability to extract the oil will pass through an optimum period before the oil becomes difficult to recover.

This is the first scientific study of its type to relate harvest timing to olive oil quality and stability in Australia. The results provide growers with valuable information on how to achieve specific quality in their olives by targeting fruit maturity. These findings show that the greatest variation in oil quality and oil yield between the cultivars studied is due to fruit maturity. As a result a grower may choose to produce pungent and peppery oil or a mellow to bland oil. The opportunity is there to select oils with increased shelf life stability when antioxidants are at a high level. The oil may be processed when it is green or left to golden yellow. Even the level of polyunsaturated fatty acids can be influenced by monitoring fruit quality and selecting the optimum harvest date.

This study has identified the changes in many of the oil components during the stages of change. The information gained will provide growers with the tools to improve their oil yield by optimising the harvest time and water application. Olive oil sensory characteristics such as pungency, bitterness and fruitiness, may also be determined by harvest timing. Shelf life stability is also sensitive to fruit maturity and is of critical importance to the product to ensure it retains quality long after the oil is bottled.

An olive grove based in Southern NSW was utilised for this study. The grove with 100,000 four year old trees included three common cultivars, Mission, Corregiola and Paragon. Furrow irrigation was established to allow a comparison of irrigation treatments. Environmental conditions during the three years of this project were less than ideal for an irrigation study. Drought conditions persisted over the entire period and water application was limited. All of the trees suffered different levels of water stress. The majority of parameters measured therefore showed no significant differences between high irrigation, moderate irrigation and severe deficit irrigation. Despite this, the study provided considerable information on soil water movements and the effects of harvest timing on some of the numerous parameters tested.

A major aim of the project was to relate physical and easy to measure parameters with oil quality and oil recovery. It is necessary to define for the grower, precise times when the fruit are at their peak and before the quality begins to decline. The tests traditionally used to identify quality in the field include fruit colour, weight, detachment force and hardness. All of these characteristics were monitored over the three years of this study, at regular dates throughout the maturation period. Maturity index (MI), the most common European method to determine fruit condition, was shown here to provide little reliable information. Fruit of different cultivars changed colour at different times. In fact, the variability of fruit colour within cultivars, and even within individual trees ranged from green to black (MI 3 to MI7). While fruit were still green oil quality and yield were changing. Although the skin of Corregiola turned black, the flesh often remained green throughout. Reliance on this character under Australian conditions would lead to late harvesting and loss of fruit and oil

quality. Variations in MI from year to year, despite consistent oil accumulation over the same time, show that MI would not produce consistent product.

It is known that the growth of the fruit follows a similar pattern to oil accumulation and has been discussed in the report (4.2.1). As such, it should be possible to design harvest timing on fruit weight. Fresh fruit weight in olives was shown to be variable, increasing and decreasing with rainfall or irrigation events. The fruit take up water when it is available and release it when it is limited. Fresh fruit weights were useless in determining fruit development, rising and falling across the growth period. A more useful tool was found to be the dry weight, which was calculated from fruit moisture contents. Fruit dry weight followed oil production closely, producing similar curves and levelling out as fruit weight reached its maximum. The limitation was that fruit weight continues to increase for a period after optimum oil content has been reached. As a result, reliance totally on this method would result in late harvests with reduced fruit yield. Fruit firmness also showed useful trends with similarities to oil accumulation. Firmness dropped rapidly at point when oil content had reached a maximum. This test has potential for further application.

It is more difficult to apply chemical tests to determine harvest timing as they are often expensive and time consuming. By the time chemical analysis has been carried out, the fruit may have missed the optimum time. This analysis is however necessary to help growers relate physical testing to oil quality under their own conditions. Chemical testing is recommended for individual conditions, at least in the initial years of generating a crop. Moisture is variable over the growing period but generally the total moisture will drop as the fruit reaches maturity. This was not always the case however and Mission tended to hold moisture levels throughout the growing period. Paragon and Corregiola also showed variation from year to year. It is not likely therefore to be a reliable indicator for harvest timing.

Solvent extraction of the oil from olives is the required method to compare oil contents between harvests, treatments or cultivars. It removes 100% of the oil and can show when the oil production has reached a maximum. This is also useful when comparing the efficiency of cold-pressed systems and determining how much oil has been lost in the process. It does not indicate however what the cold-press extraction will yield. Cold-press extraction is the best indicator of when to harvest as it imitates the oil extraction process in commercial situations. It was found that cold-press extraction reached a plateau well before the olive had ceased producing oil. This is due to the changing properties of the olives which make the oil harder to extract. It is therefore necessary to harvest before the solvent extraction would indicate to get the maximum oil recovery.

Harvest timing may be targeted toward producing particular quality traits in the oil rather than just trying to increase yield. This report describes the major quality components and the changes which take place over the growing period. The major contributor to oil quality is the polyphenol content. This plays a strong role in determining the stability of olive oil. The close relationship with shelf life stability and induction time has been discussed. Organoleptic quality is also dependent on polyphenols which determines pungency and bitterness. The high polyphenols shown to be present in young olives rapidly decreases as the fruit mature. The oil concurrently changes from strong pungent oil to a light mellow product. The relationship between induction time and several other components in the oil, including fatty acids and chlorophyll has also been discussed.

The fatty acid profile is important in determining oil stability but also in the oils nutritional quality. Oleic acid is considered beneficial and high levels are encouraged. Although oleic acid changed little over the maturity period, linoleic acid (polyunsaturated) was found to increase while palmitic acid (saturated) decreased. Higher levels of polyunsaturated fatty acids are nutritionally beneficial but reduce oil stability. Harvest timing can be used to select for stability or nutritive value. Linolenic acid also decreased with maturity. This component is unstable and can increase the rate of oxidation. However, in Australia it has been found that linolenic acid levels often exceed international standards, thus making it a possible problem in international trade. The linolenic acid needs to be below 1.0% which is usually achieved early in the maturity process. Free fatty acids must be less

than 0.8% in extra virgin olive oil. We have shown that overripe olives can exceed this limit while still on the tree. It is necessary to harvest, particular cultivars such as Paragon, before free fatty acids increase. Peroxide value was not found to be a problem and did not change during maturation.

Chlorophyll, which provides the green colour to oil, has some benefits. However, when exposed to light, chlorophyll becomes a pro-oxidant and can contribute to oil instability. The rate of chlorophyll reduction with maturity is discussed and decisions can be made to select for early harvested green oil or late oil of lighter colour but better stability. Tocopherol, which decreases with maturity, also acts as an antioxidant and provides stability to the oil.

As olive production increases as a result of the massive plantings that have taken place in recent years, olive oil sales will be more competitive. Although yield is of major importance, oil character and quality are gaining recognition. Already, olive oil competitions are in abundance in every state in Australia with many commercial bottles now carrying medals to indicate the quality standards. Fruit stability is also gaining importance and oil that oxidises quickly will be unacceptable to consumers. Also of major importance are the nutritional benefits of monounsaturated oil with reduced levels of saturated fatty acids. Growers and processors can have significant controls over all of these factors through monitoring crop maturity and adapting harvesting and processing management to suit their requirements.

Despite having achieved most of the goals of this study, it was apparent that there are more factors which need to be better understood. The irrigation system at this grove was of limited value to the study as it was furrow irrigation and difficult to monitor and apply at regulated levels. As a result, comparisons between deficit and adequate irrigation were not achieved. More needs to be understood about the polyphenolic compounds in the oil which provide the majority of resistance to oxidation. The individual responses of each cultivar to the rancimat test indicate that each was under the control of variable antioxidants. It is likely that the sensory characteristics are also controlled by these individual polyphenol profiles. The stability of the oil and the maintenance of good organoleptic characteristics, both with significant relationship to harvest timing, are probably the most important aspects of olive oil. This study has shown that despite the differences between olive cultivars and growing conditions, the greatest changes between olive oil quality characteristics are a result of the fruit maturity and harvest timing.

1. Introduction

1.1 Background

More than 9.5 million olive trees have been planted in Australia since 1994 and substantial production from new olive orchards is about to commence (Miller 2002). The Australian olive industry will face an oversupply situation within three to four years unless significant exports can be achieved.

Unless growers produce extra virgin olive oil (EVOO) that is better than the quality of current supermarket retail standards they will be confined to competition with highly subsidised, cheap imports. Large scale, highly mechanised production of EVOOs is the principal technical challenge facing the Australian olive industry. Information on managing crops through precise crop water management and harvest timing will advance the ability of Australian growers to consistently produce oils to high quality specifications, and through this improve domestic and export market opportunities.

It is well understood by the Australian olive industry that it has to produce EVOO to gain a share of domestic and export olive oil markets. In some cases, the quality standards of Australian olive oils are poor, even by general retail standards. A recent Choice magazine article (Choice 2004) was highly critical of some Australian olive oil, describing one as "very poor". The demand for technical information on management strategies that optimise yields without compromising quality will increase. The ability of the Australian olive industry to succeed will be dependent on two general principles: economical production and high quality.

Time of harvest can have a profound influence on the yield, quality, style and stability of olive oil (Ayton *et al.*, 2001). However there are few clear guidelines that producers can use to determine the optimum harvest period. This study advances our knowledge of the chemical and physical changes that take place during fruit ripening and from this we can define monitoring parameters that will assist olive producers to determine optimum harvest time.

1.2 Variability

Olives harvested from new orchards in Australia in past seasons have produced highly variable yields ranging from 5% to 30% oil by weight. Low yields have been attributed to a range of reasons such as incorrect variety, immature trees, harvesting too early, high fruit moisture contents and poor extraction efficiency. There is now significant demand for technical information on management and environmental factors that can be used to improve yield and quality.

There is general agreement that high quality oil requires precise harvest timing of good quality fruit although in-field determination of when to harvest can be very difficult. Numerous studies in the Mediterranean have shown that during the ripening period, oil percentage increases dramatically during early fruit ripening (Salvador *et al.*, 2001, Beltran *et al.*, 2004). It then slows as full ripeness approaches and declines slightly as fruit becomes over ripe. Oil quality improvement is concurrent with increasing oil content but peaks, and begins to decline, before maximum oil yield is reached. Fruit detachment force declines steadily as fruit ripens. It then drops sharply when fruit reaches full ripeness and fruit drop increases. Fruit colour changes from lime green to pale 'straw' green as fruit matures and changes colour to purple and then black as it becomes fully ripe. The rate, evenness and time of ripening will vary between varieties, environments, seasons and management practices. Studies from Europe into management practises such as harvest timing are often highly specific in terms of location and local genotypes and therefore of limited use for broader interpretation and application (Giametta 1992, Rahmani *et al.*, 1997, Tombesi 1990).

1.3 Previous studies

Data generated from a previous RIRDC Project, UCS-19A - Assessment of olive oil quality and cultivar identification (Robards and Mailer 2001), showed that the range of maturity times between cultivars, and the variation in quality at different stages of maturity was significant. Although the determination of optimum harvest was beyond the scope of that project, the published data established that oil content and fatty acid profiles vary considerably during development. There has been considerable discussion about the changes in oil quality with harvest timing (Ayton *et al.*, 2004, Mailer *et al.*, 2004) although there have been only limited studies on the effects in Australia.

Identification of olive cultivars in Australia has been a problem with many cultivars showing variations within the cultivar (Mailer and May 2002). This has been shown also in other studies (Sedgley 2004) in which trees identified as a particular cultivar are distributed across several branches of dendrograms produced from cluster analysis. The variation within Australian cultivars has also been established (Mailer *et al.*, 2002., Mailer 2004) with a wide range of fatty acid profiles, oil contents and polyphenol content which relates closely to the oils sensory characteristics.

Other studies (Caponio and Gomes 2001) have indicated that fruit maturity influences the organoleptic characteristics of the oil and the ultimate oil stability. Studies under Australian conditions are required to pinpoint the stage of maturity, and morphological indicators that the grower might use to determine the quality and style characteristics obtained from harvesting fruit at particular times during ripening. Harvest monitoring guidelines would focus on achieving customer specifications for extra virgin olive oil.

The maturity index is a mathematical measure of the degree of ripeness of the fruit which is still commonly used in Europe (Shimon Lavee *pers. com.*) to determine when to harvest fruit. The method uses a visual colour assessment of 100 randomly selected olives to obtain a numerical value of between 1 to 7. The proportions of each category are then calculated as described by Boskou (1996). This method is useful where oil yield is the only consideration, but it does not provide an accurate guide where oil quality is an important consideration.

1.4 Oil-stability

There is substantial commercial emphasis placed on the chemical and organoleptic stability (shelf life potential) of extra virgin olive oils (EVOOs). EVOOs vary substantially in stability between varieties, time of harvest, crop management and climate. This is related to the level of antioxidants in the oil, predominantly polyphenolic compounds. These compounds are also implicated with nutritional benefits for consumers in addition to providing the pungent sensory characteristics in olive oil. The assessment of stability is an important component of olive oils which must remain at a high quality during storage and prior to consumption. Other components reported to contribute to stability are chlorophyll, tocopherols and fatty acid profiles, particularly polyunsaturated fatty acids. These issues have been included in this study.

1.5 Irrigation

Management of irrigation requirements for olive orchards in Australia has been derived from Californian, Israeli and South Australian studies where the principle focus has been to maximise fruit yield. Irrigation guidelines describe a crop water requirement for olives of 9 to 11 megaliters per hectare (Fernandez and Moreno 1999). Studies on irrigation from Europe have a generally pointed towards the necessity of full irrigation to maximise production from olives, yet recent work by Goldhamer (1999) suggests that significant crop water use deficits can be imposed without loss of oil yield. When combined with Australian experience it shows full irrigation can result in high fruit moisture contents, reduced extraction efficiency and reduced chemical and organoleptic quality.

Benefits in quality and yield can be gained from careful management of irrigation during fruit development and ripening periods (d'Andria *et al.*, 1999, Goldhamer 1999, Moltiva *et al.*, 1999, Tovar *et al.*, 2002).

1.6 Industry support

Determination of harvest and irrigation management practices requires direct involvement of industry and research groups. Irrigation management, harvest timing and extraction efficiency are currently areas of high priority for the Australian Olive Association. Olive growers have demonstrated the immediate relevance of the research through extensive discussion and debate in email discussion lists and industry journals.

This project will be enhanced by the direct involvement of the Nugan Group Pty Ltd, an industry leader that focuses on high quality, and large scale mechanised production of EVOO. Adoption of the findings will not require changes to infrastructure but rather changes in management that can be used by all olive growers. The adoption of regulated deficit irrigation practices being assessed in this project will contribute to significant reductions in olive orchard water use.

2. Aims and Objectives

The aim of this project has been to determine the optimal harvest timing for olives at which to achieve maximum oil quality, extraction efficiency and organoleptic characteristics for olive oil. The study identifies and explains the changes that take place during maturity stages in olive fruit from the onset of ripening through to full ripeness. A wide range of chemical quality characteristics are used to describe fruit quality. Fatty acid profiles, free fatty acids, peroxide values and rancimat tests indicate quality characteristics. The comparison between solvent extraction and cold press extraction will provide efficiencies of extraction over the ripening period and between varieties and water management treatments.

Field guidelines for harvest parameters such as detachment force, uniformity of ripening, fruit mass and fruit maturity index through the fruit ripening period are developed for the assessment of fruit to indicate optimal harvest. The project also assesses the effects of harvest timing and irrigation on chemical and organoleptic stability of olive oils and extraction efficiency. The project will define for the first time in Australia, the timing of harvest for olives and crop water management strategies for high quality olive oil production. Methods estimate ripeness and quality of oil which can be produced from a particular harvest time given the prevailing seasonal conditions and management practices used.

3. Materials and Methods

3.1 Wagga Wagga Oil Research Laboratory

The Wagga Oil Research Laboratory (WORL) has been involved with the analysis of edible oils since 1995 and has become increasingly involved in olive oil analysis over the last 10 years. The facilities at Wagga encompass oil research, commercial oil analysis, education and recently organoleptic testing. The laboratory holds several accreditations including accreditation by the IOOC to test olive oil. In addition the WORL has ISO 9002:1994 certification and also holds NATA accreditation for several oil testing methods. The laboratory has held approved chemist status from the American Oil Chemists' Society for many years. Generally all of the methods used are IOOC methods where available. For some tests, such as oil and polyphenols content, the methods are adopted from other sources such as the American Oil Chemists' Society Standard Methods.

3.2 Grove Site and Trial Plan

The site for this project was the Nugan Group Pty. Ltd. property 'Cookathama', a commercial olive grove situated at Darlington Point (longitude 146°0', latitude 34° 57'), 33km south of Griffith in south-western New South Wales. The 'Cookathama' olive orchards were planted in the period from 1997, to 2001. They are modern young orchards containing approximately 80 thousand trees, the oldest of which have reached commercial production age. The layout, large scale of plantings and uniformity of field conditions was ideal for biometrical analysis of separate cultivars, unlike the majority of established olive groves in Australia.

The grove contains four main cultivars, Mission, Paragon, Corregiola and Leccino. Three of these Mission, Paragon and Corregiola were selected to be included in this study due to the grove layout and the differences between the cultivars. Each of the three cultivars had been planted in individual blocks, and to aid pollination, every tenth row was planted to *cv.* Pendolino, which flowers early and abundantly, and has a lengthy flowering period. Trees were planted in a 5 metre by 8 metre square grid pattern in rows 400 metres long. The trees used in the trial are located in a block of about 11,000 trees, planted in 1997-8.

The trial design was a randomized block with three replications (Fig. 1). Six rows of trees were included in each of three blocks identified as replicate 1, 2 and 3. The six rows included three treatments: full irrigation, partial irrigation and nil irrigation and a buffer row between each treatment. The trees were watered using furrow irrigation, with the furrows approximately 1.5 metres from the centre of the trunk on each side.

3.3 Irrigation

The trial was located on a large area of medium to heavy self mulching cracking grey clay loam soil. Prior to experimental design the trial area was surveyed using an EM31 device along with soil sampling and texturing. As a result of the survey, the trial area was located to maximise uniformity of soil type and depth.

Irrigation was applied through furrows positioned approximately 1.5 metres outside of the tree line. The furrows and under tree area were maintained in a weed free state through the use of herbicides while the inter-row area was planted to fescue and perennial rye grass. Irrigation water was applied by syphons with each watering event applying approximately 0.5 megalitres per hectare, the equivalent of 50 mm of rainfall. There was variability in the supply of water within each row due to variations in furrow depth and distance from the trees.

Irrigation treatments on olive orchards on Cookatharma were given lower priority for watering in the first two years due to water limitations caused by drought. A further complication was the failure of the gypsum blocks, installed to monitor soil moisture. In year three of the project distinct irrigation treatments were applied, but again water availability for irrigation was restricted resulting in no irrigation being applied beyond mid March 2004. Irrigation was deficient during late fruit growth and oil synthesis and fruit maturation periods.

Three irrigation treatments were applied. The treatments were: high irrigation (G), moderate deficit irrigation (Y) and severe deficit irrigation (R). The high irrigation regime received a total of six irrigations between November 03 and Mid March 04, applying approximately 3 megalitres of water to the crop. At an irrigation efficiency of 80% this represents about 2.4 megalitres of plant available water being applied. The moderate deficit treatment was watered 4 times while the severe deficit treatment was watered three times, supplying total water volumes of 1.6 and 1.2 MI/ha respectively.

3.4 Soil moisture monitoring.

Gypsum blocks: A total of 108 heavy soil gypsum blocks supplied by TAIN electronics were installed across the trial area. The blocks were located at 30, 60 and 90 cm depth at 1 metre and two metres distance from the base selected trees in each treatment. The blocks did not function satisfactorily. There were two principal reasons for this: firstly, the deficit treatments involved drying the soil to fairly extreme levels resulting in cracking and shrinkage which led to loss of soil-gypsum block contact. The blocks were often slow to re-wet and many ceased to work at all. The second reason was that the root system of the trees was concentrated in the top 50 cm and blocks located at 60 cm depth were below most of the roots and the depth of water infiltration on an average irrigation event. Only in the event of substantial rainfall events or watering and rainfall combined was there sufficient prolonged wetting of the gypsum blocks at 60 cm for them to re-wet. The tendency of the soil is to shrink and crack when dry, rendering the gypsum blocks ineffective and their use was discontinued.

Neutron Probe: Neutron probe tubes were installed in September –October 2003 and the soil water was monitored with a model 503 Hydroprobe on a weekly basis from October 2003 through to May 2004.

Neutron probe tubes were located in one replication of each irrigation treatment of each variety. The tubes were installed 1.2 metres from the base of the tree and 0.5 m from the centre of an irrigation furrow. The probe was calibrated at the time of tube installation and the calibration equation was applied to readings taken over the course of the irrigation period.

Readings were taken at depths of 10, 20, 30, 40, 50, 70, 90, 105 cm below the soil surface. This provided a detailed picture of water movement in the soil and the depth of penetration of soil moisture following irrigation or rainfall events. It also provided information on the zone of root activity in the soil from the varying rates of soil moisture depletion from different depths. Soil moisture depletion from various soil depths following irrigation was recorded.

3.5 Sample collection

Olives were harvested according to statistical design (Fig. 3.1). At each of four harvests, a total of 3kg fruit was taken from ten trees within each of three replicates and for each cultivar. Trees were harvested only once so as not to change natural conditions which may influence the next harvest.

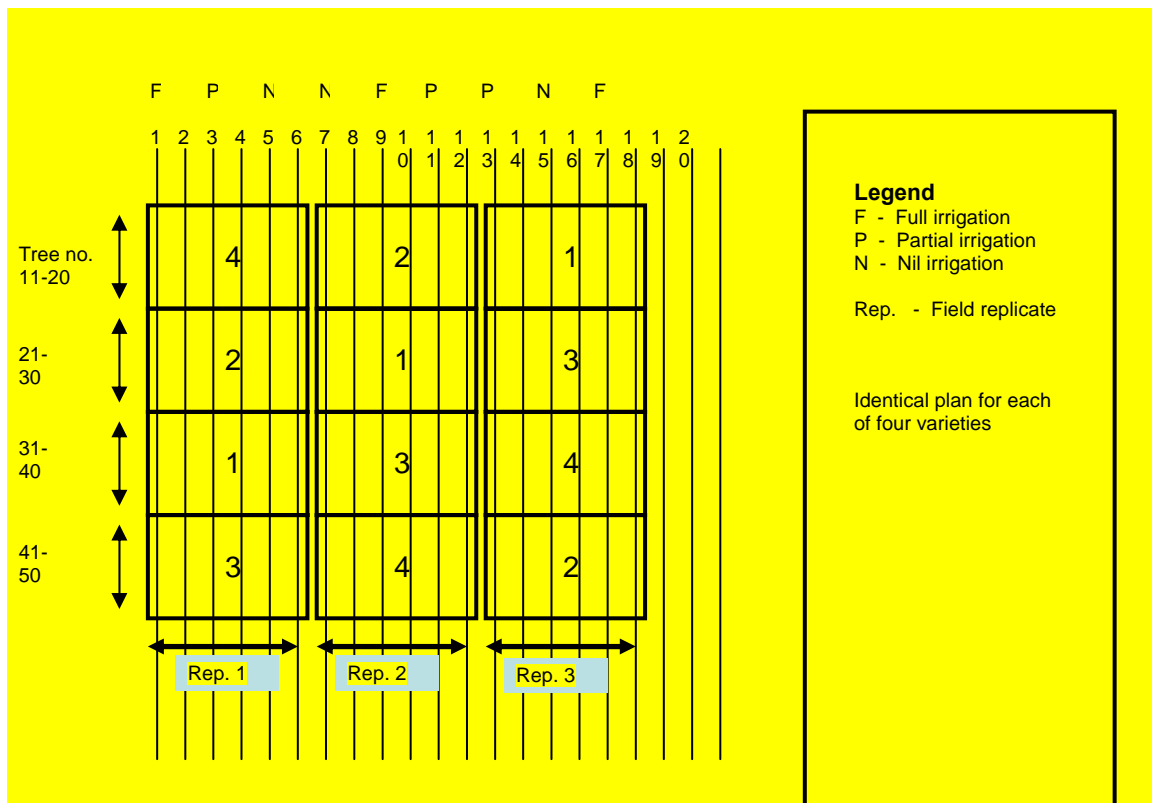
In addition to the four main harvest dates, two initial “early” harvests of 300 g of fruit were carried out in mid to late February and mid to late March, to provide additional data on fruit size and weight, maturity index, solvent extracted oil content, moisture content and fatty acid profile

Four harvests were carried out:

- (1) prior to the normal commercial harvest window (early April),
- (2) and (3) during the normal commercial harvest period (early to mid April and late May) and
- (4) post commercial harvest period (mid July) (Table 1).

Wagga Wagga laboratory staff travelled to the olive grove on the dates shown in Table 1 and sampled olives as described. Physical testing, including fruit removal force, was measured in the field at the time of harvest. Fruit size, weight and all of the chemical parameters were measured in the Oil Research Laboratory in Wagga Wagga. Fruit were packed in cotton bags and packed gently into plastic trays. They were transported back to Wagga on the same day and placed into cool rooms at 8°C until testing. Fruit was processed within four days after harvest. Examples of fruit from each cultivar and at each harvest were photographed for future reference, including external and internal fruit colour and kernel shape.

Fig. 3.1 Harvest Plan for each cultivar. Randomised block designs were used for each of the four harvests undertaken each year.



Analyses carried out on these four harvests included:

- fruit size and weight
- fruit removal force
- maturity index
- solvent-extracted oil content
- cold-press oil content
- moisture content
- fatty acid profile
- total polyphenol content
- free fatty acids
- chlorophyll content
- peroxide value
- induction time
- α-tocopherols – 2004 only
- fruit firmness – 2004 only

Table 3.1 Harvest dates for the years 2002, 2003 and 2004

	2002	2003	2004
Harvest 1	28 February 02	17 February 03	18 February 04
Harvest 2	25 March 02	18 March 03	18 March 04
Harvest 3	8 April 02	7 April 03	13 April 04
Harvest 4	30 April 02	30 April 03	5 May 04
Harvest 5	23 May 02	28 May 03	31 May 04
Harvest 6	8 July 02	15 July 03	12 July 04

3.6 Fruit detachment force (FDF) and fruit firmness

Fruit detachment force was measured on 50 fruit from each ten tree block using a Push-Pull Dynamometer (Effegi, Italy). The force, measured in grams, required to remove the fruit from the tree was recorded. Fruit firmness was measured using the same device with a 1 mm point early in the season and a 1.5 mm point late in the season as fruit became soft.

3.7 Maturity index

The maturity index was determined on 100 randomly selected olives in each sample to obtain a numerical value for the olive sample appearance. Olives were cut in half to expose the internal flesh and to permit grading.

