Delayed Fruit Harvest on Yield, Nutritional Value and Post-harvest Quality of Late Maturing Reed Avocado (*Persea americana* Mill)

by

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DECLARATION

I declare that the dissertation submitted to the University of Limpopo, for the degree Master of Agricultural Management (Plant Production) has not previously been submitted by me for a degree at this or any other university; it is my work in design and in execution, and that all materials contained herein had been duly acknowledged.

Novela P. **Date**

DEDICATION

I would like to dedicate this dissertation to my mother, Mrs Salphinah Novela and my late father, Mr Famanda Willy Novela. Thank you for moulding me into a woman I am today.

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ABSTRACT

Delayed fruit harvest (DFH) or on-tree-fruit storage is a strategy being considered to extend the harvest season of late maturing ‘Reed’ avocado. However, avocado fruit growth beyond physiological maturity is accompanied by alterations in chemical and physical properties. Thus, the study aimed to investigate the effects of delayed harvest on tree productivity, mineral nutrition and post-harvest quality of late maturing ‘Reed’ avocado and further to evaluate the fruit quality response to varying ripening temperature regimes. As treatments, fruit were harvested early, mid and late in the season, with one month apart from each harvest during 2013 and 2014 seasons. Yield and fruit size were recorded at harvest. During each harvest time, two sets of fruit samples were collected to determine the fruit nutritional content while the second set was cold stored at 5.5°C for 28 days. After storage, fruit were ripened at 16, 21 or 25°C and evaluated for post-harvest disorders, pathological diseases and fruit physico-chemical properties. Yield dropped from 42 to 12 kg/tree from early to late harvest during 2013, while treatments showed no effect during 2014. Furthermore, treatments had no effect on fruit size during both seasons. Phosphorus content decreased by 40 and 23% from early to late harvest time during 2013 and 2014 seasons, respectively. Similar decreasing patterns were observed for K, Ca and Mg content. Zinc content remained constant during 2013; however, mid-season fruit maintained higher zinc levels during 2014 season when compared to early and late harvest time. Protein content was constant for early and mid-season fruit but significantly decreased during the late harvest in both seasons. Moisture content decreased from 74.0 to 65.0% from early to late harvest during both seasons. Starch, oil and ash content were high during late harvest time in both seasons; while crude fibre was high during the mid-harvest time when compared with early and late harvest time. Internal chilling injury increased with maturity and ripening temperature. In both seasons, higher temperatures induced higher anthracnose incidences during early and mid-harvest compared with late harvest time. Similarly, vascular browning was high on fruit ripened at 16°C during early and mid-harvest time but decreased on late harvest fruit during 2013. However, low ripening temperatures induced high incidences of vascular browning during 2014 season. Stem-end rot was high on fruit ripened at 16°C than 21 and 25°C during 2014 season.

Physico-chemical quality parameters showed similar behaviour in both seasons. Fruit lightness increased with ripening time regardless of ripening temperature during the early harvest, but remained constant during mid and late harvest. Hue angle (h^0) and Chroma (C*) values were slightly reduced during ripening. Fruit ripened at 25°C had the highest respiration rate and reached a climacteric peak earlier (day 2) than fruit ripened at 21 and 16°C (day 4). Fruit firmness and weight loss were high and rapid at 25°C followed by 21 and 16°C regardless of the harvest time. Delayed harvest had no effect on yield and fruit size. Generally, nutritional content of avocado fruit increased with fruit maturity. Furthermore, fruit maturity played a major role in the response of fruit to ripening temperature. Higher temperature enhanced fruit ripening, but was conducive for development of post-harvest diseases.

Keywords*: Harvesting time, nutrition, yield, fruit size, ripening temperature, post-harvest quality*

CHAPTER 1
GENERAL INTRODUCTION

1.1 Background

1.1.1 Description of research problem

The South African avocado season extends from February to October with no supply in November to January (FPEF, 2015) (Figure 1.1). Therefore, the industry is currently not self-sufficient to supply fruit outside this traditional production window. Consequently, avocado fruit has to be imported to meet the local demand during the off-season (Lemmer *et al*., 2006). Characteristically, the South African avocado market price show a distinct seasonal variation, with the lowest price in autumn and winter, rising through spring and peaking in summer. This trend exhibits a demand and supply function during these times (Graham and Wolstenholme, 1991). Thus, the low availability of the fruit presents a very distinct price advantage available to the producer who markets fruit from November to January. The industry has since adopted (1) breeding and selecting for early and late maturing cultivars and (2) delaying harvest of some late cultivars such as 'Reed' as two strategies to address this identified avocado fruit availability gap.