The olives are sorted into categories using the following parameters:

- 0 = skin is a deep or dark green colour.
- 1 = skin is a yellow or yellowish-green colour.
- 2 = skin is a yellowish colour with reddish spots.
- 3 = skin is a reddish or light violet colour.
- 4 = skin is black and the flesh is completely green.
- 5 = skin is black and the flesh is a violet colour halfway through.
- 6 = skin is black and the flesh is a violet colour almost through to the stone.
- 7 = skin is black and the flesh is completely dark.

The total number of olives in each category was counted and recorded. The following equation was then applied to determine the maturity index:

$$\text{Maturity Index} = \frac{(0 \times n_0) + (1 \times n_1) \dots + (7 \times n_7)}{100}$$

where n is the number of fruit with that score (Boskou, D. 1996).

Fig. 3.2 Olive fruit at various stages of ripeness based on the maturity index. It can be seen by M3 to M5 that black skin is not a good indicator that fruit have matured. (MI = maturity index).



3.8 Moisture and oil analysis

3.8.1 Moisture content

Approximately 1kg of fruit was crushed using a hammer mill. After mixing the sample thoroughly, approximately 30 g of paste was transferred to a previously weighed Petri dish. The sample was dried in a fan-forced oven at 80°C for 24 hours. The sample was removed from the oven, placed in a desiccator and cooled to room temperature. The dry weight of the sample was recorded and the moisture content of the fruit calculated as a percentage of the fruit weight.

3.8.2 Oil content - cold press extraction

A cold press extraction unit, similar to that used in the IOOC accredited laboratory, Ministerio de Agricultura Pesca, Y Alimentacion, in Madrid, Spain, was purchased from Abencor, Spain. The Abencor unit imitates the process used by the industry to extract olive oil. It consists of three units: a hammer mill, a thermo-malaxer and a centrifuge.

Approximately 1kg of fruit was ground to a paste using the hammer mill. The sample was thoroughly mixed and 700g of the pulp was weighed into a mixing jar. The jar was placed in the thermo-malaxer and allowed to stir for 20 minutes with the water bath set at 25°C. Following this, 300 ml of boiling water was added to the sample, and stirred for 10 minutes. The sample was then centrifuged and spun for 1 minute. After collecting the “oily must” in a measuring cylinder, the pomace was rinsed with 100 ml of boiling water, centrifuged for 1 minute, and the “must” again collected. After allowing some time for the sample to settle, the quantity of oil was measured and the percentage of oil calculated as “cold pressed oil”.

3.8.3 Oil content - solvent extraction

Solvent extraction is the total removal of oil from each sample. This measurement is reported as a percentage of the dry weight of the olive fruit which removes the influence of moisture variability to allow the trend in oil accumulation across the season to be shown.

Approximately 1kg of fruit was crushed using a hammer mill. The sample was mixed thoroughly and approximately 30 g of paste was placed into a Petri dish, and dried at 80°C in a fan forced oven for 24 hours. The dried sample was then reground using a coffee grinder and transferred to a cellulose extraction thimble. The oil was extracted overnight (16 hours) using a Goldfische extraction apparatus and petroleum ether (100 ml; b.p. 40-60°C). The mass of the oil was determined gravimetrically after removal of solvent. Results are expressed as a percentage of the fruit weight. This is referred to as solvent extracted oil or in industry it would be called pomace oil.

Fig. 3.3 Abencor hammer mill, malaxer and centrifuge used to extract the oil from olives. This is a Spanish system used to replicate industrial scale cold press extraction of olive oil.



3.8.4 Oil recovery

The rate of oil recovery, or efficiency of extraction, was determined by the difference between solvent extracted oil and cold press extracted oil. Solvent extraction determines the total amount of oil in the fruit whereas cold press extraction will imitate the industrial process in which some oil is left in the pomace. Oil recovery is important in determining percentage oil yield per tree and therefore productivity of the grove.

3.8.5 Fruit weight and size

One hundred olives from each sample were randomly selected and weighed, with the average weight of the fruit calculated and recorded. The length and width of fifty randomly selected olives from each sample was measured using Vernier callipers. The average width and length of the fruit at that harvest was subsequently calculated.

3.9 Olive Oil Quality

3.9.1 Total polyphenol content

A modification of the Gutfinger (1981) method, using caffeic acid as the standard, was used to determine total polyphenol content. Oil (10 g) was dissolved in hexane (50 ml) and extracted 3 times with 20 ml portions of 80% aqueous methanol. The mixture was shaken for 2 min for each extraction. The sample was made up to 100 ml with water and left to stand in a dark cupboard overnight. An aliquot (1 ml) was transferred to a 10 ml volumetric flask to which 5 ml of water was added. Folin-Ciocalteu reagent (0.5 ml) was then added and the sample shaken and left for 3 minutes. Saturated Na_2CO_3 (1 ml) was added and the sample shaken again. The sample was made up to volume with water and allowed to stand for 1 hour. The absorption was read at 725 nm. Solutions of caffeic acid were prepared and used to produce a standard calibration curve. The standards were prepared and analysed in the same way as the sample solutions.

3.9.2 Induction time

A Metrohm 679 Rancimat was utilised to determine the induction time of the oil. A block temperature of 130°C and airflow of 20 L/hour was used. Volatile components which develop as a result of oxidation are measured. When the oil begins to oxidise, the change is recorded on the Rancimat. These results were reported as induction time in hours.

3.9.3 Fatty acid profile

Oil (100mg) was mixed with petroleum spirit (3 ml b.p. 40-60°C) in a small test tube. Sodium methoxide (0.5ml, 1.15% sodium in methanol solution) was added, and mixed for 15 seconds. The sample was allowed stand for 10 minutes and bromothymol blue (0.1ml; 0.1% w/v in methanol) was added followed by hydrochloric acid (0.4ml; 1M). Sodium carbonate (0.6ml, 1.5%) was added and the solution mixed thoroughly. Distilled water was added to bring the solvent layer to the top of the test tube and allowed to stand for 5 minutes to allow phase separation. The solvent layer was transferred to GC vials. The fatty acid profile was determined by gas chromatography using a SGE BPX70 capillary column (30m, 0.22mm, 0.25 μ m film) and a flame ionisation detector. The column temperature was programmed at 185°C for 8 minutes then increased at 10°C/minute to a final temperature of 220°C and held for 3 minutes. Total run time was 13.5 minutes. The injector (split mode) temperature was set at 250°C with a split ratio of 1:50. Detector temperature was 260°C. Data was analysed using Star[®] Workstation Chromatography software (Version 6.20).

3.9.4 Free fatty acids (FFA)

Free fatty acids were determined by a modified method of the American Oil Chemists Society (Aa 6-38) (AOCS 1998). Oil (approximately 7.05 g) was dissolved in neutralised isopropanol (about 50 ml). Two drops of phenolphthalein (1% in ethanol) was added to the solution. The sample was then titrated with 0.1N NaOH, previously standardised against HCl. The volume of titrant was recorded and the results calculated as a percentage FFA of the total oil (expressed as oleic acid).

3.9.5 Peroxide value

Peroxide value was determined using the International Union of Applied Chemistry (IUPAC 1992) method 2-501. Oil (2.50 g) was dissolved in acetic acid/chloroform mixture (3:2). To this solution, 1ml of saturated KI (70g KI in 40ml water) was added, and shaken for 1 minute. The sample was then placed inside a dark cupboard for 5 minutes. Water (75 ml) of was added, followed by two drops of starch solution (2.5g starch/100ml water). The solution was titrated with previously standardised 0.01N sodium thiosulphate ($\text{Na}_2\text{S}_2\text{O}_3$). The volume of titrant used was recorded and the peroxide value calculated and reported as mEq of active oxygen/kg oil.

3.9.6 Chlorophyll

Chlorophyll was measured using the method of the American Oil Chemists Society, Ch 4-91 (AOCS, 1998). The absorbance of the oil sample was measured, using dichloromethane as a reference, at 630, 670 and 710nm. The chlorophyll content was then calculated as described in the method and reported as mg chlorophyll/kg oil.

3.9.7 α -Tocopherols

α -Tocopherols were added to the list of analyses carried out in 2004 to further understand the changes that had occurred in induction time, peroxide value and free fatty acids.

Tocopherols were measured using IUPAC method 2-432 (IUPAC, 1992) with slight modification. Oil (2 g) was weighed into a 25 ml volumetric flask, and made to volume with hexane. The samples were filtered and transferred to HPLC vials. The α -tocopherol concentration was determined by HPLC, with hexane/isopropanol (99:1) (stated as 99.5/0.5 in IUPAC method) as solvent with a flow rate of 1 ml/min. A Phenomenex Luna 5 μ Silica column (250 by 4.60 mm) was used. The peaks were measured using a UV detector set at 292 nm. Data were analysed using Waters Empower Pro[®] version 5.00.

4. Results and Discussion

4.1 Climatic conditions

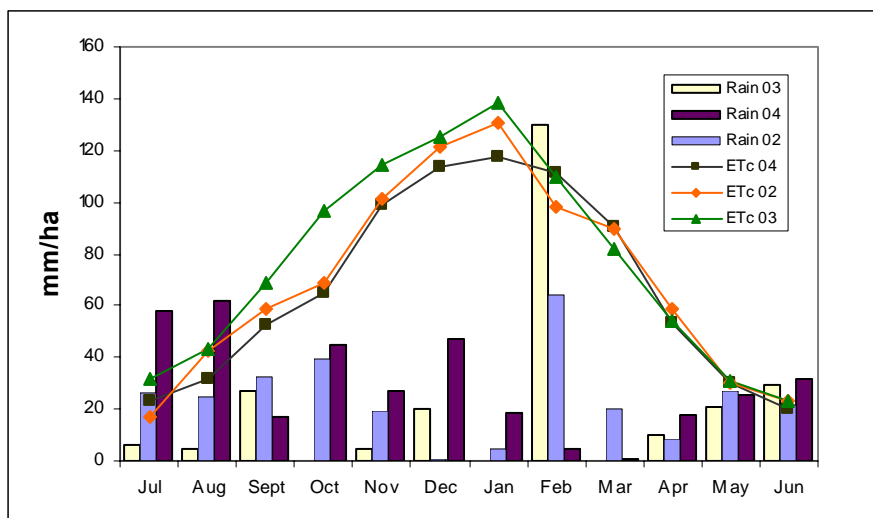
4.1.1 Climatic conditions

The trial was conducted during three years of severe drought in the Riverina. Total rainfall for the July to June periods over the course of the project was 288, 253 and 354 mm for 01/02, 02/03 and 03/04 respectively. The rainfall and potential crop evapotranspiration ETp are shown in Fig. 4.1 illustrating a high level of dependence on irrigation to supply crop water requirements. In 2004 this demand for irrigation was particularly high in the period from late December 2003 to late May 2004.

4.1.2 Crop evapotranspiration potential

We have calculated potential crop evapotranspiration ETp for olives by multiplying reference crop evapotranspiration by crop coefficients (Kc) of 0.5 to 0.65 (Fernandez and Moreno 1999). Given the trees used in this study are not full size and have a canopy area of approximately 40% of the orchard floor area this Kc was reduced by 33% based on the assumption that full canopy size would cover 60% of the orchard floor area. Reference crop evapotranspiration (ETo) used to calculate (ETp) were supplied by CSIRO in Griffith NSW, the closest weather monitoring station to the trial.

Fig. 4.1 Rainfall and crop potential evapotranspiration ETp for 2001/02, 2002/03 & 2003/04.



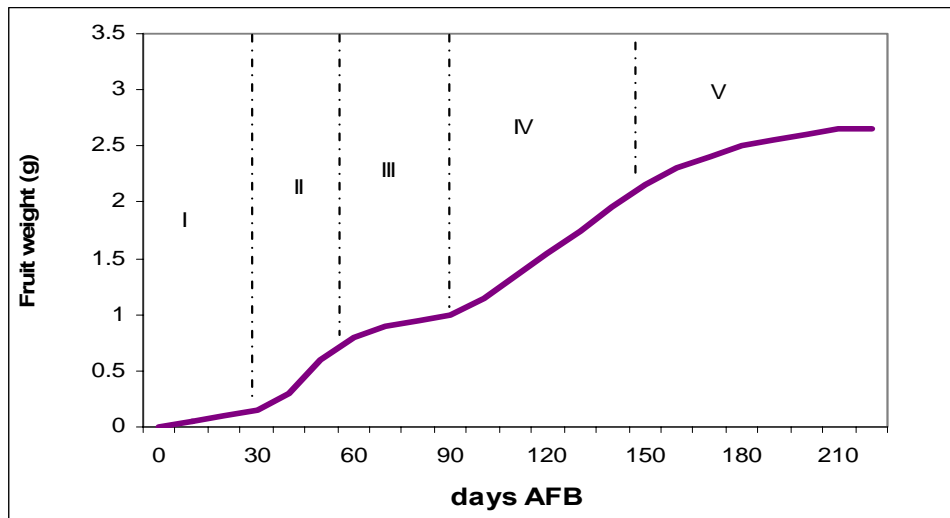
4.2 Irrigation

4.2.1 Olive fruit development and moisture

Olive fruit development goes through 5 main stages (Levee 1996). Crop management at each of these stages was reported to have an effect on the eventual yield and quality of the olive oil produced.

- a) *Fertilization and fruit set.* Rapid early cell division involves growth of the embryo. Enlargement of the new fruits becomes noticeable after about two weeks. A large number of fruit and flower parts are dropped in this stage, and severe moisture stress imposed on the tree will increase the level of abscission and reduce the crop potential. When flowering starts in early November this period will last until late November to early December.

Fig. 4.2 Schematic graph of olive fruit growth and stages of fruit development corresponding to days after full bloom (AFB) (adapted from world olive encyclopaedia)



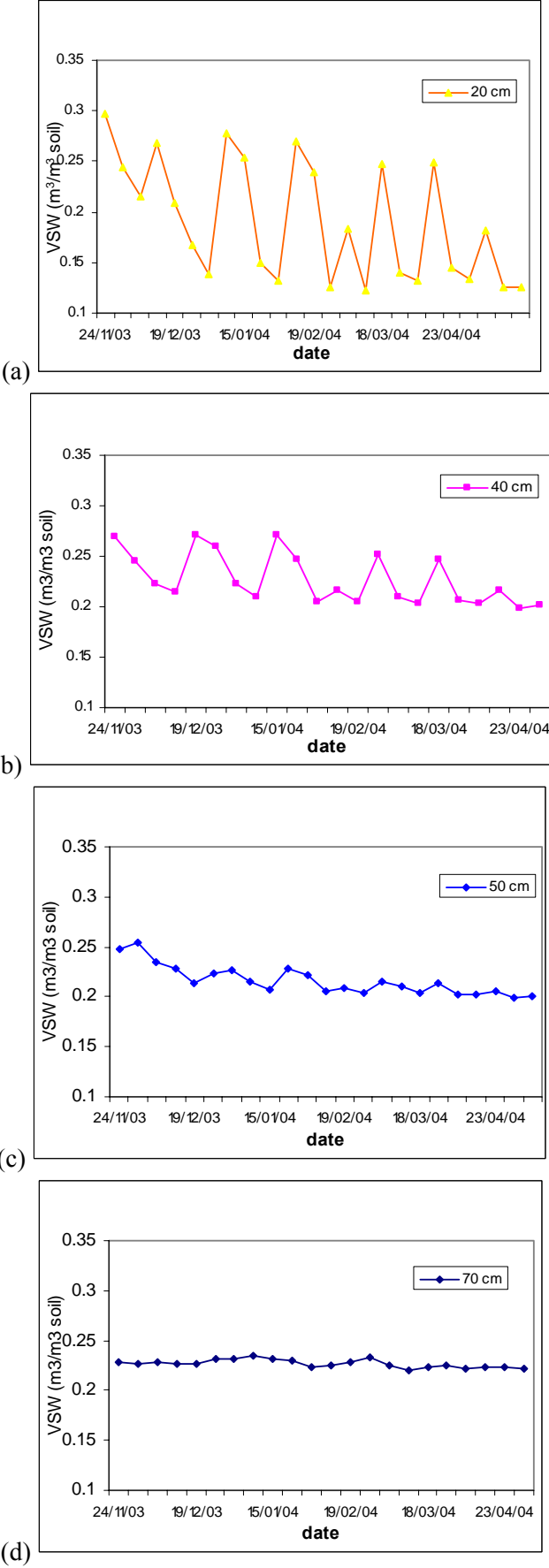
- b) *Seed (endocarp) development.* With early to mid November flowering, stage II often runs from early December to early and mid January. Stage II is a period of rapid fruit growth due to both cell division and enlargement that mainly involves the growth and development of the endocarp (seed/pit). There is little development of the flesh (mesocarp), nor is there significant oil production in the fruit at this time. Lavee (1996) suggests that imposing moisture stress on the crop in the latter part of stage two can reduce seed size and improve the flesh-pit ratio in table olives. Where irrigation water is limited some water stress late in this period can also reduce vegetative vigour and reduce competition for the developing crop. Moderate moisture stress is not thought to impact on oil production potential. However, stress, sufficient to limit cell division, has been reported to potentially limit eventual fruit size and yield potential.
- c) *Seed/pit hardening (endocarp sclerification).* Fruit growth slows during stage III as the fruit undergoes a hardening of the endocarp. This process can last 4-5 weeks running into early to mid February. There is little growth of the fruit and little oil synthesis during this period. The crop can tolerate reasonable moisture stress during this period and it is a time when fruit respiration is low. The tolerance of dry conditions also applies to the vegetative parts of the tree and vegetative growth will also slow or stop when stomata close up and transpiration slows in the hot conditions of January to early February.
- d) *Mesocarp (fruit flesh) development (cell enlargement oil synthesis).* Beginning in early February stage IV is the second period of rapid fruit growth. This growth is due to development of the mesocarp or flesh of the fruit and mainly involves cell enlargement. This is also the main period of oil synthesis with oil accumulation in the fruit being directly correlated with fruit growth (dry matter increase) during this period. Moisture stress can restrict growth of the fruit and oil production potential. Lavee (1996) reports that where irrigation water is limiting the most benefit to productivity is gained in this period. We observed in the field this year that as stage IV progresses the fruit becomes increasingly susceptible to moisture stress and desiccation. This may be related to the progressive increase in fruit respiration rate (Ranalli *et al*, 1998) and the increasing mass of soft tissue in the fruit. Where irrigation water supply is limited it maybe possible to impose a moderate deficit at the beginning of this period and increase water inputs as fruit growth progresses.
- e) *Ripening.* Ripening commences with green maturation. This is described as the change from dark lime green to lighter green but it can be determined more accurately in the field by observing the onset of fruit softening. This rapid change in fruit texture takes place over a period of one to two weeks and can be observed as a change from hard (Granny Smith apple texture) where the fruit is difficult to squash between thumb and index finger, to softer texture where the fruit is easily

squashed and milky juice is released. As the fruit ripens, dry matter continues to increase along with oil synthesis, although at a slower rate than in stage IV. As with stage three, the respiratory demand from the fruit lessens and with the weather cooling, drops off. Crop water management in stage V can have a dramatic impact on yield and quality characteristics. Fruit moisture content tends to drop off as ripening commences and imposing a water deficit on the trees would be expected to reduce fruit moisture further. It is possible that imposing water stress combined with the onset of colder weather can reduce metabolic activity in the fruit leading to reduced utilization and greater retention of phenolic compounds in the ripening fruit. In addition low fruit moisture at oil extraction time may increase the proportion of phenols in the oil due to reduced loss in vegetation water. Monitoring fruit moisture levels in the ripening period can provide a useful guide to crop water management to ensure that the required fruit moisture levels are attained at harvest time. Varieties will vary substantially in their response to water deficits and different irrigation management may be necessary for different varieties.

4.2.2 Zone of root activity

Soil moisture depletion was most rapid in the top soil from surface to 30 cm due to evaporation and water extraction by plant roots (Fig. 4.3a). At 40 cm depth soil moisture depletion is primarily by plant roots (Fig. 4.3b) and shows rapid soil moisture depletion followed by a much slower rate of depletion as VSW approaches the limit of plant available water at about $0.2 \text{ m}^3/\text{m}^3$. Fluctuation in soil moisture at 50 cm depth was less due to limited infiltration of water and reduced root activity causing much slower depletion at this depth. By late in the season (March –April VSW at 50 cm was maintained very close the lower limit of plant available water due to an extended crop water deficit period.. At 70 cm soil moisture did not fluctuate indicating no plant water uptake from this depth. Soil moisture depletion from the 0-50cm zone was used to monitor crop water use.

Fig. 4.3. Volumetric soil moisture (VSM) readings from (a) 20 cm, (b) 40 cm, (c) 50 cm and (d) 70 cm below the soil surface from November 2003 to May 2004.



4.2.3 Crop water Inputs

Soil moisture monitoring data is presented in Fig. 4.4 (a), (b) and (c). Soil moisture was measured some days after each irrigation or rainfall event because the orchard was inaccessible after irrigation or rainfall. As a result, there is variation in the maximum peaks height in these curves.

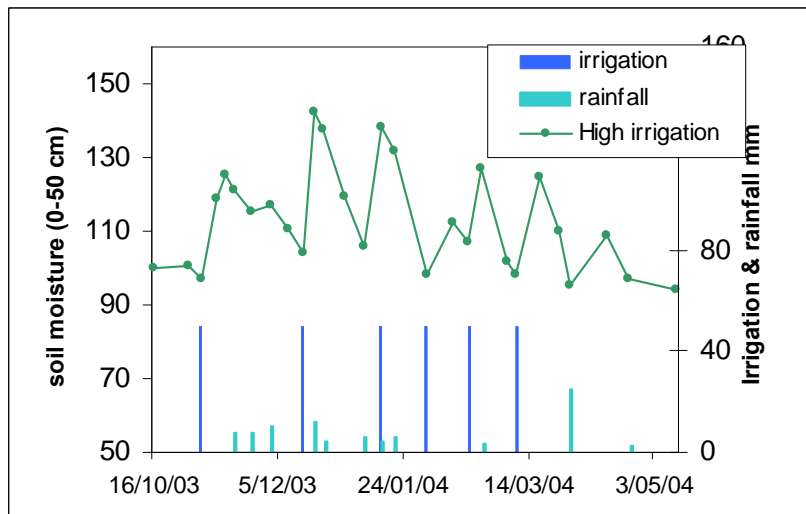
Soil moisture monitoring revealed rapid depletion of soil moisture following irrigation or rainfall. Following depletion of readily available soil moisture the rate of soil water uptake by the crop decreased as extraction from the soil became more difficult. This is illustrated by a levelling out of the soil moisture curves (Fig 4.4).

In the high irrigation treatment a total of 6 irrigations were applied at intervals of 2-4 weeks apart (Fig. 4.4 (a)). The long intervals between watering meant that trees were subjected to soil saturation followed by readily available water declining to fairly severe soil moisture deficits within each pre-irrigation interval. In the case of the deficit treatments the rate of soil moisture depletion slowed substantially as soil moisture approached the limit of plant available water. These extended dry periods produced visible signs of moisture stress in the plants such as upward turning and rolling of the leaves, as well as fruit shrivelling. Fruit shrivelling became increasingly frequent as fruit grew larger during stage IV of fruit development and was observed in all treatments and across all varieties as they were subjected to a soil moisture deficits through late fruit growth and maturation. Sanchez Raya (1983) observed fruit respiration progressively increased and peaked during this period of fruit growth which is consistent with our observations of increasing susceptibility to desiccation as stage IV progressed.

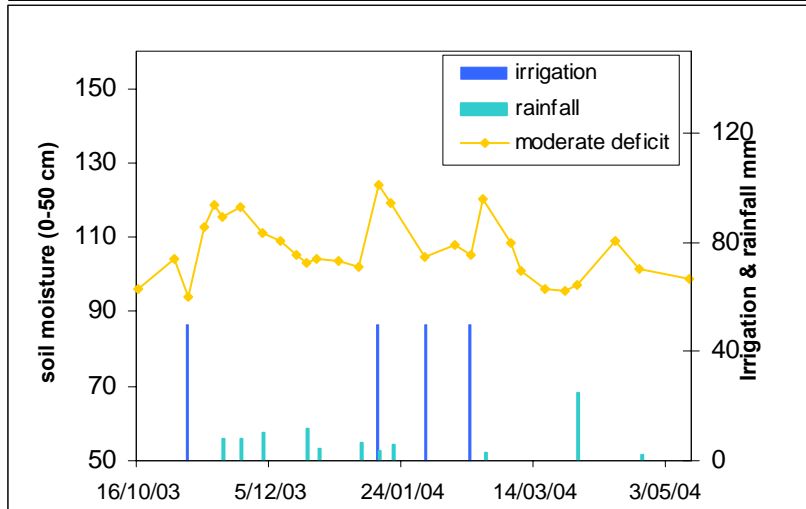
The nature of furrow irrigation, with long intervals between irrigations and the lack of precision in water application means that it is less than ideal for research into precise water management options for olive growers. However, the furrow irrigation applied in this orchard in 2004 was an efficient method of water application due to the accurate management of the irrigation system and its application by Cookatharma management. The long rows (400 m) means there is minimal excess flow of water into drainage channels (which is recycled). Infiltration of water after irrigation was to 40-50 cm depth and there was no wastage of water through deep infiltration beyond the root zone. The high irrigation regime used in the trial used approximately 3 Ml /ha of water and was sufficient to produce a 6-7 tonne/ha crop of olives from 6-7 year old trees. Interestingly there was only a moderate reduction when the irrigation was reduced to 1.5 to 2 Ml / ha of water despite the fact that the trees showed signs of severe moisture stress for extended periods under these severe deficit treatments. We will monitor crop yield next season to see if the severe moisture stress during the 2004 season has any effect on yields in 2005. It is likely that the crop would have benefited from additional irrigation in late stage IV (late March) and early stage V (early to mid April) which would be expected to increased yield potential and may have influenced the fruit polyphenol levels.

Fig. 4.4 Rainfall and irrigation events and volumetric soil moisture levels in the top 50cm of soil for high irrigation (G), moderate deficit (Y) and severe deficit irrigation (R), October 2003 to May 2004.

(G)



(Y)



(R)

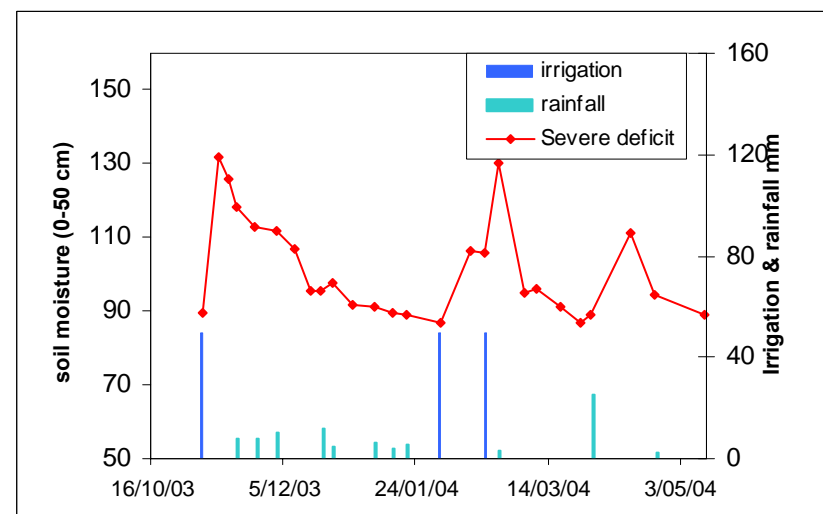


Table 4.1 Crop water inputs in relation to potential crop evapotranspiration, ETp, at the different stages of fruit development - November 2003 to 12 May 2004.

Fruit Dev Stage	ETp mm	Crop water inputs (rainfall and irrigation)					
		G		Y		R	
		Mm	%ETp	Mm	%ETp	Mm	%ETp
I	74	97	100	97	100	97	100
II	137	66	48	26	19	26	19
III	117	50	43	50	43	10	9
IV	171	142	83	102	60	102	60
V	71	26	37	26	37	26	37

Crop water inputs were not limiting during stage I in any of the treatments in stage II but only 48%, 26% and 26% of potential crop water use was supplied to the G, Y and R treatments respectively. In stage III (seed hardening) the deficit was 43% of ETp in G and Y treatments and 9% in the R treatment. In stage IV, the main period of oil synthesis the crop water supplied was 83%, 60% and 60% of crop potential water use declining to 37% in all treatments in stage V. During late stage IV and stage V the fruit and trees showed signs of moisture stress with fruit shrivelling occurring in all treatments during this period. This degree of stress in late stage IV and in much of stage V is likely to have reduced oil yield potential.

4.3 Fruit detachment force (FDF)

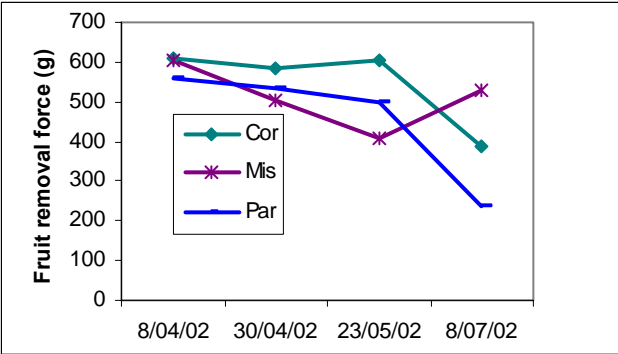
In 2002 fruit detachment force for Corregiola remained high until late May and this was reflected in the difficulties in achieving sufficient fruit removal with a tree shaker from this variety during the commercial harvest (Fig. 4.5a). Paragon had a lower FDF and fruit removal with the shaker was more successful. In 2002 softnose fruit rot in the Mission caused premature fruit drop and low average FDF. This resulted in rapid decline in average FDF through April and May. The high final detachment force in Mission in 2002 was most likely because all the diseased fruit had fallen before the final measurement was taken. In 2003 FDF remained high in Corregiola and Mission until late May while Paragon declined significantly between 30 April and 28 May (Fig. 4.5b). In 2004, FDF for Corregiola and Paragon followed a similar trend with levels increasing initially between 14 April and 5 May followed by a decline from early to late May and then an increase again by the final harvest date on 12 July (Fig. 4.5c). The final measure of FDF in 2004 was on fruit remaining after the trees had been harvested by a tree shaker and this has most likely contributed to higher than expected average value for Corregiola and Paragon. The early season increase in FDF in 2004 is unexplained.

The data show that variety and maturity stage can affect the ease of fruit detachment. It also shows that some varieties, in this case Corregiola and Mission can maintain high detachment force until well into the normal commercial harvesting period. This can contribute to the problem of insufficient mechanical fruit removal by harvesters and where this causes a delay in harvest there is potential for quality loss. It may be necessary for growers to consider using harvesting methods or machines capable of high fruit removal efficiency to reduce this risk.

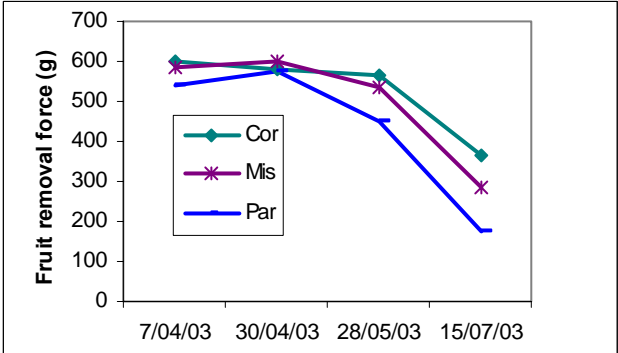
The variation in varietal behaviour over the maturation period and from season to season indicates that other crop physiology factors may influence fruit attachment. This detracts from the possibility of using this measurement to optimise harvest timing. Further research into physiological and developmental factors that affect fruit attachment could contribute positively to successful mechanical harvest of olives.

Fig. 4.5 Fruit detachment force (g) for Corregiola (Cor), Paragon (Par) and Mission (Mis) cultivars measured at 4 periods during the 2002(a), 2003(b) and 2004(c) season.

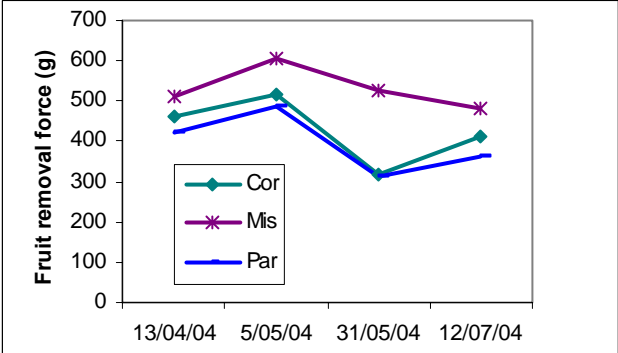
(a)



(b)



(c)

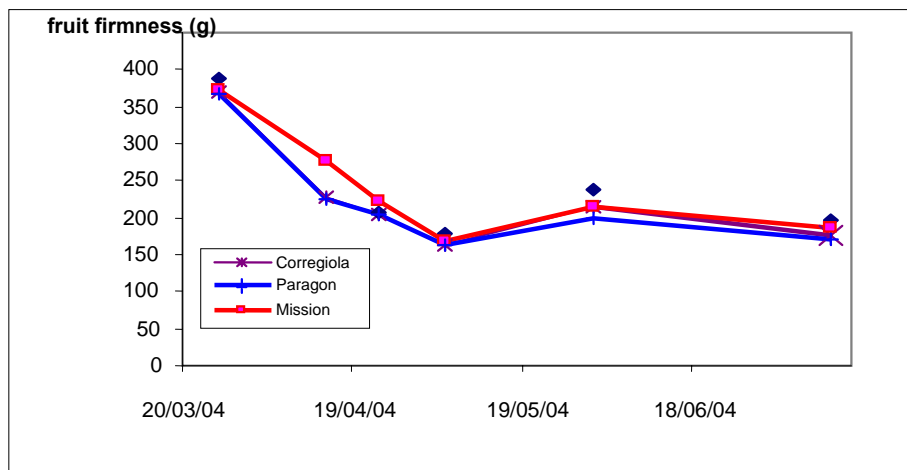


4.4 Fruit firmness

Fruit firmness was measured during the 2004 growing season to assess changes in fruit firmness associated with variety and stage of maturity. Fruit firmness declined rapidly with the onset of fruit maturation in early April. As fruit began to ripen the rate of softening slowed. We would expect another period of rapid softening as the fruit becomes over ripe prior to fruit drop. From previous experience, this can be prolonged as olive fruit remain on the tree in sound condition for several months after harvest in some varieties. These observations are consistent with Sanchez-Raya's (1983) measurements of fruit softening through maturation and ripening of olives.

The initial decline in fruit firmness with the onset of fruit maturation could be a useful indicator to assist management in the lead up to harvest. It may be used to indicate the reduction in crop irrigation following fruit softening. It may also provide a guide to a likely commencement of harvest. This would be dependent on detailed quality and yield profiling to monitor other yield and quality parameters.

Fig. 4.6 Fruit firmness for three cultivars of olives, Mission, Corregiola and Paragon in 2004.

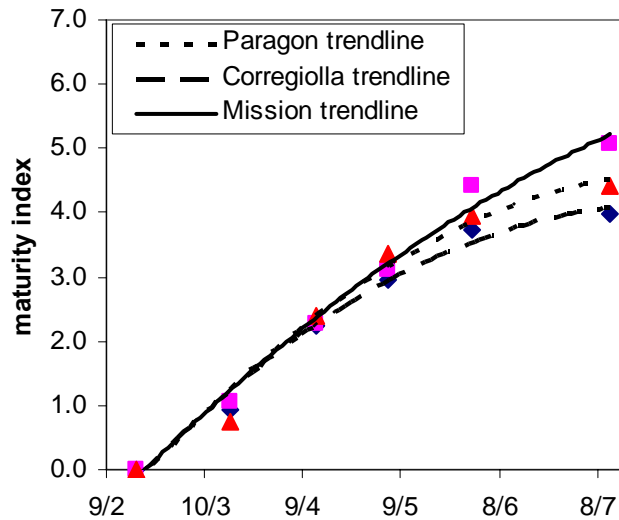


4.5 Maturity index

The most obvious physical change during maturity, and one that is often used to determine fruit ripeness, is the maturity index. The appearance of the fruit over the maturation period is illustrated in Fig. 3.2. The green skin will change gradually to become totally black. The internal flesh will also change from bright green to a milkish white and eventually become purple to black. To determine maturity index, it is necessary to cut the fruit open to examine the colour of the flesh. The early change in skin colour compared to the relatively late change in flesh colour in Fig. 3.2 illustrates the problems in predicting the maturity of the olive. The problem is further increased as the rate of change in both fruit skin colour and flesh colour may vary significantly between trees of one cultivar and even within a single tree.

In this study, fruit changed colour over a period of 3-4 months (Fig. 4.7) with some trees showing a longer period to change from green to black than others. The graph represents the average maturity index for each cultivar, based on three replicates over three years. It clearly shows that over the three years, Mission was the first to change colour in the skin and throughout the fruit. Although Mission turned purple-black relatively quickly, the skin of Corregiola tended to turn black but the flesh remained green to pale throughout maturation.

Fig. 4.7 Trendline of maturity index for three cultivars of olives. Each point represents the mean of three replicates over three years.



It may be considered that olives with black skins and green flesh would retain fruit firmness and this combination of tests may be a better indicator than just skin colour. However, it was found that fruit firmness dropped to very low levels at a time when maturity index was still low and increasing. The combination of fruit firmness and maturity index may be of limited use to determine the period when olives are reaching optimum harvest timing. It may also be an indicator for irrigation management. Fruit firmness or maturity may be combined with methods such as fruit weight gain and oil content to improve precision in determining when to harvest.

Maturity index continues to be used in Europe as a strong indicator of the optimum time to harvest (Shimon Lavee, AOA Conference 2004). In Australia however, the rate of change of maturity index for each cultivar was considerably different. For Corregiola in particular, if the colour had not changed by April, the flesh tended to remain green with a maximum maturity index of only 3-4 over the entire season. As a result, harvest timing relying on this parameter would result in very late harvest with markedly reduced quality. On the other hand, Mission showed a continual change in colour throughout the season until the flesh had become a rich purple. In this case, reliance on maturity may result in overripe fruit, from which it has been shown, oil extraction efficiency is considerably reduced.

Fruit colour changed at different rates for each season with a more rapid rate in 2003 than for the other two years. Colour change was considerably slower in 2002. Oil content on the other hand, was consistent over the three periods, showing that maximum yield for the three years would have been achieved at three different maturity indices. These factors reinforce the difficulty in using maturity index as a determinant for harvest timing to produce a consistent product. Premature harvesting may result with loss in oil yield together with reductions in oil quality as described in other sections of this report.

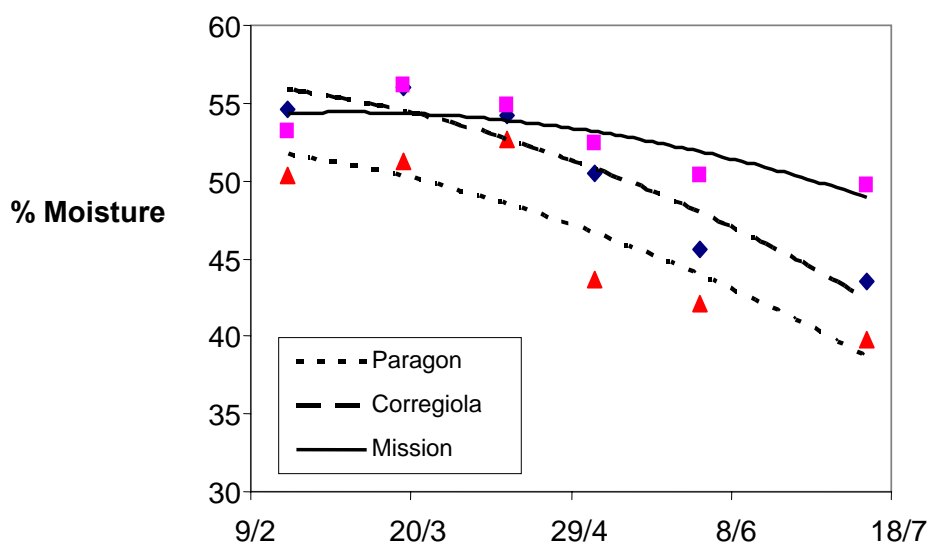
Oil extracted from green olives, or olives with black skins and green flesh generally exhibited grassy and pungent sensory characteristics whilst being processed whereas olives with a maturity index of 5 to 7 had lost this characteristic.

4.6 Moisture and oil content

4.6.1 Moisture content

Moisture content is a major factor for olives as it generally contributes to more than 50% of the fruit weight. It therefore has several effects on the fruit and oil quality. Moisture content is very variable across the maturation period as the olives take up water when it is abundant but will release moisture when it is in short supply. The appearance of the fruit changes as they become round and plump after an irrigation or rainfall event but they may be shrivelled and smaller with water stress.

Fig. 4.8 Trendline of moisture content for three cultivars of olives. Each point represents the mean of three replicates over three years.



In addition to the variability in moisture availability during maturation, there is a tendency for moisture content to reduce as the fruit mature. This study showed that moisture was generally high, around 55%, in all of the cultivars at fruit set and as the fruit began to develop. However, the moisture levels showed an overall downward trend across the season (Fig. 4.8). This was particularly evident in Paragon, and, to a degree, in Corregiola. Mission however was very different to the other cultivars with moisture levels remaining relatively constant throughout fruit development with a maximum average value of 56.1% down to a minimum of 49.7% in mid July. The Paragon cultivar showed the lowest average moisture level with a maximum of 52.7% moisture in mid April down to 39.8% in July.

In Fig. 4.9, 4.10 and 4.11, a comparison of irrigation treatments and time of harvest is shown for three cultivars for each of the years 2002, 2003 and 2004 is shown. It can be seen that there is a scatter of points about the line of best fit, with no particular pattern across the varieties or the years. This is an indication of the variability in moisture content with time due to irrigation and or rainfall events.

Corregiola (Fig 4.9) showed a significant decline in moisture content over each of the periods of harvest for each of the three years ($p < 0.001$). The initial moisture content at the first harvest was highest in 2002 and lowest in 2004 indicating progressively lower moisture levels in the soil for the

three years. Moisture content was significantly different over the three years ($p = 0.053$) although there was no effect of irrigation on moisture content. This would suggest irrigation was insufficient.

Mission (Fig 4.10) moisture concentration also declined for each harvest time over the first two years ($p = 0.019$) although in 2004 there was very little change in moisture content throughout fruit growth. There was a significant difference between the three years ($p < 0.001$) but the lack of significant difference between irrigation treatments ($p = 0.470$) would suggest that irrigation was deficient.

Paragon (Fig 4.11) showed the greatest range in moisture content of each of the cultivars although the year effect was insignificant ($p = 0.452$). The dramatic difference in moisture for different harvest times shown in 2002 was quite different to the constant moisture observed in 2004. Despite that, the effect of harvest timing overall indicates that it is significant ($p = 0.026$). There were no irrigation effects ($p = 0.477$).

There are no statistical data between the three cultivars as the blocks were independent and there was no control between the blocks. The trends are similar for each of the cultivars with reductions in moisture with maturity. In all cases this reduction was less for the third year. Additionally, the data for all three cultivars was less than 60% throughout the season which is a clear indication that the trees were experiencing some water stress for each of three low rainfall seasons.

In the prevailing drought conditions of the 2004 season, fruit moisture content changed in response to irrigation and rainfall events and the extremely dry conditions at this time. The fact that in premature fruit, moisture levels were at or below 50% in several of the varieties studied in late February gives an indication of the severity of soil moisture deficits imposed on the crop early in the season. There is a clear response to the mid March irrigation in the full irrigation (G) treatments and a clear response to the April 6 rainfall in all varieties and treatment. This is apparent from the (G) points above the line of best fit which in reality would have been a spike in fruit moisture level in an otherwise extended period of low fruit moisture. There was variation between varieties in their response to soil moisture deficits with fruit moisture in Paragon (2004) responding to changes in soil moisture conditions much more dramatically than the other two varieties. In general, the fruit moisture levels illustrate that the crop experienced moisture deficit throughout late fruit growth, maturation and ripening. There was no significant effect of irrigation for any of the cultivars.

Moisture content of the fruit is important to oil quality for a number of reasons. If the fruit moisture level drops to a point where desiccation occurs, cell breakdown can follow leading to increased free fatty acids and therefore lower oil quality. Stress conditions during mesocarp development (Stage IV) may also result in reduced oil yields by limiting oil synthesis and accumulation. Fruit growth may also be restricted. On the other hand, if the moisture content of the fruit is very high at the time of harvest, low yields result from cold press extractions. Moisture levels at the time of harvest may also have effects on fruit quality which will in turn affect oil quality. The moisture content of the fruit can be influenced by numerous factors including rainfall, evaporation, irrigation events, soil type and tree health.

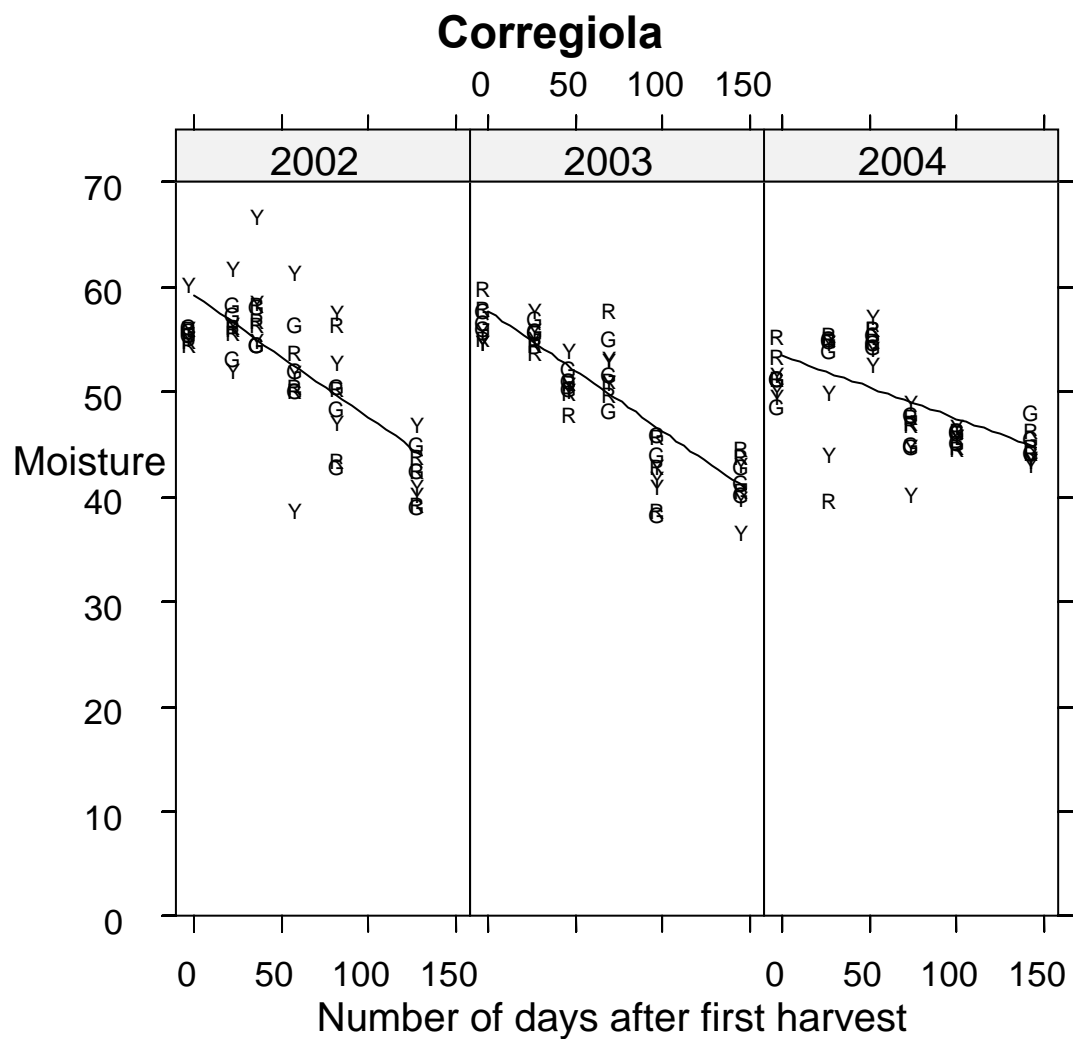


Fig. 4.9 Moisture content for Corregiola at six harvest dates over three years including three irrigation treatments. G = full ; Y = moderate; R = deficit irrigation. There was limited effect between years ($p = 0.053$) and no irrigation effects ($p = 0.986$) although there was significant time of harvest effects ($p < 0.001$).

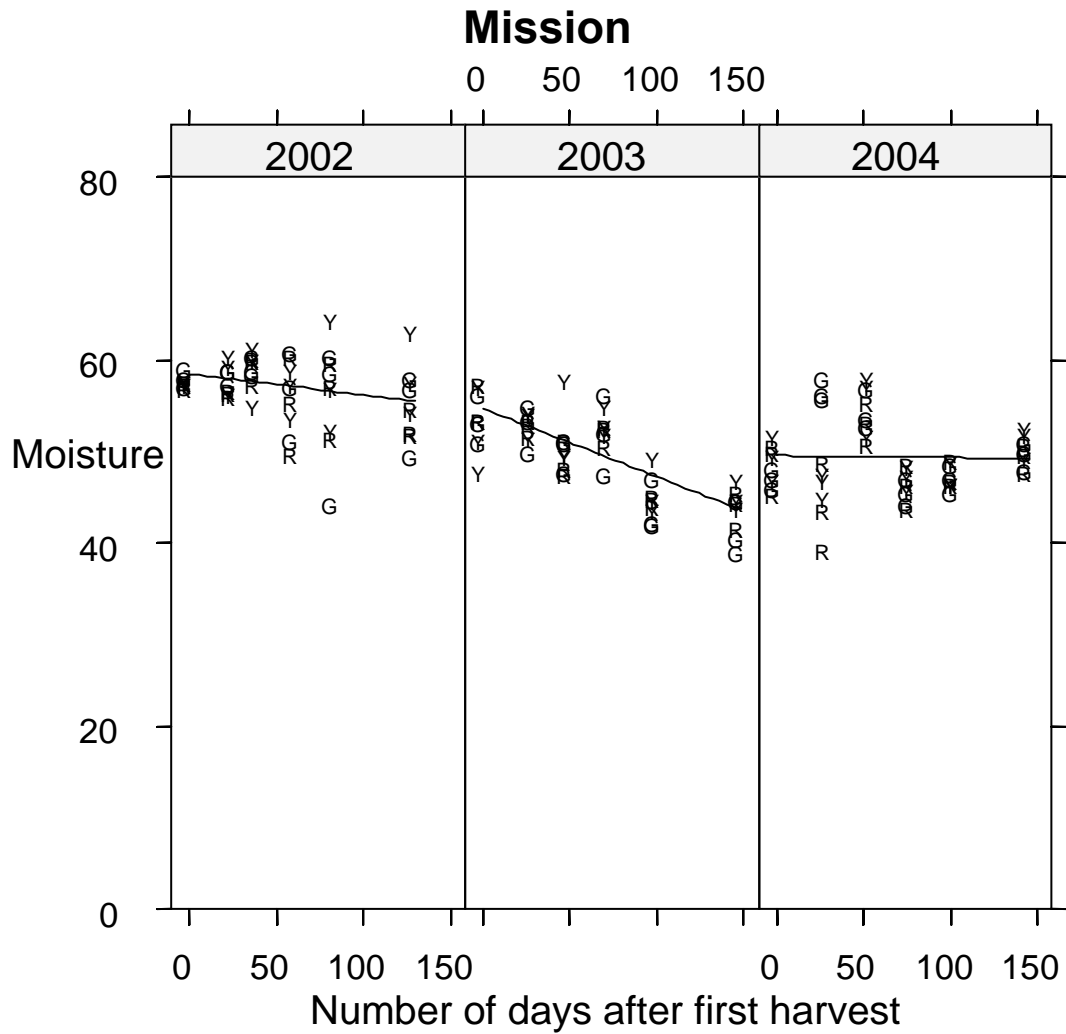


Fig. 4.10 Moisture content for Mission over three years and including three irrigation treatments. G = full ; Y = moderate; R = deficit irrigation. There was a significant year effect ($p < 0.001$) but no effect for irrigation treatment ($p = 0.470$). There was significant time of harvest effect ($p = 0.019$).