Harvest month

Cultivar	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
'Fuerte'												
'Hass'												
'Pinkerton'												
'Ryan'												

Table 1.1 Seasonal spread of major avocado cultivars in South Africa (adopted from FPEF, 2015).

1.1.2 The impact of research problem

The subtropical fruit industry is the sixth largest contributor to the gross value of horticultural products (Scheepers *et al.,* 2007). The South African avocado industry (SAAI), as the main contributor to the gross value of production of subtropical crops, has progressively grown in recent years, and currently, ranked fourth in the world in terms of world export trade (NAMAC, 2014). Currently, the domestic market occupies 51.4% of the total avocado production volume, whereby 48.6% of the total avocado production volume is exported to the European market (DAFF, 2014). In 2013, SAAI constituted an international market share of 3.3% (DAFF, 2014). Most of the fruit is exported during the first export season with little contribution from the industry late in the season. This gives an advantage to other exporting countries such as Peru and Spain, which export most of their crop during the South African off-season.

1.1.3 Possible cause/s of research problem

Different cultivars are cultivated in different climatic regions, but early season fruit usually dominate the market, followed by mid-season cultivars while tail end of the season is left with very little or no supply of the fruit (DAAF, 2014; FPEF, 2015). According to the South African Avocado Growers Association (SAAGA, 2007) and FPEF (2015), 'Fuerte' fruit usually dominates the market early in the season (February-June), followed by mid-season 'Pinkerton' fruit, while 'Hass' (March-September) and 'Ryan' (August-October) serve as a filler from early to tail end of the season. Due to the export orientation nature of the industry, the growers and research workers have focused mainly on European preferred cultivars ('Hass' and 'Fuerte') (SAAGA, 2007). This has resulted in negligence of late maturing cultivars such as 'Ryan', 'Reed' and 'Gwen' which could be used to fill the seasonal gaps in the local market.

1.1.4 Proposed solution/s

The inability of mature avocado fruit to ripen while attached to the tree (Graham and Wolstenholme, 1991) could be utilized as an on-tree-fruit storage strategy to fill the production gap late on the season. However, Kassim *et al*. (2013) showed that late fruit growth beyond physiological maturity was accompanied by alterations in certain physico-chemical properties of the fruit. Generally, it is important to understand these physico-chemical changes, especially late in the growing season to ensure that fruit are harvested at their optimal maturity and quality (Ferguson and Boyd, 2002). On-tree-storage or delayed harvest strategy was extensively studied only on 'Hass' and

‘Fuerte’ as major cultivars. However, this strategy still needs to be explored on minor late cultivars such as ‘Reed’.

1.2 Problem statement

The physiological effects of delayed fruit harvest on the nutritional value and yield of late maturing ‘Reed’ avocado has not yet been determined, although the practice has been studied on ‘Hass’ and ‘Fuerte’ (Graham and Wolstenholme, 1991; Whiley *et al*., 1996). Furthermore, the impact of increasing fruit maturity in relation to ripening temperature and ripening physiology of the fruit is not well defined for South African late season avocado fruit.

1.3 Motivation of the study

Empirical based information on developmental alterations during the advancing fruit maturity and their effects on fruit quality would enhance farm management decisions and post-harvest treatments commonly used in the avocado industry. This will also allow for the extension of the production season and reduce the need to import fruit from the Northern hemisphere production areas during the South African off-season.

1.4 Aim

The study aimed to investigate effects of delayed harvest of late maturing ‘Reed’ on tree productivity, nutritional value and post-harvest quality in response to different ripening temperature regimes.

1.5 Objectives of the study were to:

a) Determine the effects of delayed harvest on fruit nutritional content and tree productivity of late maturing ‘Reed’ avocado.
b) Investigate the influence of ripening temperature on post-harvest quality of avocado fruit harvested at different maturity stages.

1.6 Hypotheses

a) Delaying fruit harvest has no effect on nutritional content and productivity of late maturing 'Reed' avocado.

b) Delaying fruit harvest has no influence on post-harvest quality of fruit in response to ripening temperature.

1.7 Structure of the dissertation

The dissertation was designed using the Senate-approved technical format of the University of Limpopo. Findings were summarised in the abstract, followed by detailed background of the research problem (Chapter 1), which was in turn followed by a review of the relevant literature on the research problem (Chapter 2). Empirical studies comprised that of achieving two objectives *viz*. Objective 1 (Chapter 3) and Objective 2 (Chapter 4). Finally, findings were summarised, with related recommendations being provided regarding delayed fruit harvest impact in avocado production (Chapter 5).