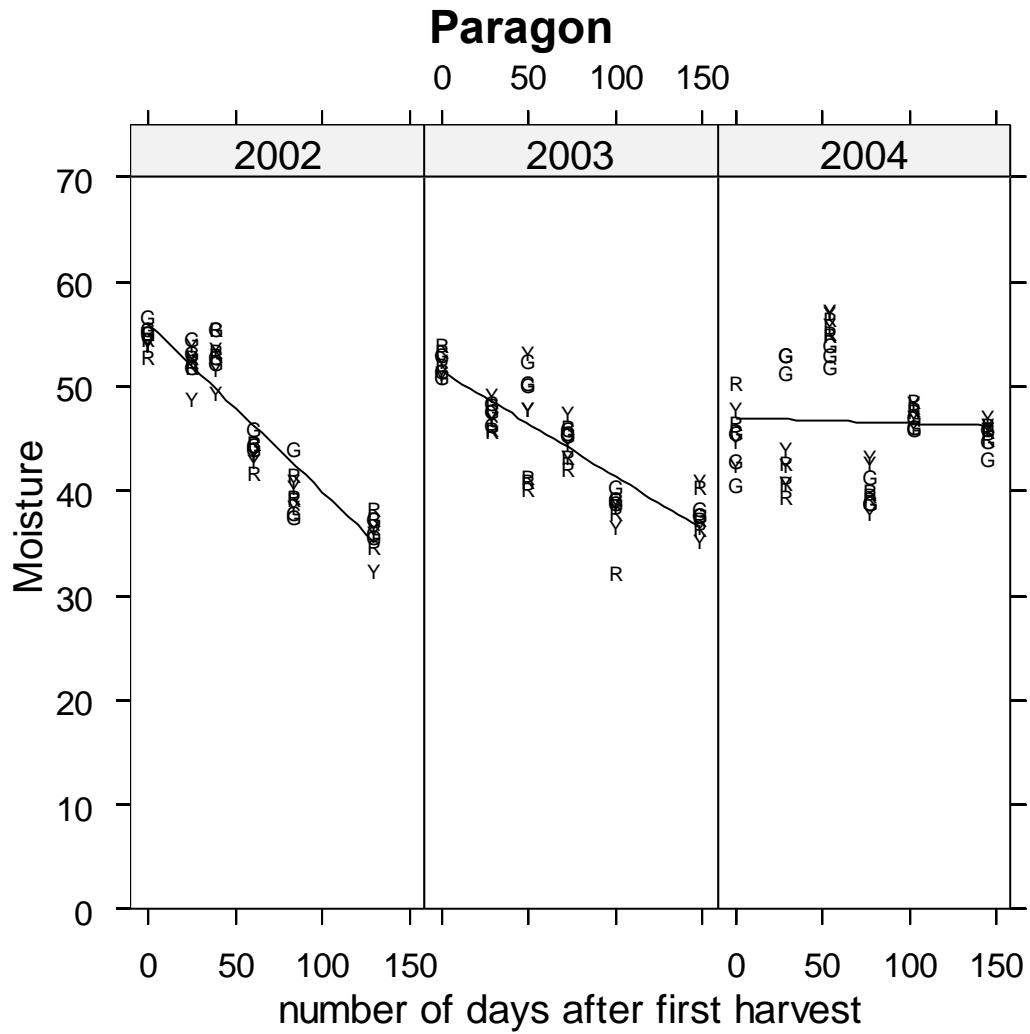


Fig. 4.11 Moisture content for Paragon over three years and including three irrigation treatments. G = full ; Y = moderate; R = deficit irrigation. There was no significant year effect ($p = 0.452$) and no effect for irrigation treatment ($p = 0.477$) although there was a time of harvest effect ($p = 0.026$).

4.6.2 Oil content - cold press extraction

Olive fruit yield and oil content are the major contributors to profitability for olive growers where currently no premium is paid for quality. Factors which are considered to contribute to fruit yield and oil content would include cultivar, environment and seasonal effects. There have been a considerable number of publications which have studied factors which contribute to oil content in olives.

Extra virgin olive oil must be extracted by mechanical means without the use of chemicals including solvents. The oil content for growers is therefore “*the total amount of oil which can be extracted by cold press extraction*” rather than the total amount of oil in the fruit. It is understood however that cold press extraction leaves a variable amount of oil in the pomace, or waste product. This waste can be considerable and it is in the processors interests to try to keep it to a minimum.

The Abencor cold press oil extractor used at the Wagga Wagga Oil Laboratory simulates the industrial extraction process. The fruit is ground to a paste, mixed and centrifuged to separate the three components, oil, water and solids. The three extractor units, a hammer mill, a malaxer and a centrifuge, separates most of the oil from the other components but, as for industrial extractors, leaves varying amounts of oil in the pomace.

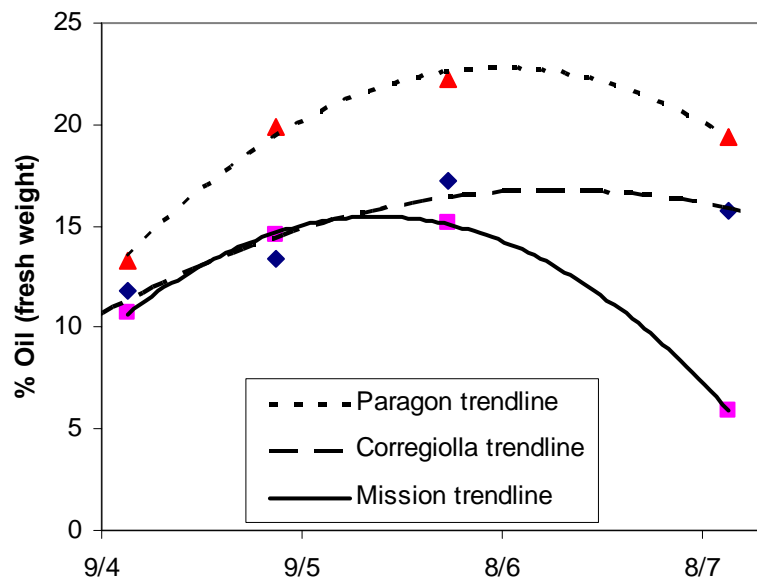
Samples of olives at each harvest in this study were analysed for oil content using this cold pressed system. The average weight of oil, extracted by cold press, was determined and is illustrated in Fig. 4.12. Oil content is expressed as a percentage of the fresh weight of olives. Each point represents the mean value for oil content for three replicates over three years for each cultivar.

The trend for oil content was similar for all cultivars with the amount of oil extracted tending to increase to a maximum at mid maturity and then decrease. It can be seen from the results (Fig 4.12) that the amount of oil recovered for each cultivar in the early stages of maturity was very similar. However, as the fruit matured, the difference in the amount of oil extracted from each cultivar became greater. The highest amount of oil extracted was from Paragon which peaked in early June and then showed a reduction in the oil extracted from then on. Mission produced reasonable yields up to mid May but oil recovery then reduced. Corregiola extraction efficiency was also reduced but not to the same degree of reduction as Mission.

When extracting older fruit, a stable emulsion developed between the oil/water phases which consisted of fine solids from the fruit. This emulsion contained oil, water and solids. As a result, a considerable portion of the oil could not be separated. In the case of Mission, although it yielded similar oil contents to the other cultivars in young, green olives, it had very low recovery rates at late maturity. Corregiola had the least of this emulsion and the best oil extraction percentage, although the oil yield was less than Paragon. It is of interest that Corregiola was also the fruit which had the lowest maturity index at the end of the maturation period. This would suggest that separation of the fruit from the pomace was more complete in Corregiola, possibly because the fruit flesh had not degraded to the same extent as Mission.

The maximum oil recovery of 28.8% was achieved by Paragon on 20/05/2003. The lowest oil recovery was Mission with only 1.31% on 12/7/2004. This highlights the importance of understanding the changes during fruit maturation and the effects that it can have on oil yield.

Fig. 4.12 Trendline of oil content, cold-pressed for three cultivars of olives. Each point represents the mean of three replicates over three years. Extraction of oil is achieved using an Abencor cold press oil extraction unit.



A statistical analysis of the variation in cold-pressed oil content for the three cultivars over three years is illustrated in Fig. 4.13, 4.14 and 4.15. The figures show a comparison between years, harvest timing and irrigation treatments. The statistical significance of each of the variables is shown in the Appendices. There were dramatic fluctuations in the oil contents for each of the cultivars over the harvest periods and for the three years.

Corregiola (Fig 4.13) showed no significant effect across years ($p = 0.687$) with rapid oil accumulation up to 100 days after first harvest. There was a significant reduction in the cold-pressed oil recovered at the last harvest period in years 2003 and 2004 although this reduction was not obvious in 2002. The reason for this is not apparent and is not explained by moisture contents which were similar to the other two cultivars. The reduction in oil recovery however, was found to be less influenced by fruit age between the third and fourth harvest dates (Table 4.2). There was an effect of time of harvest ($p = 0.032$) but there were limited effects of irrigation ($p = 0.229$).

Mission performed quite differently over the three years although the trend of rapid oil accumulation followed by reduced recovery after fruit maturity was common. The difference between years was significant ($p = 0.010$) although the time of harvest was not ($p = 0.586$). Irrigation effects were limited ($p = 0.158$).

Paragon cold pressed oil content also decreased after maturity in the last two years but remained high in 2002. The year effect was significant ($p=0.012$) and the time of harvest showed differences ($p = 0.032$). The irrigation effect was apparent ($p = 0.229$).

The dramatic changes that occur with the cold-pressed oil extraction for the three cultivars, particularly Mission and Paragon, enforce the fact that harvest timing is critical to achieve optimum, and even acceptable returns. Delayed harvest has a strong influence on oil extraction efficiency. The emulsion that forms when olives are ground is much more stable in old fruit and retains a higher percentage of the oil. Maximum extraction appeared to be in mid May which is generally before the maximum oil is produced. However, due to the higher extraction efficiency, this may be a more profitable harvest period. After this period, oil recovery begins to decrease even though the actual oil content may still be increasing.

The laboratory scale cold-press extractor used in many laboratories, and also used in this study, is considered to be less efficient than commercial sized olive extraction equipment. Our findings therefore may show larger differences in oil content between cold press and solvent extraction than would occur in an industrial olive press. However, although the actual percentage extraction may be less, the effects that cause poor extraction efficiency have been highlighted.

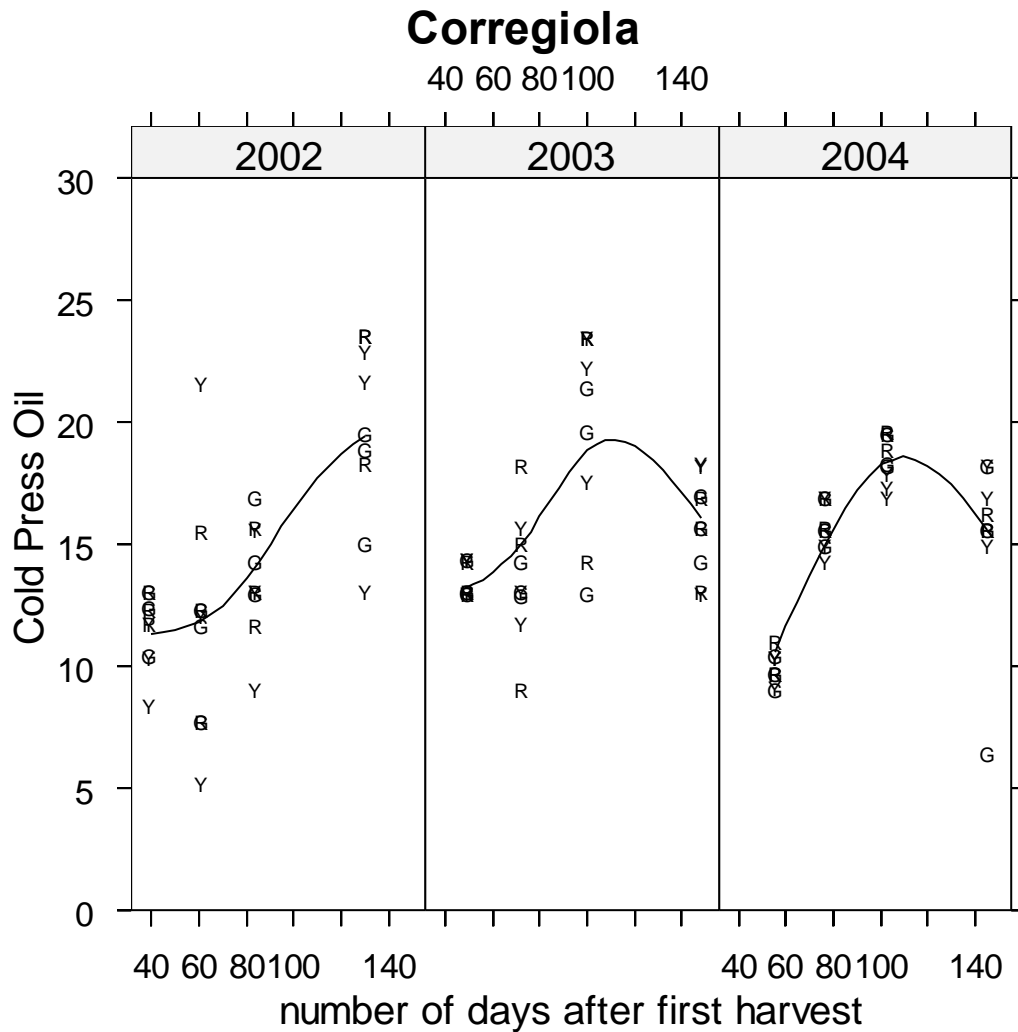


Fig. 4.13 Cold-pressed oil content for Corregiola over three years and including three irrigation treatments. G = full ; Y = moderate; R = deficit irrigation. There was no significant year effect ($p = 0.687$) and no effect for irrigation treatment ($p = 0.537$) although there was a time of harvest effect ($p = 0.075$).

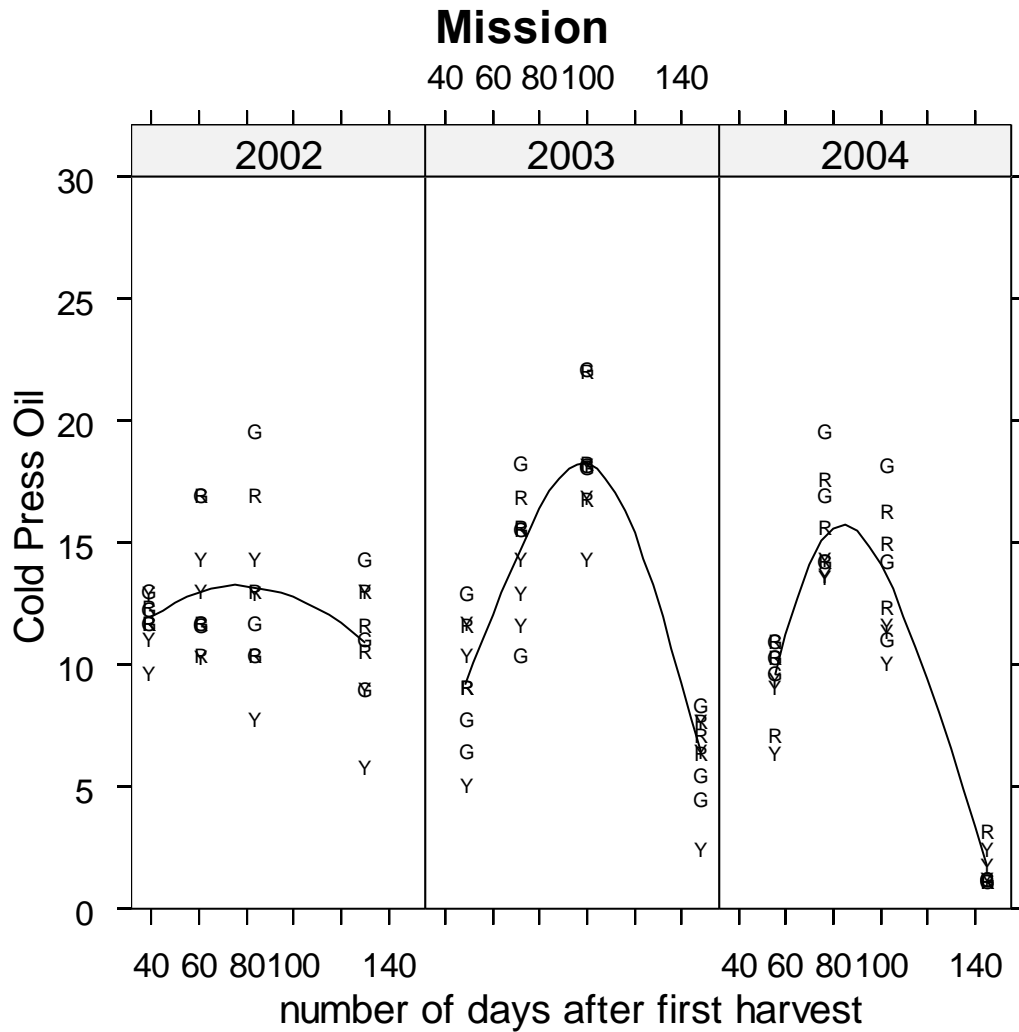


Fig. 4.14 Cold-pressed oil content for Mission over three years and including three irrigation treatments. G = full ; Y = moderate; R = deficit irrigation. There was a year effect ($p = 0.010$) but little effect for irrigation treatment ($p = 0.158$) or time of harvest ($p = 0.586$).

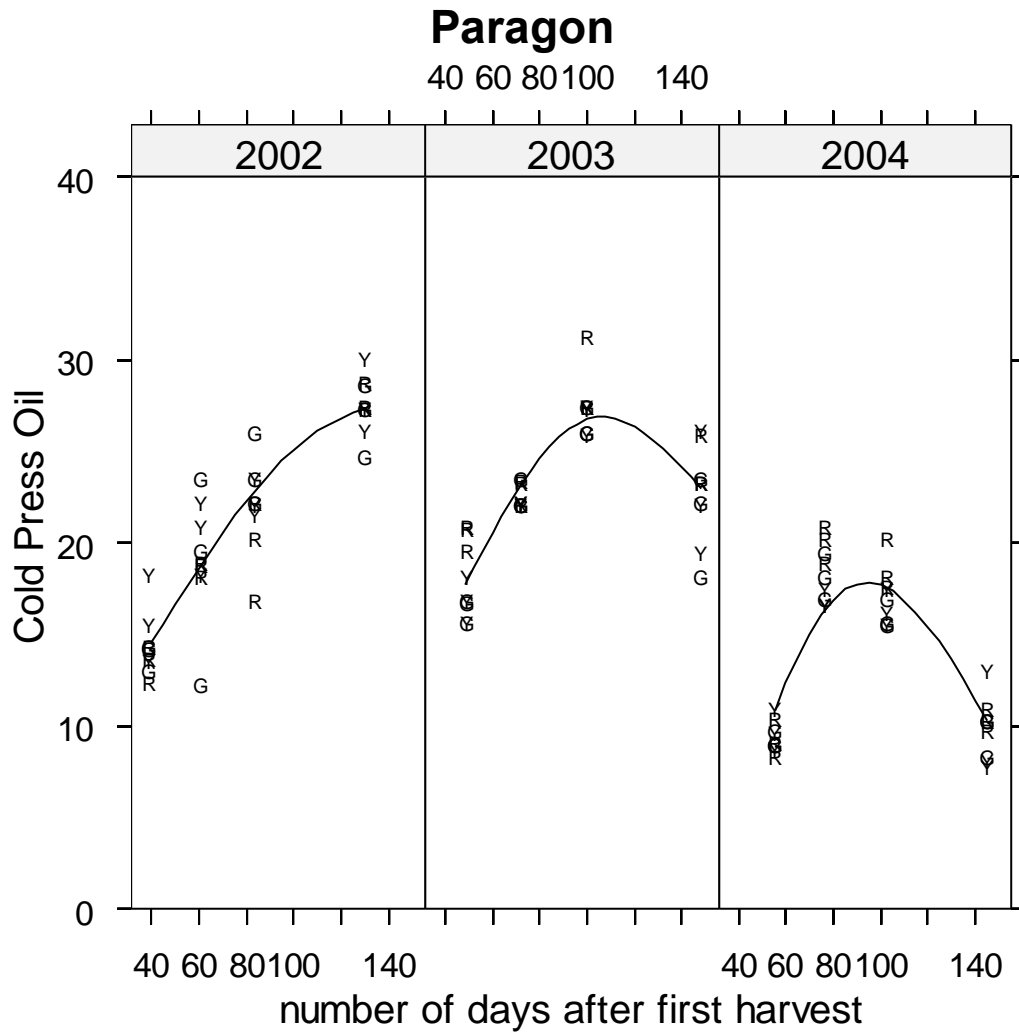


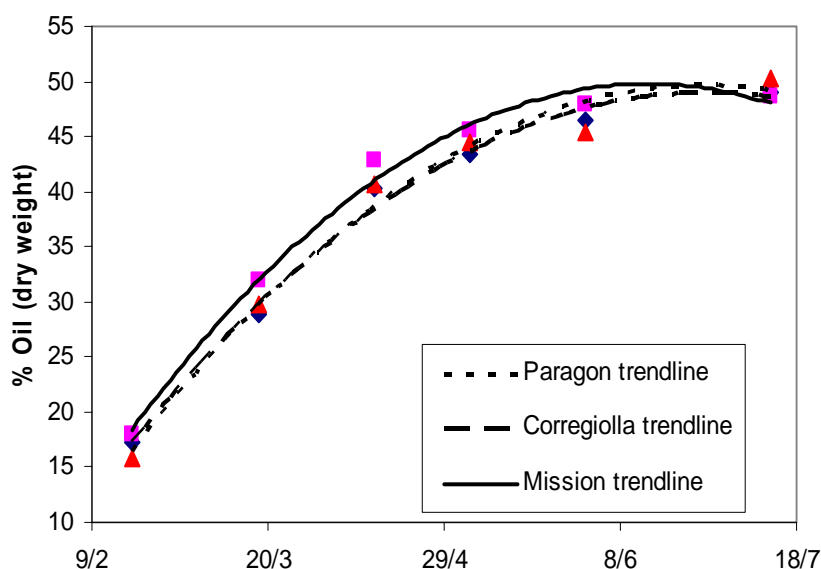
Fig. 4.15 Cold-pressed oil content for Paragon over three years and including three irrigation treatments. G = full ; Y = moderate; R = deficit irrigation. There was a year effect ($p = 0.012$) but little effect for irrigation treatment ($p = 0.229$). The time of harvest had an effect ($p = 0.032$).

4.6.3 Oil content–solvent extraction

Cold press extractions systems leave some oil in the waste and this amount may vary depending on the extraction system, the moisture content, temperature and other factors. Solvent extracted oil is referred to as pomace oil. The oil is totally removed from the fruit by washing with solvents. The oil content is then calculated by evaporating the solvent to determine the absolute amount of oil in the fruit, rather than the recoverable oil discussed in the previous section. The results for solvent extracted oil shown here are therefore not representative of industrial olive extraction processes but merely provide a comparison between cultivars and treatments of the total oil produced. This data can be used to compare oil productivity. It can also be used to determine oil recovery by measuring the difference between cold-press extraction and solvent extraction values.

Solvent extracted oil contents on a dry-weight basis reached almost 50% in all three cultivars. The trendline for oil content of three cultivars over six harvest times is shown in Fig 4.16. There was rapid accumulation of oil at the beginning of the season up to the end of April at which time the rate decreases. It can be seen that the absolute oil content at any point during the six stages was very similar for all three cultivars.

Fig. 4.16 Trendline of oil content-solvent extraction for three cultivars of olives. Each point represents the mean of three replicates over three years.



Similar trends for oil accumulation rates are shown for each year and for each cultivar (Fig. 4.17, 4.18 and 4.19). These were in stark contrast to the cold-press extraction in which cultivars varied significantly in oil content between cultivars. The results of cold press extraction show Paragon to be far superior due to the more efficient oil recovery. It was therefore unexpected that all of the cultivars in this study produced very comparable quantities of oil when averaged over the three years.

Corregiola (Fig 4.17) produced close to 50% oil for each of the three years although there was a significant difference between the three years data ($p = 0.016$). There was clearly an effect of harvest timing ($p < 0.001$) and also some effect for irrigation treatment ($p = 0.075$).

Mission (Fig 4.18) showed similar trends with a strong year effect ($p = 0.008$) and time of harvest ($p < 0.001$). There was no difference in irrigation treatments.

Paragon (Fig 4.19) did not plateau or show a levelling effect but had a continual increase in oil content over the period of the four harvests. The year effect was significant ($p < 0.001$) but irrigation had no effect. The time of harvest had an effect ($p = 0.039$).

It was expected that irrigation effects would have been more obvious between the treatments. A previous study by Goldhamer (1998) reported higher oil content from olives grown under deficit conditions. However, oil content was not significantly affected by irrigation treatments in this study. This may be due to the severe deficits imposed on all of the treatments too late in the season which have overridden potential irrigation effects.

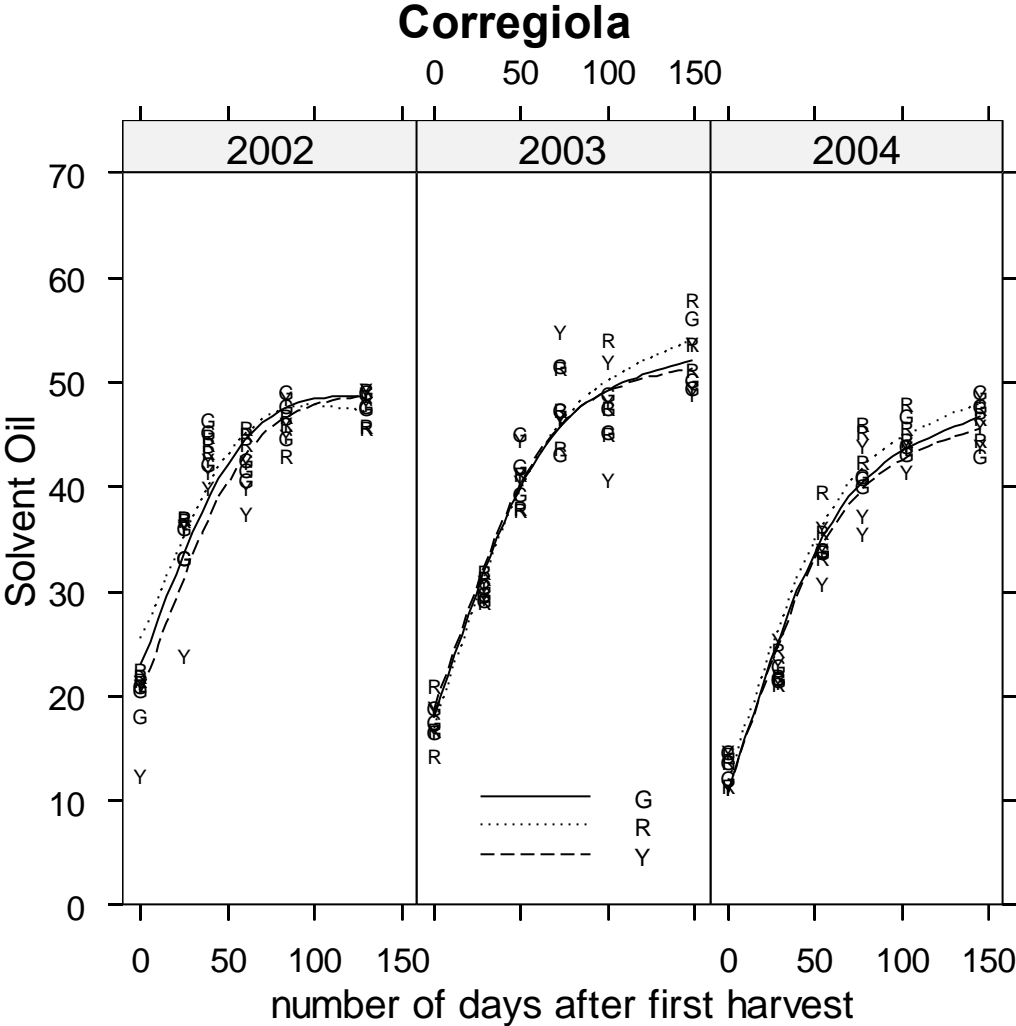


Fig. 4.17 Solvent extracted oil content for Corregiola over three years and including three irrigation treatments. G = full ; Y = moderate; R = deficit irrigation. There was a significant year effect ($p = 0.016$) and irrigation treatment effect ($p = 0.075$). There was also a significant time of harvest effect ($p < 0.001$).

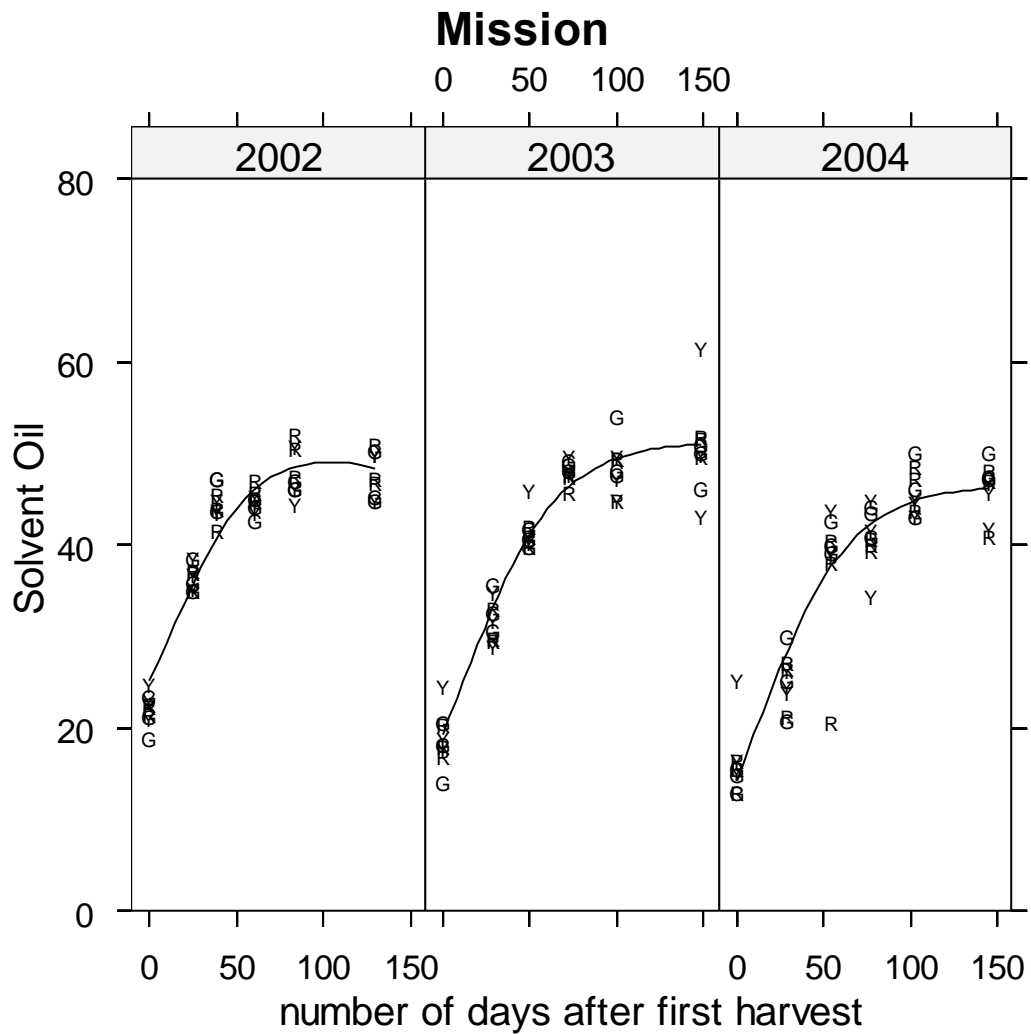


Fig. 4.18 Solvent extracted oil content for Mission over three years and including three irrigation treatments. G = full ; Y = moderate; R = deficit irrigation. There was a significant year effect ($p = 0.008$) no irrigation treatment effect ($p = 0.644$) but there was significant time of harvest effect ($p < 0.001$).

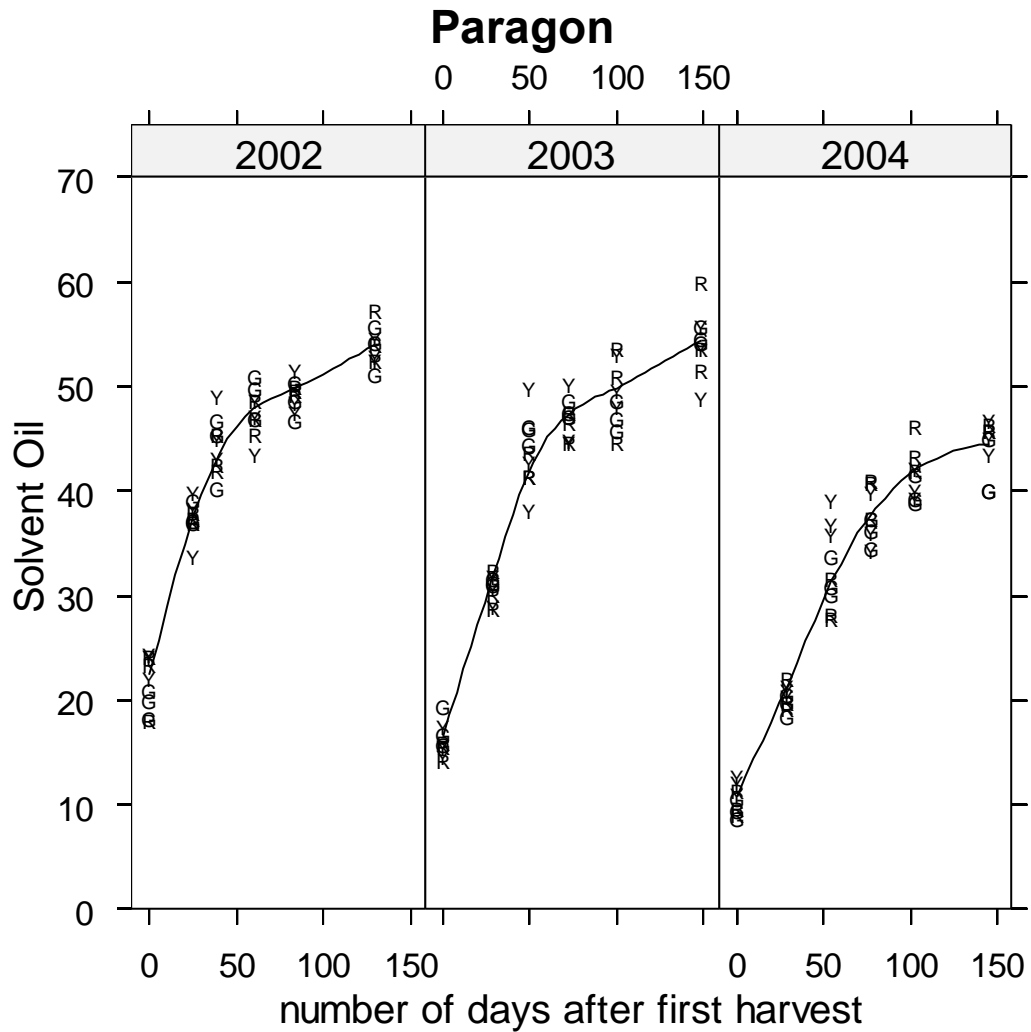
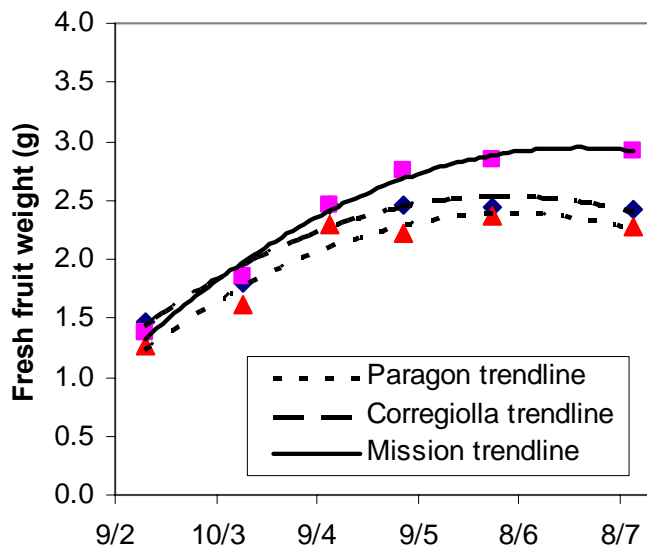


Fig. 4.19 Solvent extracted oil content for Paragon over three years and including three irrigation treatments. G = full ; Y = moderate; R = deficit irrigation. There was a significant year effect ($p < 0.001$) no irrigation treatment effect ($p = 0.589$) and there was also a harvest effect ($p < 0.039$).

4.6.4 Fruit weight

In this study it was shown (Fig. 4.20) that fresh fruit weight increases significantly in olives during the first part of the season, around the time of seed hardening, and then plateaus, and even decreases, during the latter stages of ripening. This is at least partly because, although the dry fruit weight continues to increase throughout the season, the moisture content of the fruit is decreasing (Fig. 4.8), therefore slowing the rate of increase of the fresh fruit weight.

Fig. 4.20 Trendlines of fresh fruit weight (includes moisture) in three cultivars of olives. Each point represents the mean of three replicates over three years.



Mission fresh fruit weight was consistently higher than the other two cultivars, Corregiola and Paragon (Fig. 4.20). However, Mission oil recovery from cold-press extraction was the lowest at the final harvest date. It can be seen therefore that the measurement of fresh fruit weight is misleading due to the variability of moisture differences which can significantly alter the fresh fruit weight.

Corregiola (Fig 4.21) varied considerably between years ($p < 0.001$) and also showed a large difference between harvest dates ($p < 0.001$). Irrigation had no influence on fresh fruit weight ($p = 0.820$).

Mission (Fig 4.22) had almost the same response as *Corregiola* over the three years with year effects ($p < 0.001$) and harvest timing effects ($p < 0.001$) but no significance between irrigation treatments ($p = 0.283$).

Paragon was different to the other two cultivars in that it had significant effects for timing ($p < 0.001$) although less significant for years ($p = 0.027$) but it also showed clear irrigation effects ($p < 0.001$). It is difficult to explain the reason for this other than the fact that Paragon may be more sensitive to changes in soil moisture than the other cultivars. The effect was not so clear in the first two years but very obvious in 2004 (Fig 4.23).

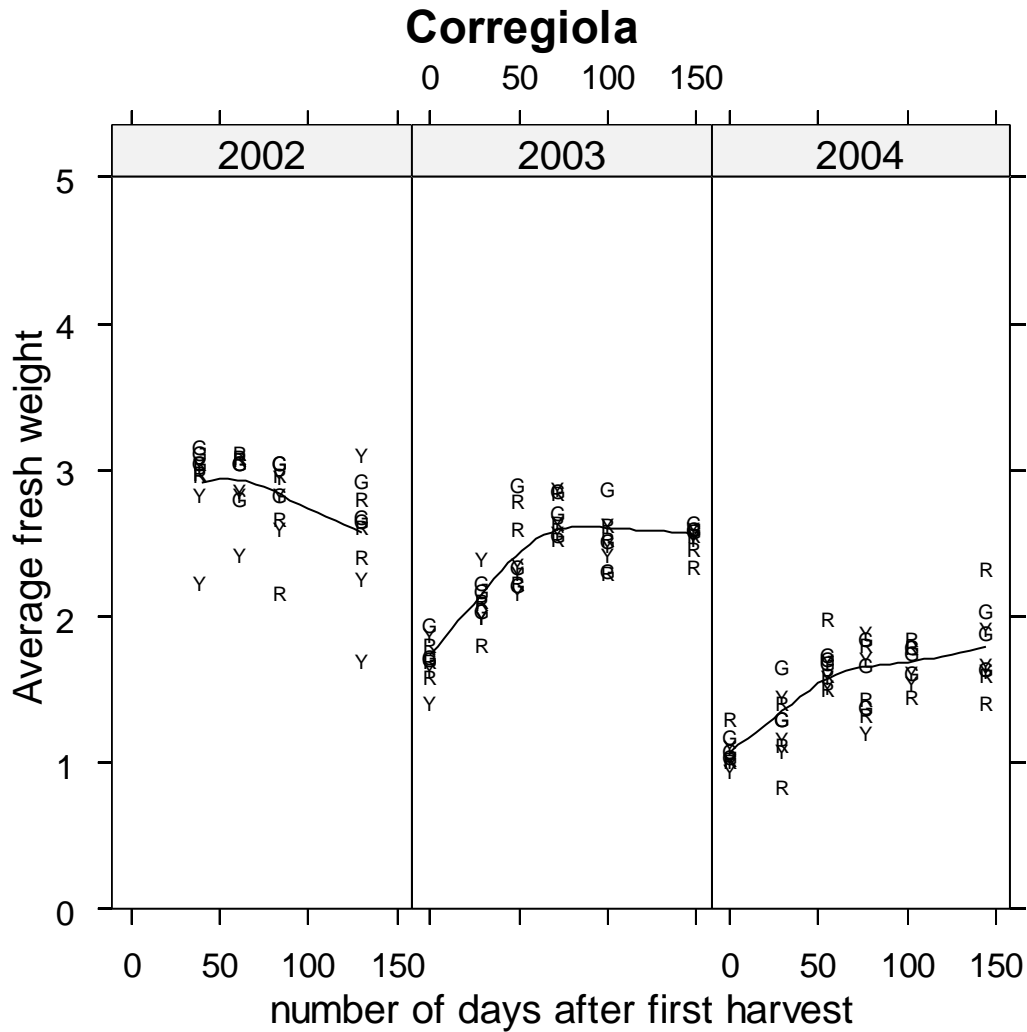


Fig. 4.21 Fresh weight of Corregiola at six harvest dates over three years including three irrigation treatments. G = full ; Y = moderate; R = deficit irrigation. There was a significant effect between years ($p < 0.001$) and no irrigation effects ($p = 0.820$). There was also a significant time of harvest effect ($p < 0.001$).

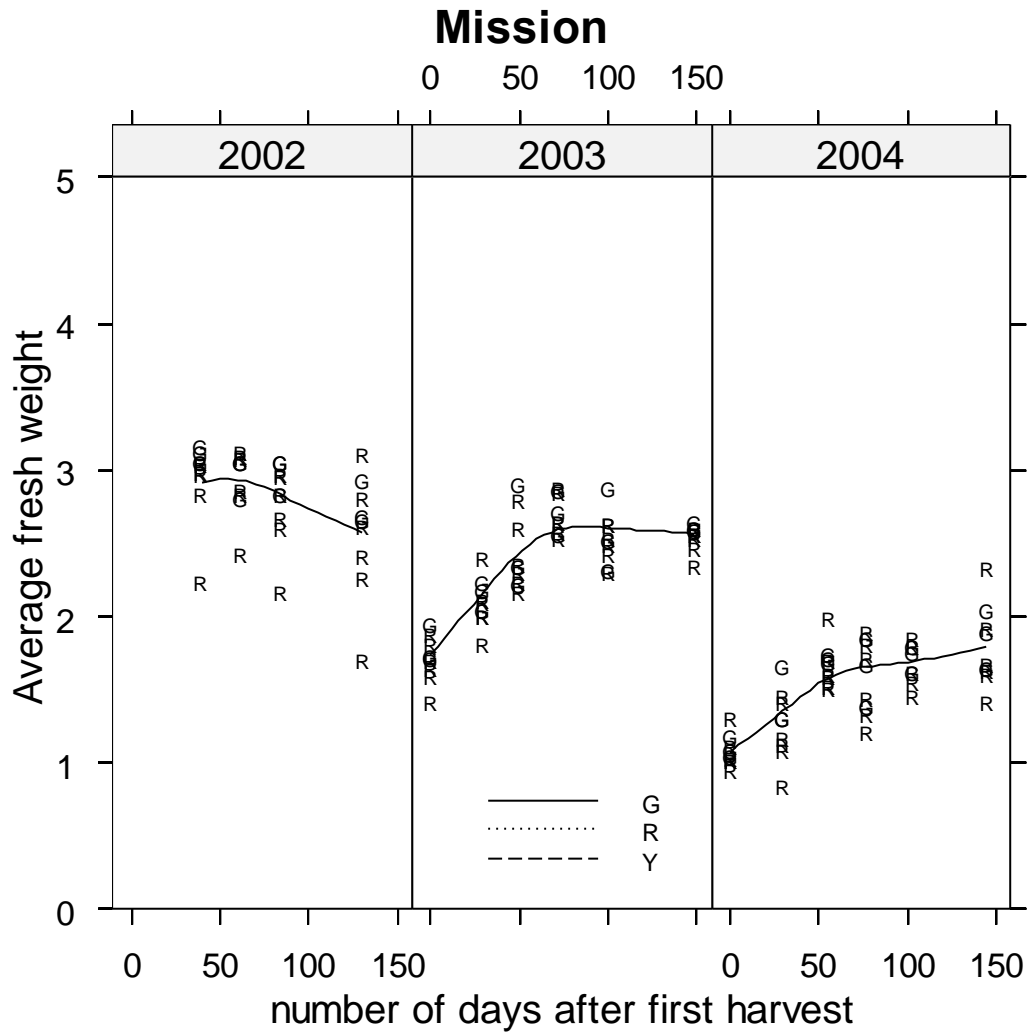


Fig. 4.22 Fresh weight of Mission at six harvest dates over three years including three irrigation treatments. G = full ; Y = moderate; R = deficit irrigation. There was a significant effect between years ($p < 0.001$) and no irrigation effects ($p = 0.283$). There was also a significant time of harvest effect ($p < 0.001$).

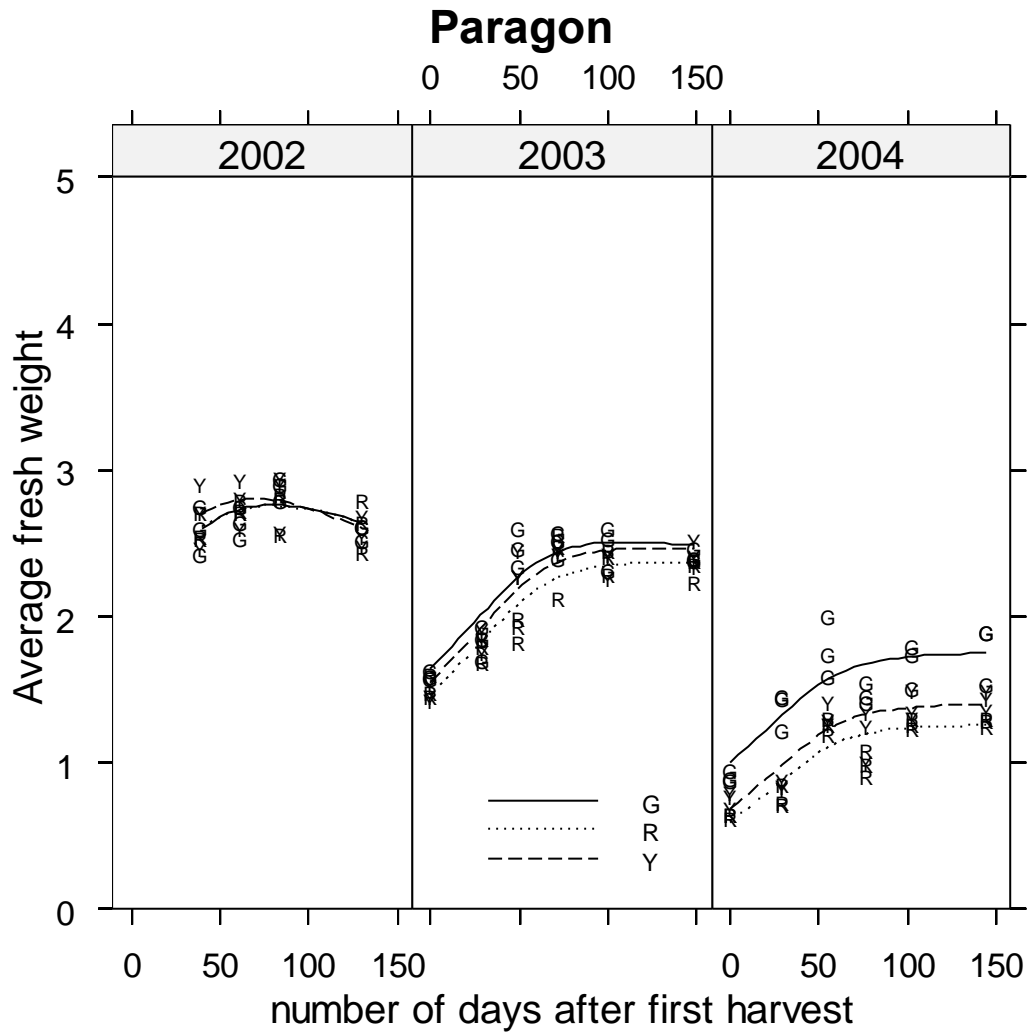
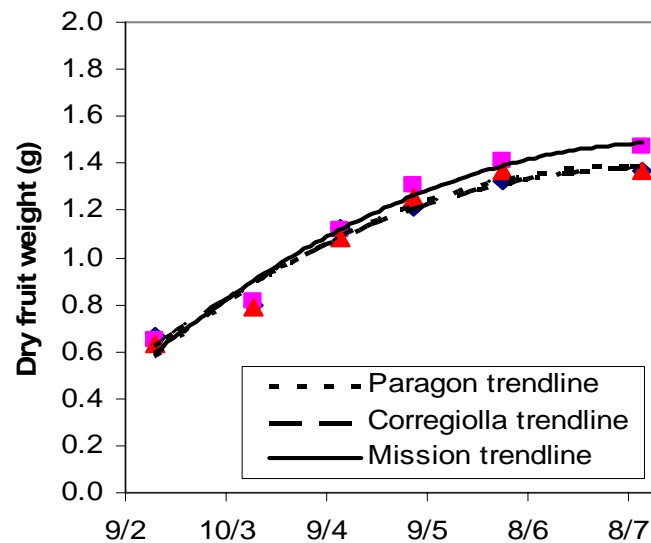


Fig. 4.23 Fresh weight of Paragon at six harvest dates over three years including three irrigation treatments. G = full ; Y = moderate; R = deficit irrigation. There was a significant effect between years ($p < 0.001$) and an irrigation effect ($p < 0.001$). There was also an effect for harvest timing ($p=0.027$).

Fig. 4.24. Trendlines of dry fruit weight in three cultivars of olives. Each point represents the mean of three replicates over three years.



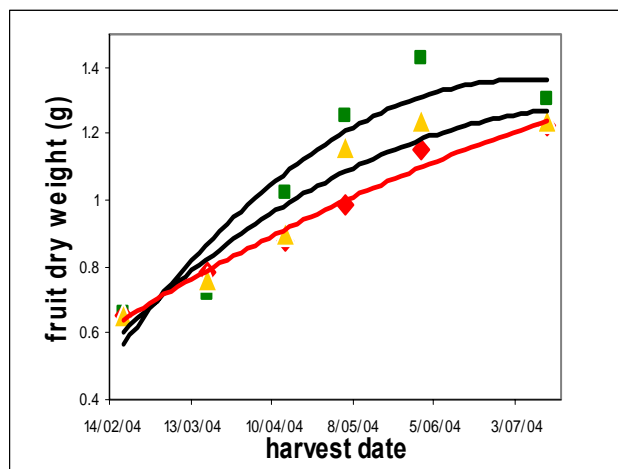
There is a requirement by olive growers to have indicators which can be used in the field to identify when oil content has reached its maximum. Previous studies have suggested that fruit weight increases simultaneously with oil accumulation in the fruit. The increase in fruit weight is masked by the variability in changes in moisture content which have been shown to occur (4.6.1). The use of dry weight may therefore be a better indicator. To determine this, we needed to determine both fresh fruit weight and the moisture content.

When the fruit is dried, the weights of the three cultivars were found to be the same, as shown in Fig. 4.24. This difference in fresh and dry weights would be expected based on the data discussed previously, showing Mission moisture to remain relatively higher than the other cultivars throughout the maturation process (Fig. 4.10). Dry fruit weight is therefore a more accurate measure of the increase in the weight of the fruit, as other influences have been removed. The variation shown in fresh fruit weight in Fig. 4.20 is due to moisture in the fruit.

Dry weight continues to increase throughout the season, with no plateau as was observed with fresh fruit weight. As expected, the responses of solvent extracted oil content (Fig 4.16) and dry fruit weight (4.24) showed similar trends. This close relationship shows the possibility of using dry fruit weight as an indicator of oil accumulation and therefore fruit maturity.

The overall effect of irrigation on dry weight was not significant between treatments. The data in 2004, at which time better control had been established with irrigation, has been plotted in Fig. 4.25. In this year alone, Corregiola, Paragon and Mission showed some differences in fruit dry weight between irrigation treatments. Fruit dry weight of the low irrigation treatments in Paragon and Corregiola were low at the time of the first harvest on 18 February and remained lower than that of the high irrigation treatment for the duration of the season. This indicates that moisture stress applied in stage II (cell division) and stage III (seed hardening) has reduced potential fruit weight which has then remained low due to continuing water deficits. On 5 May fruit dry weight of the 'R' treatment was 32 percent lower than the 'G' treatment in Paragon and 18 % lower in Corregiola. There was no difference for Mission in fruit dry weight between irrigation treatments on 18 February. However dry weight differences developed as the season progressed and on 5 May fruit dry weight of the 'R' treatment was 22% lower than in the 'G' treatment.

Fig. 4.25 Trendlines of dry fruit weight in *cv. Mission* for three irrigation treatments for the year 2004. Each point represents the mean of three replicates. R = — ; Y= — ; G= —



4.6.5 Oil Recovery

Oil recovery from cold-press extraction is a major consideration for olive producers and, as shown here (4.6.2) is as important as oil production. The issue of poor oil recovery are important to consider when deciding which cultivars to grow and when to harvest to achieve the highest possible returns.

Oil recovery refers to the amount of oil obtained from cold-press extraction. This is in contrast to the total oil content which refers to the total amount of oil in the fruit. This is equivalent to the oil from the cold-press extraction plus the oil remaining in the waste (pomace) after processing. Ideally, the oil from the cold-press should be maximised with minimal waste.

Data presented in Table 4.2 are an average of oil and moisture contents over three years. They include four harvest times for each of three cultivars. As shown in Fig. 4.9, 4.10 and 4.11, the moisture content decreases over the maturation period. In the same period, total oil content for all cultivars, that is the solvent extracted oil, is seen to increase over the entire maturation period. This has also been illustrated in Fig. 4.17, 4.18 and 4.19.

Cold press extracted oil content (dry weight) however, shows that oil recovery diminishes in all cases in the final harvest. Oil recovery dropped from 67 to 24% in Mission and from 85 to 64% in Paragon. This was been shown to be a result of the inefficiency in the cold-press system to remove oil from the matured fruit. Although this is often related to the moisture content of the fruit at the time of processing, it can be seen from Table 4.2 that the moisture content of Mission at the third harvest is 50.3% and the oil recovery was 63.6%. By the fourth harvest, the moisture content had dropped only 0.6% to 49.7%. At this time the oil recovery had dropped to 24.1%. This would indicate that moisture plays only a minor role in the poor recovery of oil from Mission.

Oil recovery by cold-press is much higher in Paragon than Corregiola and to an even greater extent than Mission. Again this may be related to moisture but these results suggest that the moisture is not strongly related to oil recovery. At the third harvest, Paragon had 42.1% oil and 84.8% recovery. At the fourth harvest, Corregiola had 43.5% moisture and only 56.7% oil recovered.

These figures suggest that moisture content is not the major factor in determining oil recovery. The only apparent link between moisture content and extraction efficiency by cold press extraction is that Paragon, with consistently the lowest moisture content, also had the highest oil extraction rate. This is in contrast to Mission which maintained high moisture content throughout the maturity period and had very low oil recovery.

Management of irrigation toward the end of fruit maturity is considered important to help reduce moisture content in the fruit and therefore increase oil recovery. This has not been shown in this study where moisture availability was low for all crops at the last harvest but perhaps highest in Paragon (Fig 4.11). Despite this, the fruit moisture content in Paragon dropped to the lowest level (Fig 4.8) of the three cultivars. It would appear that although moisture was available to Paragon, the moisture level reduced. In contrast, Mission, with limited moisture availability remained high in moisture throughout the maturation period. These findings indicate there are genetic differences between the cultivars, similar to the differences shown for moisture and oil recovery discussed previously.

It is possible that high moisture contributes to poorer grinding and rupture of oil cells to access the oil. It may also provide less resistance in the malaxer. However, it is unlikely that a relatively small difference in moisture content explains the dramatic differences seen here (Table 4.2) for oil recovery.

Table 4.2 Comparison of moisture content and recovery of oil from three olive cultivars over six harvest periods.

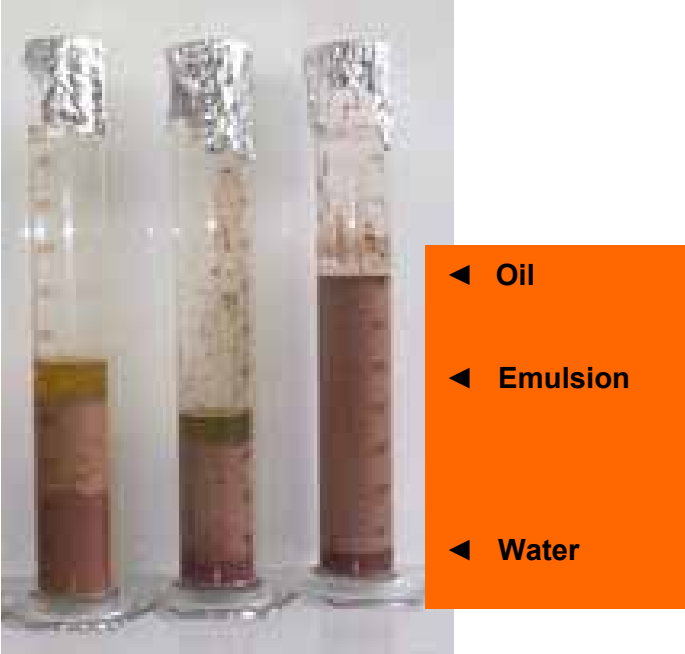
Harvest Date	Moisture (%)	Oil - Solvent (% dry wt)	Oil -Cold Press (% dry wt)	Oil recovery (%)
Mission				
13/4	54.80	42.75	23.67	55.37
5/5	52.42	45.66	30.6	67.02
31/5	50.33	47.92	30.46	63.57
12/7	49.71	48.61	11.73	24.13
Corregiola				
13/4	54.21	40.37	25.77	63.83
5/5	50.42	43.33	26.97	62.24
31/5	45.53	46.47	31.63	68.07
12/7	43.46	49.05	27.80	56.68
Paragon				
13/4	52.69	40.58	28.13	69.33
5/5	43.60	44.56	35.25	79.10
31/5	42.15	45.41	38.50	84.77
12/7	39.83	50.26	32.28	64.22

Cold-press processing produces three phases including oil and water separated by an emulsion. Fig. 4.26 shows examples of three cold-press extractions typical of three stages of maturity. The left cylinder is from young olives whereas the right cylinder is an extract from well matured Mission fruit. The middle cylinder is from a mid maturity group.

The less mature fruit has good oil recovery and low emulsion content. The slightly more mature fruit produces more emulsion and less oil on top. The right hand cylinder of very mature olives formed an emulsion immediately on malaxing and almost all of the oil remains trapped in the emulsion. Sample one showed reasonable recovery whereas samples two and three had progressively stronger emulsions and less oil was recovered.

The poor recovery of oil is related to the consistency of the emulsion. This emulsion may be the result of decomposing flesh in the old fruit. It may also be caused by surfactants or emulsifying reagents within the fruit extract. It is clear by these studies that the problem is more pronounced in some cultivars than others.

Fig. 4.26 Oil recovery from cold-pressed olive oil. The cylinders from left to right contain three olive samples, young fruit, mid maturity and old fruit. The oil layer on top of each cylinder is less from left to right with the age of the fruit. At the same time, the proportion of emulsion in the middle between the oil and water becomes greater and more stable.



4.6.6 Oil potential

In industry the most common form of yield potential assessment is done by measuring oil content of the fruit. Because oil content tends to increase slowly during the ripening period many producers assume there will be little further increase in yield when oil content reaches 18-20% of fresh weight. But this assumption does not take into account that fruit dry weight continues to increase during ripening and this growth combined with moderate increases in oil percentage can lead to substantial yield increases during the ripening period.

To accurately assess changes in yield potential between harvest dates we have multiplied fruit dry weight by oil percentage (expressed as a % of fruit dry weight) to give us oil yield per fruit. While the yield potential for all harvests is presented to provide an overview of yield development, the discussion of this data (Table 4.3) is confined to the last four harvests covering the period from early April to early July as these have relevance to commercial harvest period for olives.

The data show an increase in oil yield per 100 g fruit from early April to late April (3 week interval) of 16, 27 and 26 % for Corregiola, Paragon and Mission respectively. This is followed by an increase of 17, 11 and 13 percent between early and late May. From the end of May until early July there is a further 9, 11 and 6 percent increase for the three varieties although it is likely that the maximum yield is reached before this final harvest date.

The total potential yield increase between the mid April harvest and the late May harvest is 36%, 41% and 42 % for Corregiola, Paragon and Mission respectively. The early harvest date, in early to mid April is just 2 weeks after the beginning of fruit maturation and would be too early for most growers. This study illustrates that it is clearly a period where rapid oil synthesis and fruit growth is still occurring. The harvest window could commence by the fourth week in April and extend until the third or fourth week of May. In this scenario the yield compromise associated with the harvest start date could be over 20%. Consideration needs to be given to the loss of ability to recover some of this increased production from fruit as it becomes more mature.

It is clear signal that there is a quantifiable yield cost associated with harvesting olives too early. There is a requirement for the development of price differentials based on fruit maturity and quality specifications. A later start date for harvest will yield more olive oil in all of the varieties tested in this study. Harvesting from late May into June would achieve the highest yield. However there is potential for quality and crop losses the longer the fruit is left on the trees.

Table 4.3 Average oil yield (oil content x dry fruit weight) at 6 harvest dates for Corregiola, Paragon and Mission, over 3 years.

Oil yield potential per 100 g fruit			
	Corregiola	Paragon	Mission
18/2/2004	11	10	12
18/3/2004	23	23	26
13/4/2004	45	44	48
5/5/2004	53	56	60
31/5/2004	62	62	68
12/7/2004	67	69	71

4.7 Olive oil quality

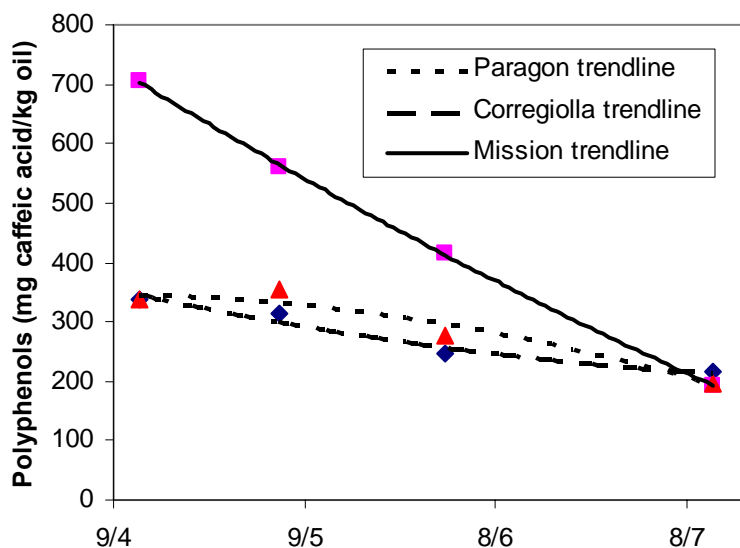
4.7.1 Total polyphenol content

Polyphenols are perhaps the most important of the minor components in olive oil due to the powerful antioxidant effect they have on the oil and the resulting contribution to shelf life stability. Young olives are typically high in polyphenol content and this component has been shown to relate closely to oil stability (Mailer *et al.*, 2002). The polyphenols also have been shown to be closely associated to the organoleptic characteristics of the oil, being largely responsible for pungency and bitterness attributes. Sensory analysis has shown that young oils are generally more pungent and bitter than oil from mature fruit and again this is related to the polyphenol content.

Polyphenols are a very broad range of chemicals clustered together under one term. Considerable research has been carried out on the individual members of this group but for the purpose of this study, the analysis carried out has been to group the compounds in one group as total polyphenol components.

This study has identified the cultivar Mission as generally having considerably higher levels of polyphenols than the other cultivars. The fruit of all cultivars had very high levels at early maturity which decreased as the fruit matured. For each cultivar, there was virtually a linear reduction with time, based on the analysis of three replicates of each harvest over three years, as shown in 4.28. It can be seen that Mission's polyphenol content decreased to levels similar to the other cultivars as the fruit matured. All cultivars maintained their relative order throughout the maturity period. Mission had the highest level at 7/4/03 with 857mg/kg with the lowest level in Corregiola at 68 mg/kg on 23/5/02.

Fig. 4.27 Trendline of total polyphenols for three cultivars of olives. Each point represents the mean of three replicates over three years.



It is often suggested that levels above 200 mg/kg are necessary to produce good quality EVOO. Levels above 400 mg/kg may be too high for the average consumer, producing a pungent oil. From Fig 4.27 it can be seen that polyphenol levels in Mission were above 400 mg/kg until early June. All cultivars in this study maintained reasonable levels of polyphenol on average, even into late maturity. This is not always the case with some Australian oil producing very low levels, below 100 mg/kg (Mailer 2004).

The individual performance of the three cultivars is illustrated in Fig. 4.28, 4.29 and 4.30. Generally, all cultivars showed similar trends, with low levels of polyphenol in the first year and increasing to higher levels in subsequent years.

Corregiola (Fig 4.28) had insignificant levels in 2002, starting at less than 200 mg/kg in young fruit and reducing to less than 100 mg/kg as they matured. In 2003 the polyphenols ranged from 350 down to 150 mg/kg. however, in 2004, polyphenols started in young fruit at above 600 mg.kg and even at full maturity, contain more than 400 mg/kg. *Corregiola* had significant effects over the three years ($p < 0.001$) and significant differences between time of harvest ($p < 0.001$). The irrigation effect was insignificant over the three years ($p = 0.184$).

Mission (Fig 4.29) also showed increased levels of polyphenols over the three years. Again they concentrations started at a high level and decreased with maturity. The final harvest of *Mission* produced only 100 mg/kg in the first year but around 300 mg/kg in the third year. Again, the year effect was significant ($p = 0.009$) and there was a time of harvest effect ($p = 0.010$). There was again no irrigation effect ($p = 0.787$).

Paragon (Fig 4.30) was quite different to the other two cultivars showing different effects for the three years. The first year concentrations remained almost constant over the year. In 2003, the levels started high, around 600 mg/kg, and decreased dramatically too around 200 mg/kg over the season. In 2004 the levels started low, increased to a maximum at mid maturity and then decreased to levels slightly lower than the original concentration of the young olives. For this cultivar alone there was an irrigation effect ($p < 0.001$) with highest moisture treatments producing the least concentration of polyphenol. As for the other two cultivars, there was a year effect ($p < 0.001$) and a time of harvest effect ($p < 0.001$).

The reasons that *Paragon* alone had an irrigation affect are not clear but it would seem that this cultivar is more sensitive to water availability/stress than the other cultivars. This cultivar also showed an irrigation effect for fresh fruit weight, unlike the other cultivars. However, the moisture content of the fruit indicated that it had higher moisture in the early part of fruit development but decreased in line with the other cultivars toward maturity (Fig. 4.11).

Table 4.4 Mean values for polyphenol concentration in *Paragon* calculated for three irrigation treatments across three years and for three replicates.

Year	Irrigation	Polyphenol mean
2002	G	110.81
2002	R	102.13
2002	Y	125.22
2003	G	334.01
2003	R	417.80
2003	Y	390.17
2004	G	442.15
2004	R	683.66
2004	Y	597.91
SED		55.50
LSD(5%)		127.983
LSD(1%)		186.202

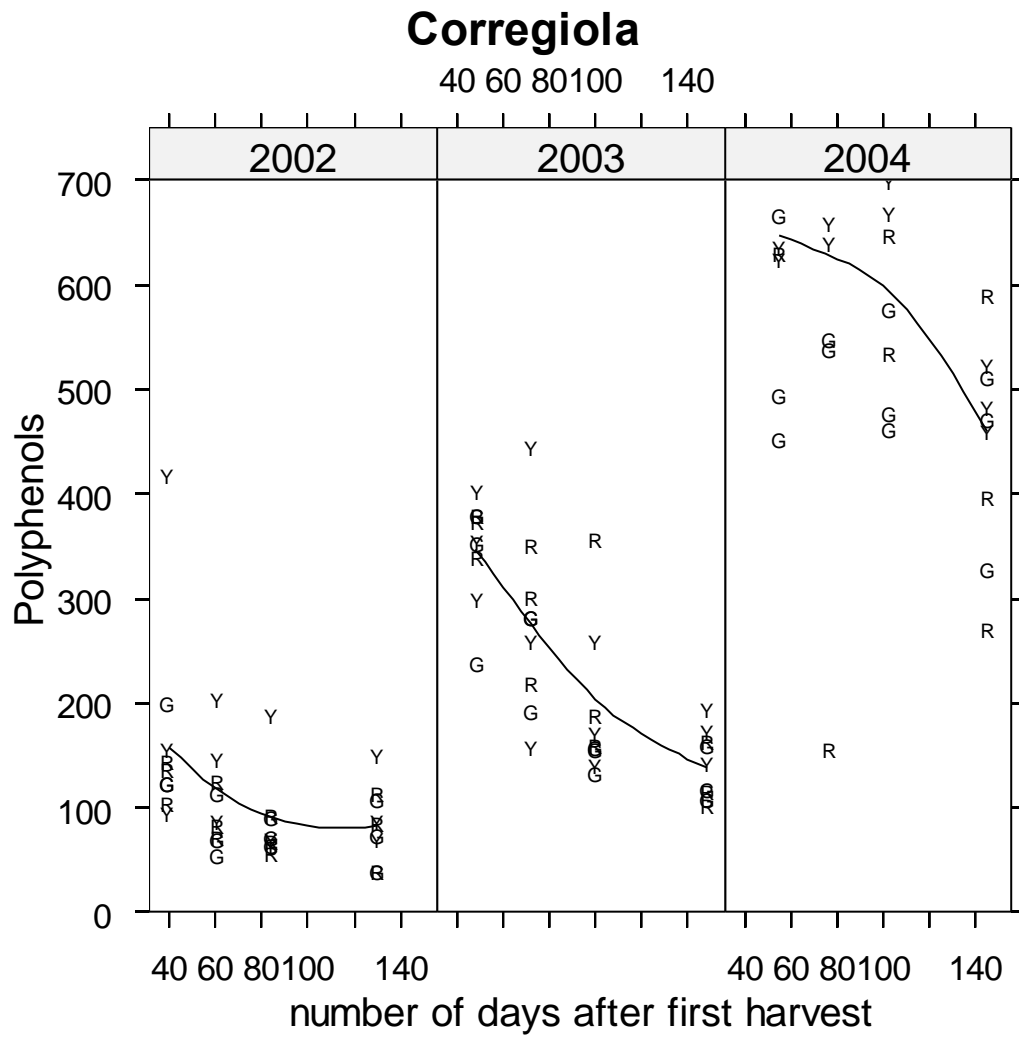


Fig. 4.28 Polyphenol content of Corregiola at four harvest dates over three years including three irrigation treatments. G = full ; Y = moderate; R = deficit irrigation. There was a significant effect between years ($p < 0.001$) but little irrigation effect ($p = 0.184$). There was a significant effect for harvest timing ($p < 0.001$).

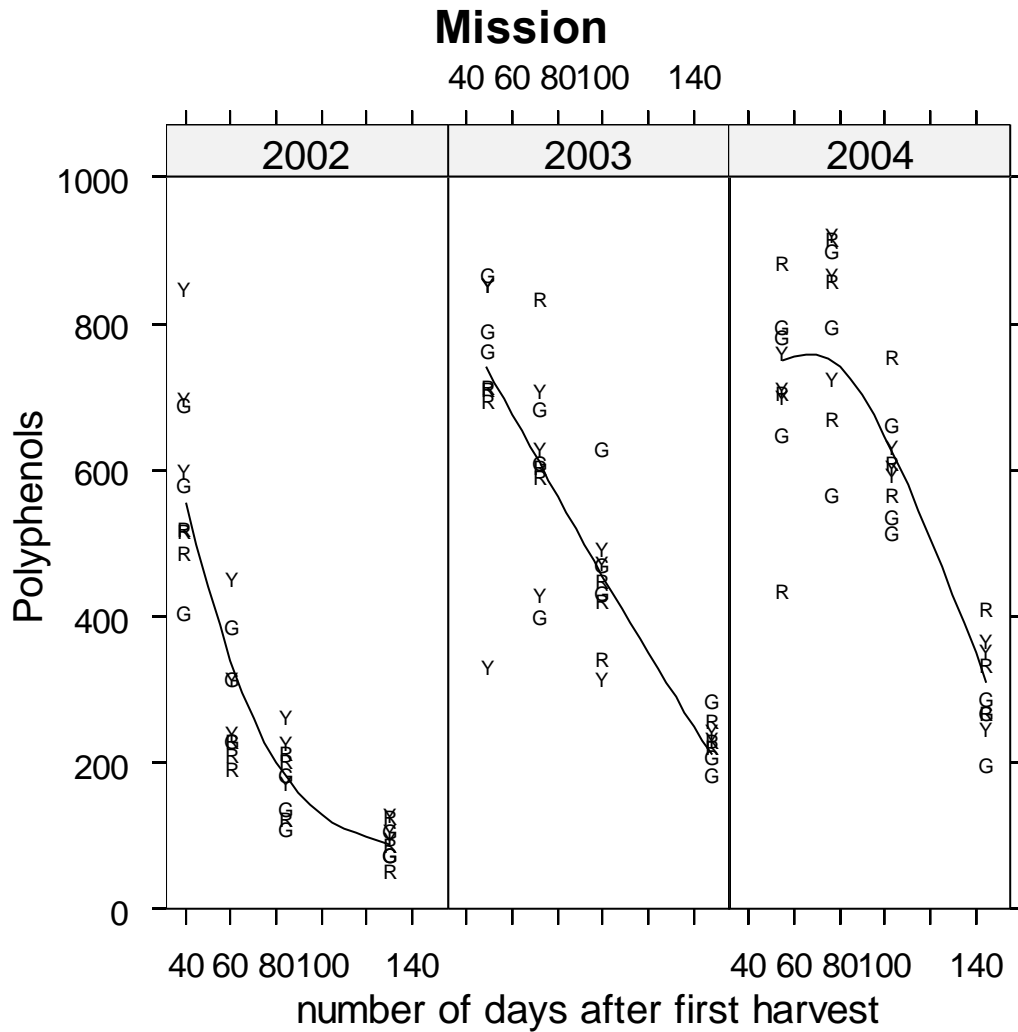


Fig. 4.29 Polyphenol content of Mission at four harvest dates over three years including three irrigation treatments. G = full ; Y = moderate; R = deficit irrigation. There was a significant effect between years ($p < 0.001$) but no irrigation effect ($p = 0.787$). There was an effect for harvest timing ($p = 0.010$).

Paragon

40 60 80 100 140

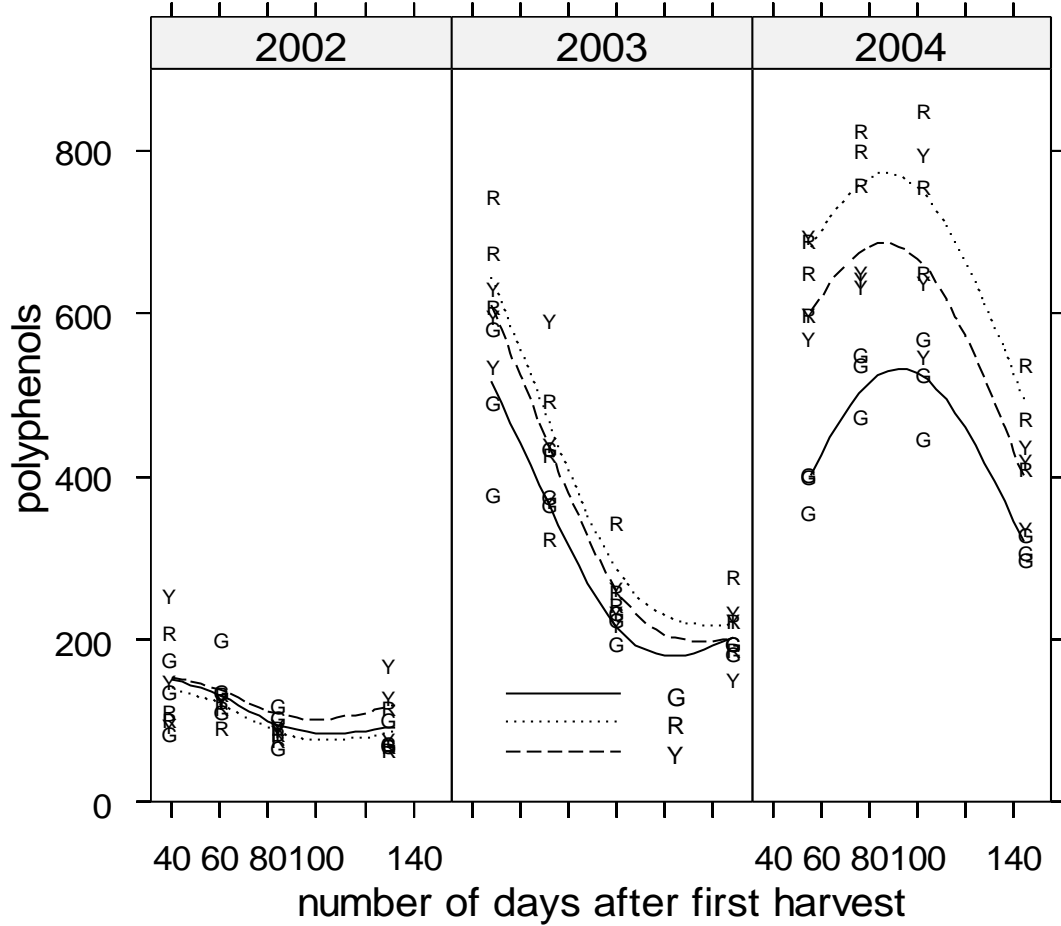


Fig. 4.30 Polyphenol content of Paragon at four harvest dates over three years including three irrigation treatments. G = full ; Y = moderate; R = deficit irrigation. There was a significant effect between years ($p < 0.001$) and an irrigation effect ($p < 0.001$). There was also an effect for harvest timing ($p < 0.001$).

An interesting observation over three years was that each year, all cultivars showed an increase in polyphenol content overall. Table 4.4 shows the case for Paragon with average levels from 102 mg/kg in the first year to 684 mg/kg in the third year. There are several possible reasons for this. Firstly, tree age could have had an influence.

- In 2002, the trees were 4-5 years of age, they had vigorous vegetative growth and variable yields from tree to tree, typical behaviour for young trees. Vegetative growth provides a dominant sink for nutrients in young trees and this will have had an influence on fruit development and growth. It is possible that this has contributed to lower than expected polyphenol levels in all varieties in 2002.
- In year 2003 polyphenol levels were similar to those observed in industry, starting high and reducing in all varieties as ripening progressed.
- In 2004 polyphenols levels were unusually high in all varieties. The trees at this time were well established and producing good yields.

Moisture stress on the developing and ripening crop is also a likely cause for increased polyphenols. Fruit moisture was 45-50% on 18 February 2004 indicating a degree of moisture stress in the developing crop which would normally have fruit moisture levels of 55-60% under stress free conditions. Long intervals between irrigation events meant that the crop was subjected to periodic moisture stress throughout the season and the moisture deficit was almost continuous from mid March onwards, (see section 4.2.3 Crop water inputs) leading to almost continuous moisture stress in late fruit development, maturation and ripening. The imposition of moisture deficits on the crop is the predominant reason for the very high polyphenol levels in all varieties in 2004 and this is consistent with the observations of other researchers (Tovar *et al.*, 2002) who observed higher polyphenols in crops subjected to irrigation deficits.

Irrigation effects on oil quality and yield were not measurable in 2002 and 2003 due to problems associated with the irrigation treatments in these seasons. In 2004 there were effects associated with the deficit irrigation treatments on polyphenol levels in Paragon but not Mission or Corregiola. This is interesting in that Paragon is a variety that appears to respond dramatically to changes in soil moisture status with more dramatic fluctuations in fruit moisture than Corregiola or Mission. Paragon was also the first variety to show foliar signs of moisture stress in the field. It was also the variety where fruit size was most dramatically affected by the deficit irrigation treatments.

In our study the main period of prolonged moisture deficit common to all treatments was in late fruit development, maturation and ripening due to the cessation of irrigation in early March. We must assume therefore that part of the reason for a dramatic elevation in polyphenol levels in 2004 (compared to 2002 and 2003) across all treatments and varieties was because of a physiological response in the fruit to soil moisture deficits in late stage 4 and throughout stage V of fruit development. The additional increase in polyphenol levels in the Paragon deficit treatments was most likely due to multiple causes. Polyphenols are concentrated in and near the skin of fruits and the surface area of skin per kg of fruit is greater when fruit size is smaller. The use of RDI in wine grapes is based on the idea of producing smaller berries which have higher levels of polyphenol per kg of fruit and more intensive flavours. This is also a likely reason for the increase in polyphenol levels in the all of the treatments in Paragon and other cultivars where moisture availability was limiting.

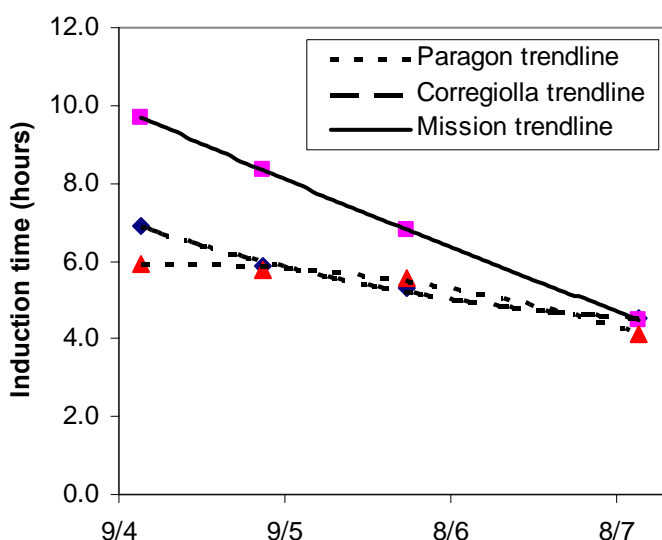
Clearly further work needs to be done to define and quantify the effects of deficit irrigation on polyphenol levels and other quality characteristics of olive oil. Many producers entering the market want olive oils that are not too bitter or pungent, so being able to prevent elevated polyphenol levels or reduce polyphenol levels will be just as important as being able to elevate polyphenol levels where a robust oil characteristics and high stability are required. The stage or stages of fruit development where crop water management will influence polyphenols synthesis is of great interest to olive oil producers. Understanding this could allow growers to exert control over the polyphenol levels in their olive oil.

4.7.2 Induction time

Induction time is a method commonly used to indicate the relative stability of oil in storage. It involves passing oxygen through the oil while the oil is held at an elevated temperature, thereby causing the oil to oxidise. If the oil is stable, it will resist oxidation and result in a long induction time. However, unstable oils will have relatively short induction times. Although induction time in hours cannot be used to precisely represent a particular shelf or storage life it is useful for comparing oils to determine relative stability. The conditions under which the oil is stored will have a major influence on the storage ability. Induction time merely indicates the stability of oil in relation to others when stored under similar conditions.

Induction time for the three cultivars in this study were very different with times ranging from 1.6 hours (Corregiola – 8/7/02) to 12.2 hours (Paragon 5/5/04). There was a consistent trend for induction time to decrease as samples matured (Fig 4.31). Mission consistently produced higher induction times on average for the three replicates over three years. Corregiola and Paragon were very similar over the study period.

Fig. 4.31 Trendline of induction time for three cultivars of olives. Each point represents the mean of three replicates over three years.



Induction time was significantly different for each of the cultivars for each of the three years as can be seen from Figs 4.32, 4.33 and 4.34.

As for polyphenol content, Paragon and Corregiola had very short induction times in 2002 and became progressively longer over the three years. Mission also increased over the three seasons, but not to the same extent. All showed significant differences between years (Tables 8.1, 8.2 and 8.3). There was again a strong effect of irrigation on Paragon but this was not apparent for the other two cultivars. The effect of harvest timing was less obvious for Mission than for the other two cultivars.

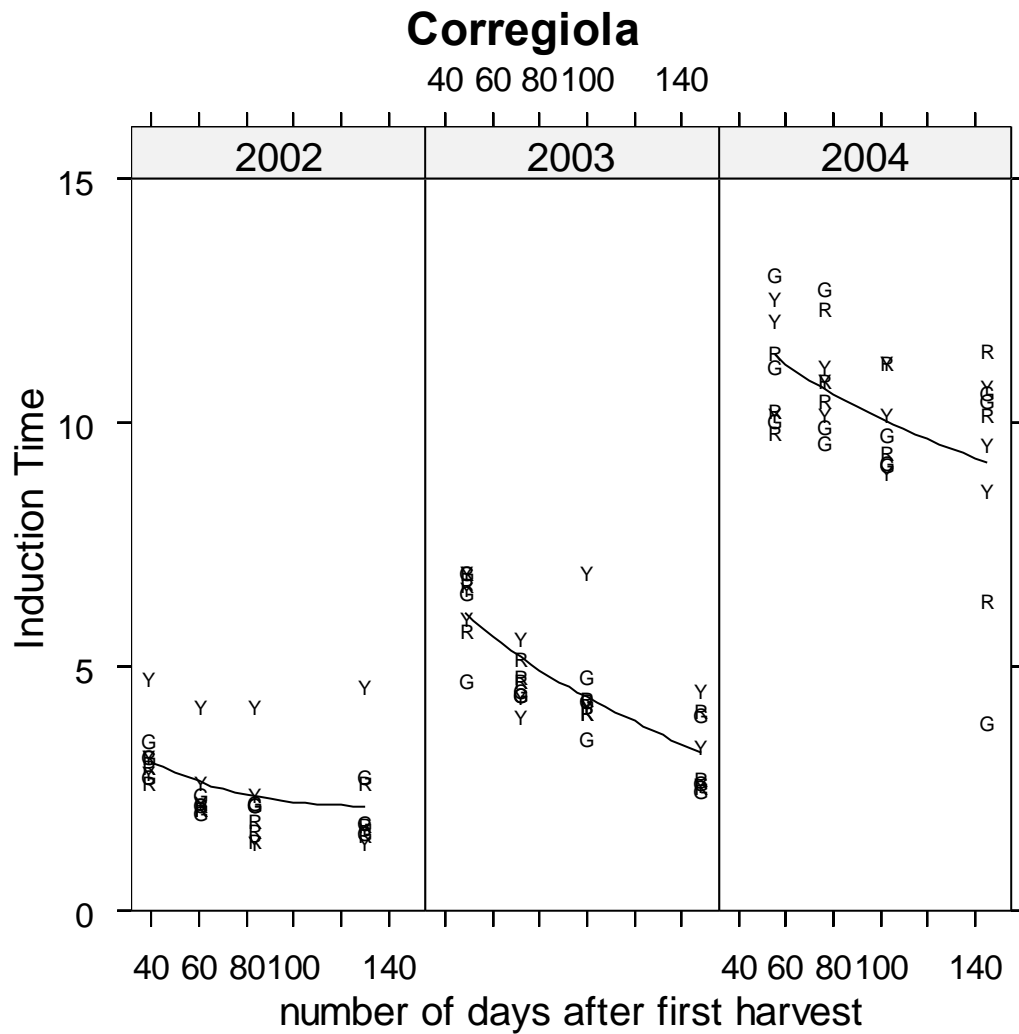


Fig. 4.32 Induction time of Corregiola olive oil at four harvest dates over three years including three irrigation treatments. G = full ; Y = moderate; R = deficit irrigation. There was a significant effect between years ($p < 0.001$) and no irrigation effect ($p = 0.417$). There was also an effect for harvest timing ($p = 0.002$).

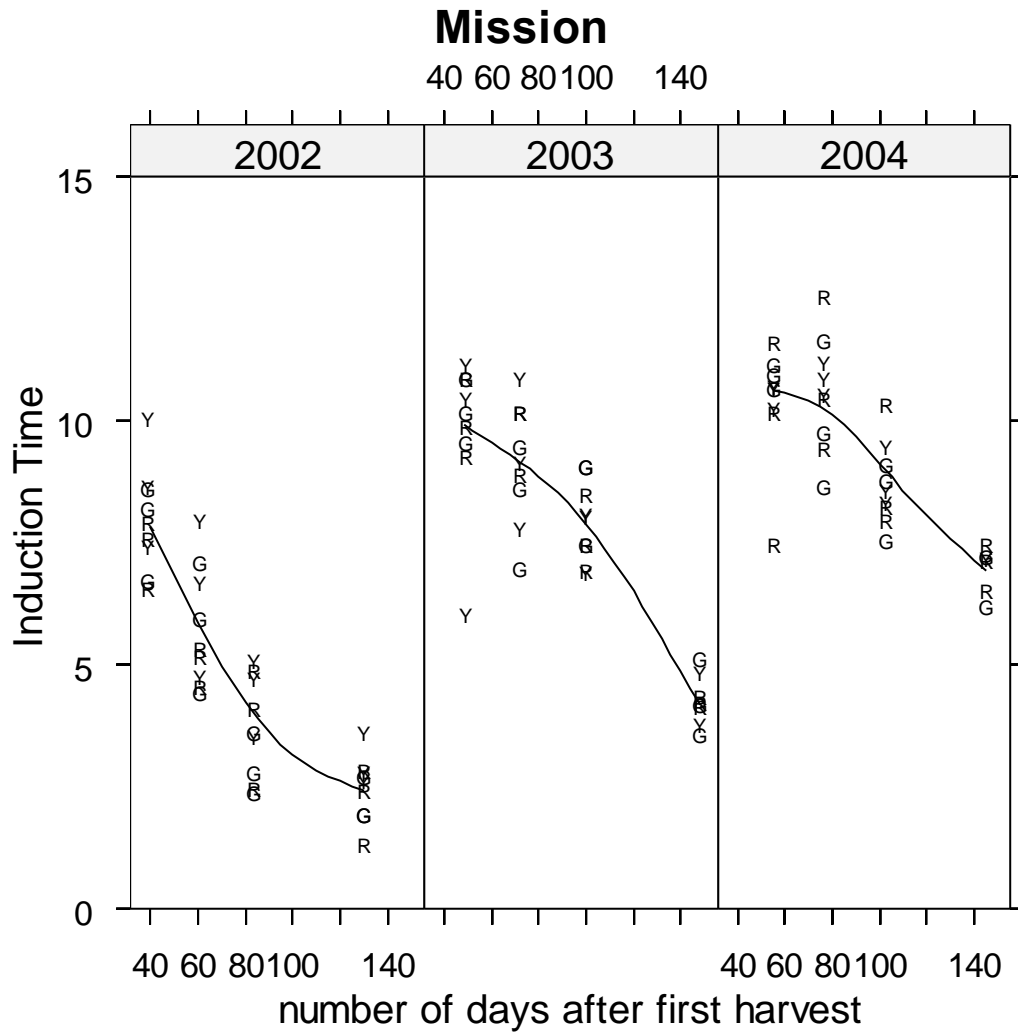


Fig. 4.33 Induction time of Mission olive oil at four harvest dates over three years including three irrigation treatments. G = full ; Y = moderate; R = deficit irrigation. There was a significant effect between years ($p = 0.002$) and no irrigation effect ($p = 0.462$). There was also an effect for harvest timing ($p = 0.043$).

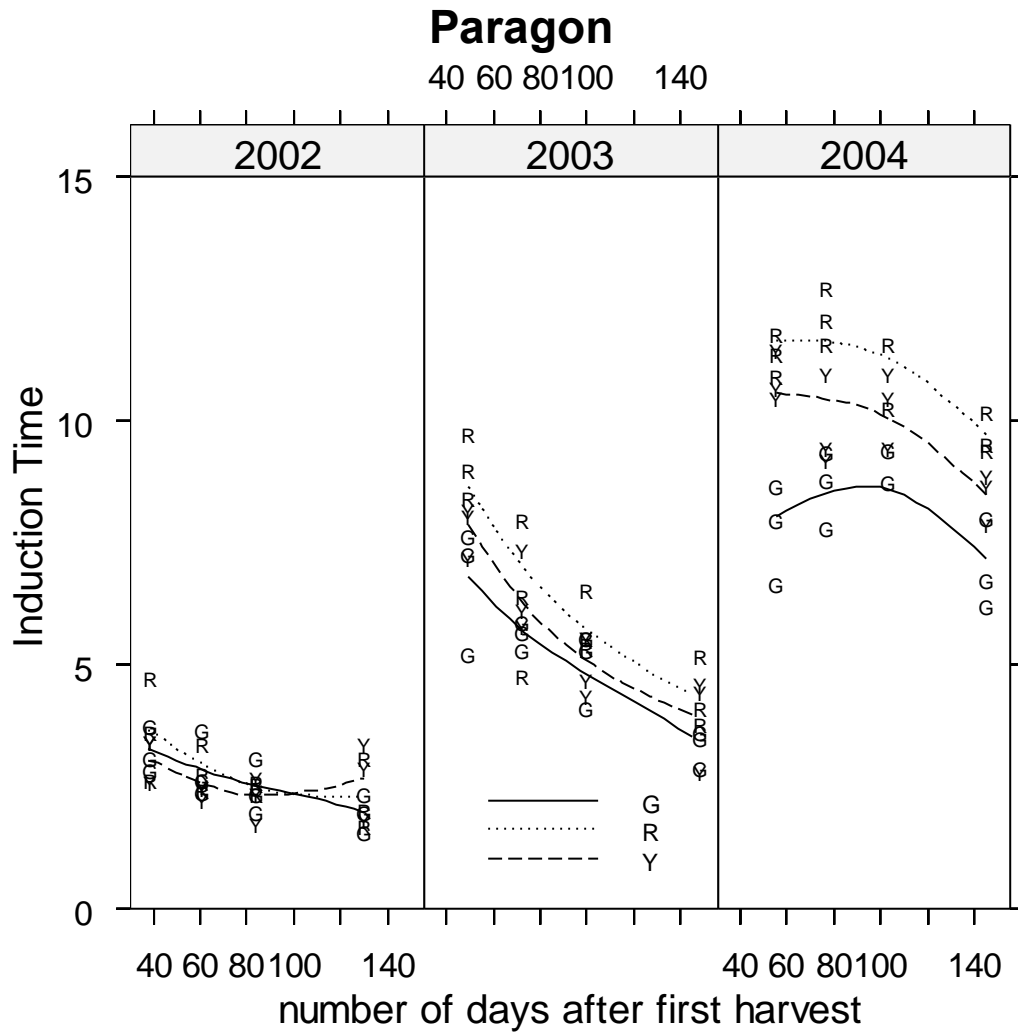
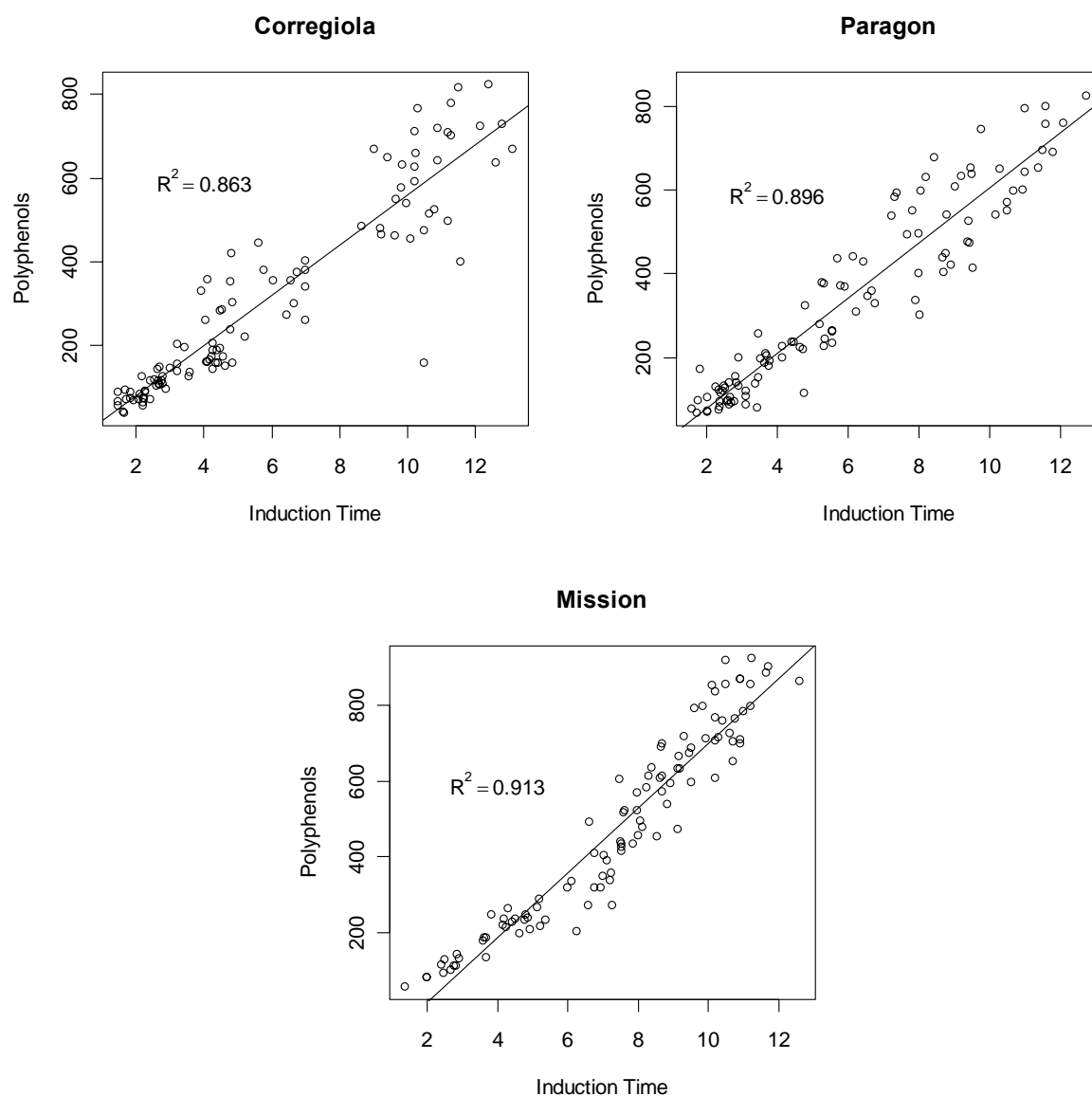


Fig. 4.34 Induction time of Paragon olive oil at four harvest dates over three years including three irrigation treatments. G = full ; Y = moderate; R = deficit irrigation. There was a significant effect between years ($p < 0.001$) and an irrigation effect ($p < 0.001$). There was also an effect for harvest timing ($p < 0.001$).

It can be seen by comparing induction time in (Fig 4.32, 4.33 and 4.34) in all cases, the induction time gets shorter with fruit maturity. Paragon had an unusual effect in 2004 with low induction time initially then increasing and eventually shortening again. This has identical to what has been observed with the polyphenol content which also increased in the middle of maturity unlike the other cultivars which showed continual declines. As for polyphenol concentration, the high irrigation treatment (G) in particular started with lowest induction and polyphenols which gives support to the theory that more water produces lower polyphenols and less oil stability.

Fig. 4.35 Relationship between induction time and polyphenol content in three cultivars of olives.



The increased induction time with the age of the trees is also important and also relates to increased polyphenols with tree age. As previously suggested, this could be presumed to be the result of better tree and root establishment with better access to soil nutrients.

Other oil characteristics which may contribute to increased induction time will include the ratio of saturated to polyunsaturated fatty acids, the level of other antioxidants such as the tocopherols, the amount of peroxide already present in the oil and the chlorophyll concentration. However, the linear regressions observed here suggest that polyphenol content is by far the greatest antioxidant influence. It also indicates that total polyphenols provide a good picture of the oil stability without the need for expensive and time consuming analysis of individual polyphenol compounds.

This relationship between polyphenols and induction time has been previously discussed (Mailer *et al.*, 2002). When induction time of all cultivars was graphed against polyphenol content there was a strong relationship, indicating the role that polyphenols play in oil stability. The relationship was even stronger when individual cultivars were analysed separately (Fig 4.35). This result would suggest that the relationship of induction to polyphenols content is different for each cultivar. It is most likely that different polyphenol profiles in each cultivar have different antioxidant effects. This has not been studied here but is of great interest to determine which polyphenol compounds provide the greatest level of stability.

Induction time was also found to relate to many other components, particularly the individual fatty acids. (Fig 8.1 - 8.2). It is also apparent that these parameters also relate to polyphenol content. These findings indicate that there is considerable interrelationship between olive oil components which are so far unknown. This study has identified some of these factors which will lead to further research.

4.7.3 Fatty Acid Profile

Fatty acid analysis at various stages of maturity indicated that individual fatty acids vary considerably in proportion during fruit development. This is of significant importance to olive oil producers in selecting oil with good stability and a superior nutritional fatty acid profile. The fatty acid profile is an important factor in consideration of oil quality. Olive oil is considered to be highly nutritional oil due in part to the high level of monounsaturated oleic acid. Australian oils however vary considerably in their fatty acid profile and sometimes do not meet the expectations of high oleic oil.

Palmitic acid: Palmitic acid (C16:0) is a saturated fatty acid which is commonly abundant in animal fats but also present in olive oil. As can be seen by Table 4.5, palmitic acid decreases in proportion to the other fatty acids over time from around 16% to more acceptable levels of 12%. Even at these levels, olive oil contains relatively high levels of saturated fat compared to seed oils. This is important to consider in future research and possible selection of cultivars of olives with a lower level of saturated fat. Although the levels of palmitic acid were similar between cultivars, Mission consistently had a slightly lower level than Paragon and Corregiola. The highest level of palmitic acid was in Paragon at 17.2% on 17/02/03. Mission was the lowest at 10.2% on 12/7/04. Palmitic acid had significant year effect ($p < 0.001$) for all cultivars. There were also significant effects for harvest timing ($p < 0.001$) in all cultivars but no effect for irrigation.

Oleic acid: Oleic acid (C18:1) was relatively consistent across the series of harvest times particularly with Mission in which there was virtually no change from early to late maturity. The maximum concentration was in Corregiola on 13/4/2004 at 74.8%. The minimum level was in Corregiola at 59.9% on 23/5/04. Corregiola and Paragon had significant year effects ($p < 0.001$) but there were no effects for harvest timing or irrigation.

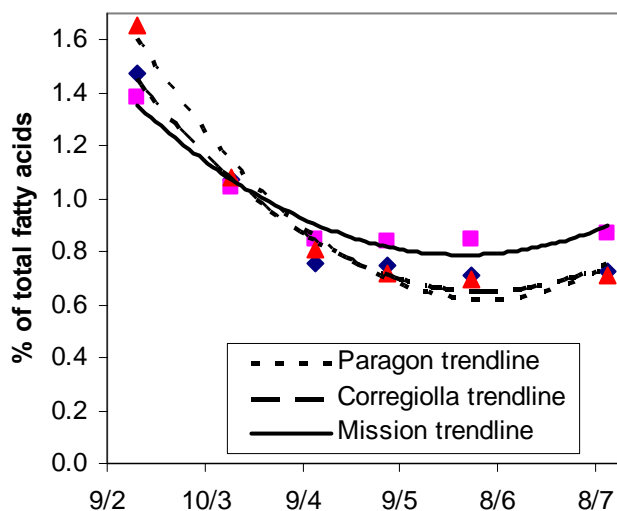
Linoleic acid: Linoleic acid (C18:2) increased as palmitic acid decreased with the oil becoming more polyunsaturated with time. Corregiola reached a maximum of 19.5% in 23/5/02 and also had the minimum level of 6.0% in 18/2/04. All cultivars increased in linoleic acid content. The rate of accumulation was similar until late April when Corregiola and Paragon slowed in comparison to Mission. Linoleic acid contributes toward oil instability due to the unstable polyunsaturated fatty acid. Again, Corregiola and Paragon had significant year effects ($p < 0.001$) but Mission didn't. Corregiola and Mission had significant time of harvest effects ($p < 0.001$) but there were no irrigation effects.

Table 4.5 Average palmitic, oleic and linoleic acid content of three olive cultivars over three years.

Harvest Date	Palmitic acid (C16:0)			Oleic acid (C18:1)			Linoleic acid (C18:2)		
	Coregiola	Mission	Paragon	Coregiola	Mission	Paragon	Coregiola	Mission	Paragon
18/2	15.24	15.70	15.48	72.08	71.11	71.01	7.46	7.09	7.76
18/3	15.10	14.94	15.23	69.82	69.16	69.69	10.11	10.31	9.95
13/4	14.18	13.47	14.75	70.19	70.72	69.46	10.81	10.50	10.86
5/5	14.11	12.56	14.26	68.37	70.37	68.64	12.54	12.09	12.02
31/5	13.36	11.69	13.74	68.68	70.01	68.63	12.82	13.21	12.36
12/7	12.80	11.06	13.00	68.58	69.76	68.56	13.53	14.20	13.08

Linolenic acid (C18:3): Olive oil has to conform to international standards and Australian oil sometimes is outside of those requirements. In particular, linolenic acid sometimes exceeds the maximum standard of 1.0%. This can occur more often in immature oil as the linolenic acid level decreases as the fruit matures. Linolenic acid was found to be very high early in the season but to decrease to acceptable levels (IOOC Standard <1.0%) by the beginning of April. All cultivars showed the same trend with Mission having a slightly higher level than the other cultivars. All three cultivars showed a tendency to increase slightly from mid June onwards. Paragon produced the highest concentration of linolenic acid in 18/2/04 at 2.2% and also the lowest on 15/2/03 at 0.5%. Shelf life and oil stability are closely related to the degree of saturation and polyunsaturation of oils. The traditional fatty acid profile of olive oil produces a relatively stable product. Palmitic acid is a saturated fat and as such provides stability. Oleic acid is monounsaturated and therefore contributes to that stability. However, linoleic and linolenic acids, despite their perceived nutritional benefits, are both susceptible to oxidation as they are polyunsaturated.

Fig. 4.36 Trendline of linolenic acid for three cultivars of olives. Each point represents the mean of three replicates over three years.



Analysis of variance of fatty acid profile (%) for the varieties Mission, Corregiolla, and Paragon are provided in Table 8.1, 8.2 and 8.3. Generally there were some year effects for linolenic acid but no irrigation effects. Time of harvesting was important for Paragon and Mission but not for Corregiolla.

4.7.4 Free fatty acids (FFA)

Free fatty acids were generally very low in immature fruit. The levels remained low until after June and the fruit had reached a high maturity index. At that stage, Paragon, showed a rapid increase in free fatty acids as the fruit aged. In 2002 and 2003, the FFAs in Paragon had exceeded the IOOC standard of 0.8% while the fruit were still on the tree. Corregiola had also increased in FFA toward late maturity but not to the same extent as Paragon. Mission remained low throughout the harvest period. The maximum level of FFA was in Paragon at 1.08% in 8/7/02. Mission had the lowest FFA in 12/7/04 at 0.10%.

Fig. 4.37 Trendline of free fatty acids for three cultivars of olives. Each point represents the mean of three replicates over three years.

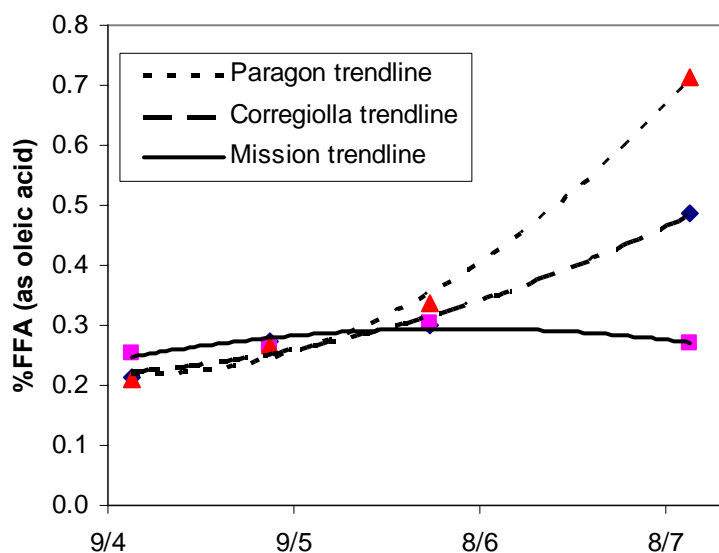


Fig. 4.38 Free fatty acid (FFA) content of three cultivars of olives over three years. Graphs are the average of three replicates in each year for each cultivar.

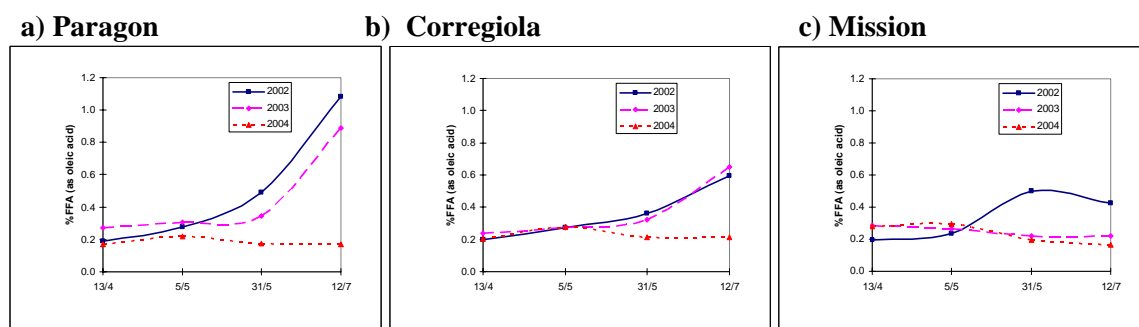


Fig. 4.38 shows individual year effects for FFAs for each cultivar. Although there were differences for the three years, Paragon consistently had the highest level of FFA.

The factors that contribute to increase in FFAs are those that bring the oil triacylglycerols into contact with endogenous lipase enzymes which can break the oil molecules down. For example, as the fruit ages, free fatty acids increase with the rupture of cell walls. Other contributors appear to be from reduction in moisture content to a level where desiccation caused cell breakdown and an increase in free fatty acids. The latter scenario appears to have been responsible for high free fatty acids at the final harvest in Paragon and possibly Corregiola in 2002 and 2003 which had very low moisture levels in those two years (Fig. 4.9 and 4.11).

4.7.5 Peroxide value

Peroxide values were included in the analysis for the years 2003 and 2004 although little change was expected. Peroxides form with oxidation, after the oil is extracted and exposed to air and therefore should not change in the fruit. Mission was consistently the lowest in peroxide and Paragon the highest, possibly due to desiccation as discussed for FFAs. Peroxide value was shown to be higher in young olives than later in the season (Table 4.6) although this was not understood.

Peroxide value was influenced by years ($p = 0.010$ to < 0.001) but not by irrigation or time of harvest.

Table 4.6 Peroxide value at four harvest dates for three cultivars of olives, average over two years.

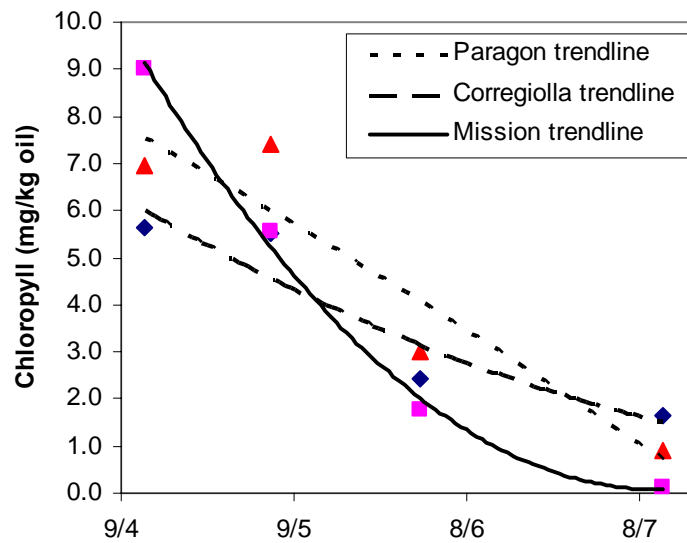
Harvest Date	Days after Jan 1	Free Fatty Acid (%)			Peroxide Value Average of two years – 2003/04		
		Corregiola	Mission	Paragon	Corregiola	Mission	Paragon
13/4	104	0.21	0.25	0.21	8	8	10
5/5	126	0.27	0.27	0.27	-	-	-
31/5	152	0.30	0.30	0.34	8	7	9
12/7	194	0.49	0.27	0.71	8	6	10

4.7.6 Chlorophyll

Chlorophyll content, as expected, was high in the immature olives and rapidly decreased with time as the colour changed from green to black. Mission started with the highest level of chlorophyll overall but ended with the lowest level (Fig. 4.39). Paragon had the highest level of chlorophyll in 5/5/2004 with 21.5 mg/kg whereas Mission was the lowest in 12/7/04 with 0 mg/kg measured. All cultivars showed a temporary increase in chlorophyll during ripening in 2004. This was unexplained but again the overall trend for the three years was a gradual decline in chlorophyll as the fruit matured. The year, irrigation and time of harvest effects for chlorophyll were low (Table 8.1, 8.2 and 8.3).

Chlorophyll has been shown in previous studies to contribute to oil stability whilst kept in the dark, or in intact fruit (Boskou 1996). However, chlorophyll captures light and will become a pro-oxidant after the oil is extracted. Chlorophyll is seen as a positive characteristic for olive oil as it provides the green colour to the oil. However, to take advantage of the aesthetic value, it is necessary to store the bottle in clear glass but this would therefore contribute to oxidation and reduced shelf life of the oil. The green colour is generally a sign that the oil is relatively fresh as oil stored in the light will gradually lose the green colour and turn golden yellow.

Fig. 4.39 Trendline of chlorophyll for three cultivars of olives at four harvest dates. Each point represents the mean of three replicates over three years.

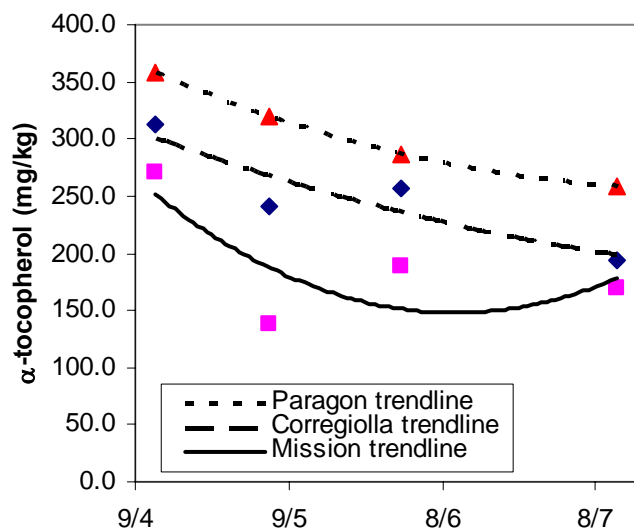


4.7.7 α -tocopherol

α -tocopherol or Vitamin E, is a strong antioxidant. This component was only measured in 2004 when the method had been established as it was not included in the original proposal. All cultivars showed a trend to reduction in tocopherol content as the fruit matured with only Mission showing a slight increase again toward the end of ripening. The order of α -tocopherols content for each cultivar was the inverse of polyphenols with Mission being the lowest concentration and Paragon the highest.

Paragon reached the highest level with 479 mg/kg in 13/4/04 and Mission was the lowest with 138 mg/kg in 5/5/04. Tocopherol showed a time of harvest effect but no irrigation effect.

Fig. 4.40 Trendline of α -tocopherols content for three cultivars of olives at four harvest dates. Each point represents the mean of three replicates for one year.



5. Implications

The major variation in oil quality and oil yield between the cultivars studied is due to fruit maturity. The grower may therefore decide to harvest to produce green oils with pungent sensory characteristics and with increased shelf life stability when antioxidants are high, or alternatively, to produce oil which is golden yellow with milder flavours. The nutritional quality will also be influenced by decisions of when to harvest.

The aim of the project was to relate physical and easy to measure parameters with oil quality and oil recovery. The tests traditionally used to identify quality in the field include fruit colour, weight, detachment force and hardness. Maturity index is the most common European method to determine fruit condition but was of little value in this study to identify harvest times. We have shown that fruit change at different rates within each cultivar, and even within individual trees and reliance on this character under Australian conditions would lead to late harvesting and loss of fruit and oil quality.

The study included irrigation effects although the environmental conditions over three years were less than ideal with drought persisting over the period. Water was limiting and there were few significant differences between high irrigation, moderate irrigation and severe deficit irrigation. The study however did provide considerable information on soil water movements and the effects on some of the numerous analyses carried out.

Fruit growth was found to follow the same pattern as oil accumulation. Although fresh fruit weight in olives was very variable, increasing and decreasing with rainfall or irrigation events, a more useful tool was the dry weight. Fruit dry weight produced similar growth curves and levelled out as fruit weight reached its maximum. The limitations are that fruit weight continues to increase after optimum oil content has been reached. Reliance totally on this method would result in late harvests with reduced fruit yield. Fruit firmness dropped rapidly at point when oil content had reached a maximum. A combination of dry weight and fruit hardness has potential for indications of harvest timing.

Chemical tests are expensive and take time but are necessary to relate physical testing to oil quality. Moisture tends to decrease over the growing period but Mission was unique in that it maintained constant moisture throughout the growing period. Paragon and Corregiola also varied showing this is not likely to be a reliable indicator. Cold-press extraction is the best indication of when to harvest as this imitates the oil extraction process in commercial situations. It was found that cold-press extraction reached a plateau well before the olive had ceased producing oil. This is because other factors are changing in the olive which makes the oil harder to extract. Solvent extraction of the oil is useful to compare the efficiency of cold-pressed systems, however, it does not indicate what the cold-press extraction will yield.

A major contributor to oil quality is the polyphenol content which plays a role in determining oil stability and organoleptic qualities of pungency and bitterness. The high polyphenols shown to be present in young olives rapidly decrease as the fruit matures. The oil changes from strong pungent oil to a mellower product. Polyphenols are closely related to many of the other oil components and most obviously to induction time. The close relationship of polyphenols to induction time, shelf life stability and many of the other oil components has been illustrated.

The fatty acid profile is important for oil stability but also in the oils nutritional quality. Although oleic acid changed little over the maturity period, linoleic acid (polyunsaturated) was found to increase while palmitic acid (saturated) decreased. Harvest timing can be used to select for stability or nutritive value. Linolenic acid also decreased with maturity. This component is unstable and can increase the rate of oxidation. The linolenic acid needs to be below 1.0 % which is usually achieved early in the maturity process.

Free fatty acids must be less than 0.8 % in extra virgin olive oil. It is necessary to harvest olives, particular cultivars such as Paragon, before free fatty acids increase. Chlorophyll provides green colour to the oil which has some benefits but becomes a pro-oxidant contributing to oxidation. Early harvesting can achieve higher levels of chlorophyll and tocopherol.

6. Recommendations

Tree age: This report has highlighted the changes which occur during fruit maturation. The trees utilised were very young at only four to seven years of age. Although this is typical of many new groves around Australia, it will not reflect the olive groves of the future. There are several theories about the advantages of old trees over young ones, such as a perceived improvement in oil sensory characteristics. Little however is known if there is any advantage.

The quality of the oil obtained from the trees in this project showed dramatic differences over the three years. Polyphenol content increased from less than 200 mg/kg to greater than 600 mg/kg over the three years. In relation to polyphenols, which contribute to pungency and bitterness in the oil, all of the samples tested showed progressive changes in oil sensory characteristics. Oil shelf life stability also increased over the three years. The changes in three years may be related to three years of progressively worsening drought or to better establishment of root growth and tree uniformity. It is difficult to confirm that this is due to tree maturity over three years and the study should be continued for several years into the future to establish the influence of tree age to oil quality.

Further research of this grove would be beneficial to provide evidence of changes in oil quality. However, if the orchard is not available, alternative groves of similar age may be used to record changes in oil characteristics over several years.

Orchard Location: This study was limited to a single orchard in Southern New South Wales. It has been shown through a study of fatty acid profiles undertaken by this laboratory for Codex International, that fatty acid profiles show significant variation between northern regions of Australia and the south. Oleic acid is consistently higher in Victoria than Queensland and palmitic acid is lower. Other factors not measured may also be variable but is currently unknown.

An expanded study to take in groves in different environments is essential to determine the results of oil produced under all conditions.

Cultivars: Only three cultivars were included in this study. It is estimated that 90% of Australian olive trees consist of ten main cultivars. Chemical analysis of oil from the range of these trees across the Australian growing region, has shown considerable variation in limited tests. Barnea for example, an Israeli bred cultivar, produces a sterol profile quite different from other Australian types.

Additional cultivars should be included in future research to provide the same information to all growers.

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8. Appendices

Analysis of variance for the varieties Mission, Corregiola, and Paragon. * The denominator degrees of freedom used in the calculation of the P-value was determined using Kenward adjustments (as implemented in ASReml).

Table 8.1: Table of P-Values from the analysis of the variety Paragon.

Term	S. Oil	Moisture	Polyphenol	Induction	C18.1	C18.2	C18.3	C16.0	Chloro	%FFA	Perox	Toco	Cp. Oil	Av. Fresh wt	Mature
Year	<0.001	0.452	<0.001	<0.001	<0.001	<0.001	0.014	<0.001	0.033	<0.001	0.001	NA	0.012	<0.001	0.163
Irr	0.589	0.477	<0.001	<0.001	0.753	0.529	0.592	0.755	0.057	0.296	0.002	0.298	0.229	<0.001	0.622
Year:Irr	0.117	0.680	0.001	<0.001	0.776	0.900	0.235	0.246	0.120	0.122	0.176	NA	0.127	0.003	0.162
Lin(Time)	0.039	0.026	<0.001	<0.001	0.123	0.011	<0.001	<0.001	0.019	<0.001	0.233	<0.001	0.032	0.027	0.015
Year:Lin(Time)	0.060	0.012	<0.001	<0.001	0.017	<0.001	0.007	0.188	0.152	0.002	0.300	NA	0.734	0.178	0.087
Irr:Lin(Time)	0.209	0.799	0.117	0.309	0.576	0.528	0.256	0.730	0.095	0.006	0.830	0.474	0.904	0.997	0.811
Year:Irr:Lin(Time)	0.614	0.956	0.470	0.244	0.055	0.229	0.082	0.423	0.073	0.056	0.282	NA	0.819	0.851	0.576

Table 8.2: Table of P-Values from the analysis of the variety Corregiola.

Term	S. Oil	Moisture	Polyphenol	Induction	C18.1	C18.2	C18.3	C16.0	Chloro	%FFA	Perox	Toco	Cp. Oil	Av. Fresh wt	Mature
Year	0.016	0.053	<0.001	<0.001	<0.001	<0.001	0.228	<0.001	0.003	0.050	0.010	NA	0.687	<0.001	0.006
Irr	0.075	0.986	0.184	0.417	0.292	0.578	0.467	0.077	0.633	0.091	0.193	0.479	0.537	0.060	0.820
Year:Irr	0.535	0.580	0.839	0.969	0.169	0.263	0.392	0.158	0.897	0.887	0.046	NA	0.940	0.584	0.495
Lin(Time)	<0.001	<0.001	<0.001	0.002	0.283	<0.001	0.087	<0.001	0.016	<0.001	0.358	<0.001	0.075	0.109	<0.001
Year:Lin(Time)	0.253	0.033	0.014	0.068	<0.001	<0.001	0.009	0.026	0.057	0.006	0.141	NA	0.607	0.014	0.169
Irr:Lin(Time)	0.996	0.880	0.533	0.633	0.579	0.643	0.230	0.687	0.764	0.077	0.350	0.362	0.421	0.781	0.948
Year:Irr:Lin(Time)	0.033	0.907	0.535	0.518	0.075	0.368	0.022	0.034	0.713	0.635	0.727	NA	0.853	0.936	0.739

Table 8.3: Table of P-Values from the analysis of the variety Mission.

Term	S. Oil	Moisture	Polyphenol	Induction	C18.1	C18.2	C18.3	C16.0	Chloro	%FFA	Perox	Toco	Cp. Oil	Av. Fresh wt	Mature
Year	0.008	<0.001	0.009	0.002	0.033	0.024	0.019	<0.001	0.322	0.044	<0.001	NA	0.010	<0.001	0.401
Irr	0.644	0.470	0.787	0.462	0.042	0.214	0.890	0.056	0.262	0.283	0.149	0.331	0.158	0.283	0.199
Year:Irr	0.601	0.868	0.508	0.506	0.476	0.596	0.797	0.203	0.284	0.372	0.276	NA	0.987	0.441	0.191
Lin(Time)	<0.001	0.019	0.010	0.043	0.599	<0.001	<0.001	<0.001	0.003	0.092	0.018	0.002	0.586	<0.001	0.005
Year:Lin(Time)	0.110	0.008	0.651	0.417	0.008	0.052	0.008	<0.001	0.519	0.002	0.075	NA	0.389	0.169	0.710
Irr:Lin(Time)	0.204	0.126	0.211	0.752	0.761	0.645	0.131	0.746	0.275	0.156	0.389	0.414	0.731	0.882	0.705
Year:Irr:Lin(Time)	0.397	0.844	0.614	0.799	0.210	0.295	0.432	0.170	0.543	0.347	0.969	NA	0.709	0.008	0.077

Corregiola

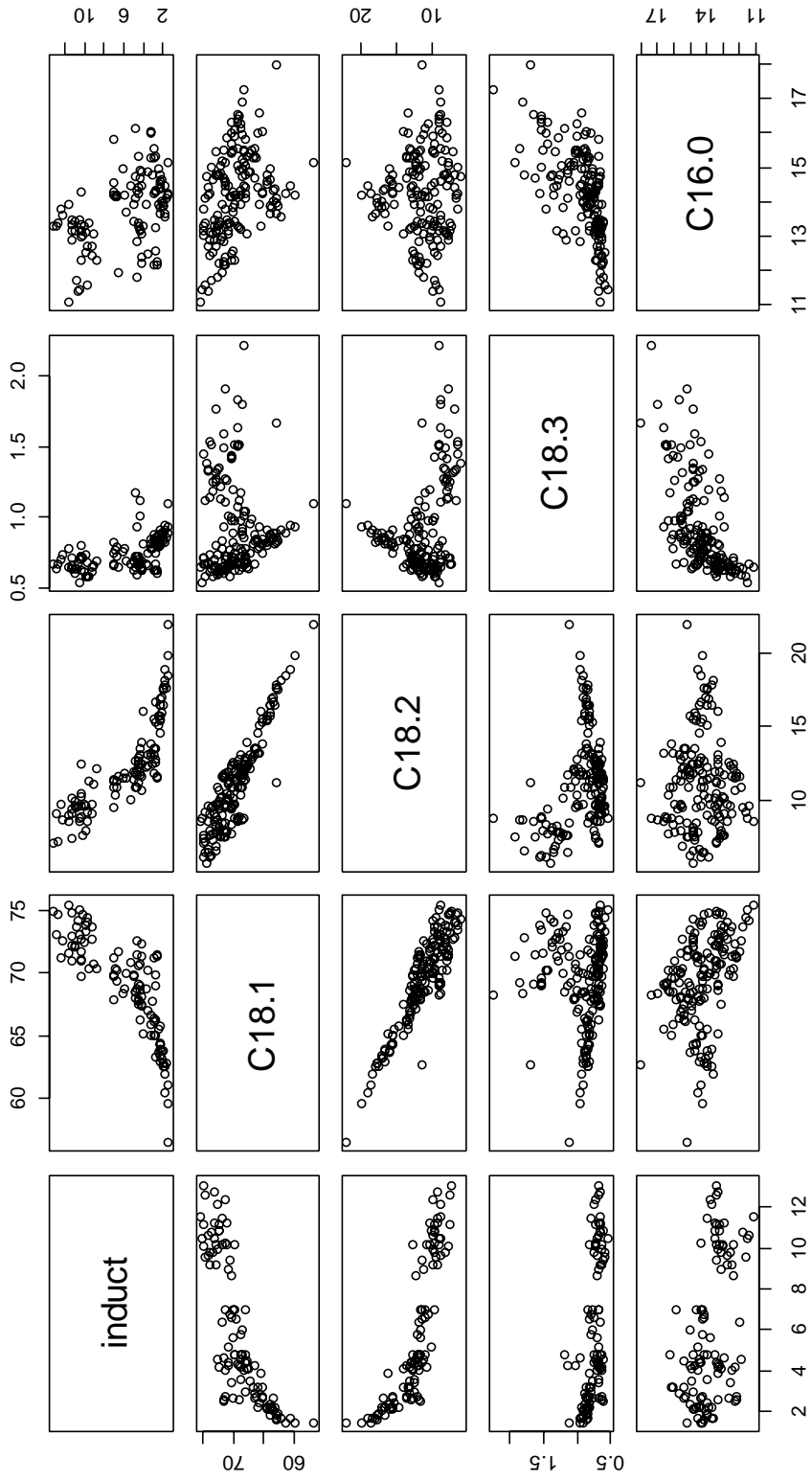


Fig. 8.1. Relationship between induction time and fatty acids C16:0, C18:1, C18:2 and C18:3.

Corregiola

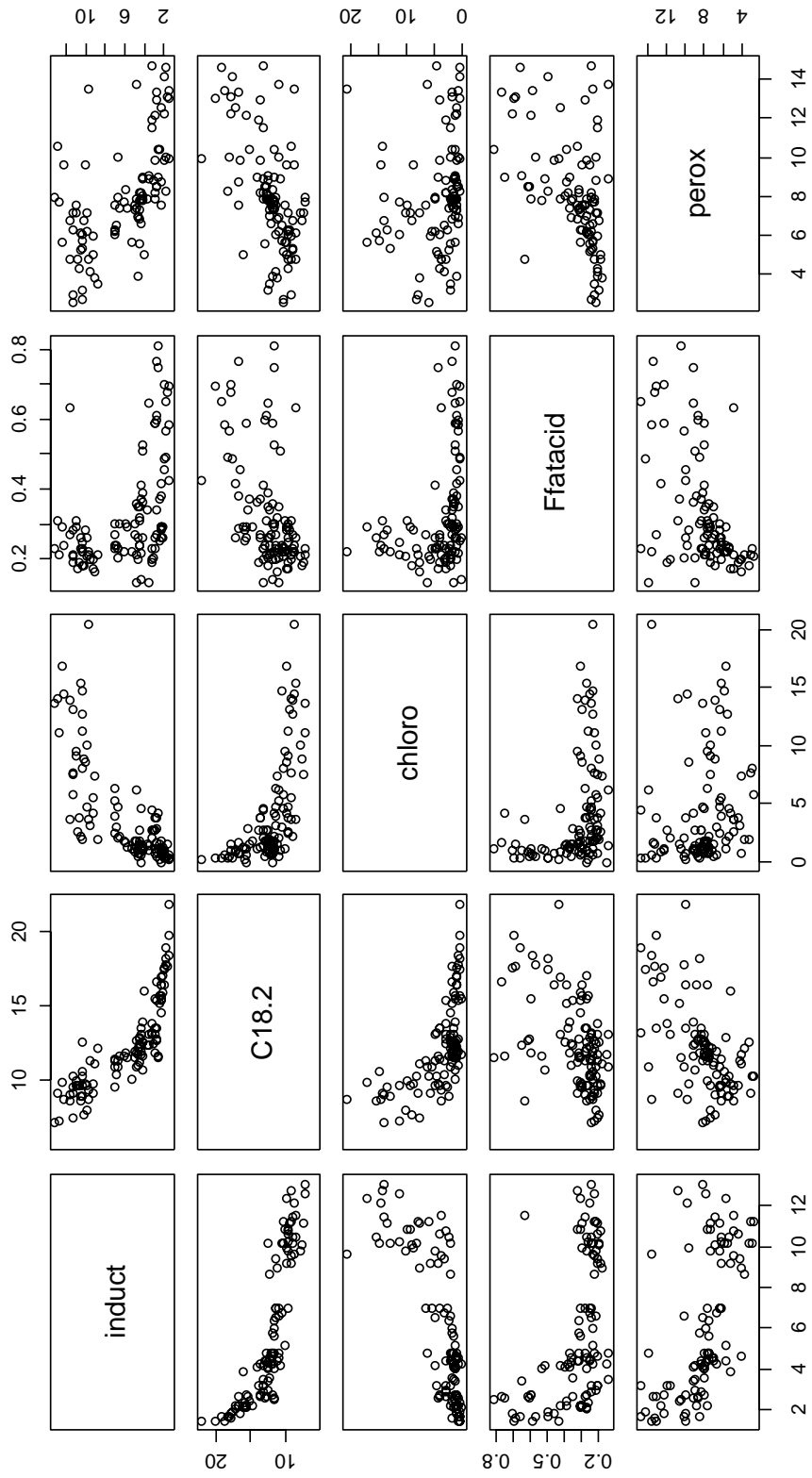


Fig. 8.2. Relationship between induction time and fatty acid C18:2, chlorophyll, free fatty acids and peroxide value.