CHAPTER 2
LITERATURE REVIEW

2.1 Introduction

Low fruit volumes produced during the summer months has been the challenge faced by the SAAI for decades (Graham and Wolstenholme, 1991). Currently, delaying fruit harvest is one of the industry's options to extend its production season, as avocado fruit does not ripen on the tree. This review on the research problem focused on three themes: (i) what has already been written on the research problem, (ii) existing gaps on the research problem and (iii) the explanation on how the gaps would be addressed.

2.2 Work done on the research problem

2.2.1 Avocado production in South Africa

In South Africa, avocado was introduced only in the 19th century. In the 1970s, the area planted with avocado was approximately 2 000 ha. However, this has expanded steadily over the past three decades to 15 388 ha in 2012 (DAFF, 2014). The production is concentrated mainly in the warm subtropical areas of the Limpopo (Tzaneen and Levubu), Mpumalanga (Nelspruit and Hazyview) and KwaZulu-Natal Provinces (De Villiers and Joubert, 2011). The commercial avocado orchards have predominantly been planted with 'Fuerte', 'Hass', 'Ryan' and 'Pinkerton' cultivars (DAFF, 2014). These different regions give the industry an ability to produce avocados from the beginning of February to the end of October (FPEF, 2015), with the bulk of the crop from the end of March until beginning of September. However, the supply gap from November to January has an influence on the industry's market share and also seasonal price variation on local markets. This has since been identified as an opportunity for South African farmers to lengthen their harvesting season in order to take advantage of relatively high summer prices.

2.2.2 Avocado nutritional value and its relation to fruit maturity

Avocados are becoming an increasingly important crop in South Africa and other countries of high economic value due to its excellent nutritional quality and easily eating characteristics by people with certain health problems (Loeillet, 1997). It is a high fat fruit, contains rare sugars of high carbon number (C-7) and is relatively rich in certain vitamins, dietary fibre, minerals and nitrogenous substances. Oil content is a key part of the sensory quality of the fruit. The quality of oil is similar to olive oil with a high proportion being approximately 71% monounsaturated, 13% saturated and 16% polyunsaturated fatty acids (Dreher and Davenport, 2013). The mono-unsaturated fats in avocados have been found to reduce blood cholesterol while preserving the level of high density lipoproteins (Dreher and Davenport, 2013). Furthermore, the edible portion of the fruit is rich in oleic, palmitic, linoleic and palmitoleic acids, while stearic acid is present only in trace amounts. However, changes in fatty acid distribution of the lipids were found to be associated with fruit development (Itoh *et al*., 1975).

Kassim *et al*. (2013) reported that fruit growth is accompanied by alterations in certain chemical properties of the fruit, and thus, some losses in mineral and nutritional components are likely to occur during late season fruit. According to Rosecrance *et al*. (2012) avocado fruit absorb nutrients according to the seasonal growth patterns, and matching cultural practices to those patterns could maximize yields, improve fruit quality and reduce the potential for nutrient loss. However, the pattern of mineral uptake is also dependent on the nature of the mineral and the transport pathway, with some minerals ceasing moving to the fruit late in the growing season (Heynes and Goh, 1980). Furthermore, Rosecrance *et al*. (2012) reported that ‘Hass’ avocado fruit accumulated N, P, and K in a double sigmoid curve while Ca accumulation follows a single sigmoid curve. The ash or mineral content and total sugars in avocados is relatively small as compared to the other constituents of the fruit, but it is noticeable higher than that recorded for other fresh fruits, with most varieties containing twice as much mineral matter as that contained in other fruits (Stahl, 1933). However, changes in these constituents during fruit maturity are too small.

2.2.3 Avocado fruit quality components and indices

Fruit quality is a prime concern for all involved in the avocado industry. Recently, quality has become one of the most important consumer decision factors in the selection among fresh produces (Khorshidi *et al*., 2010). It is essential for the industry to maintain a perfect quality of the produce in order to sustain the competition. According to Molnar *et al*. (1995), the quality of fresh produce in conformity with consumer requirements and acceptance is determined by their physical properties, sensory attributes, chemical composition, level of microbiological and toxicological contaminants, shelf-life and packaging. The major quality criteria used in the avocado industry include skin colour, fruit size, absence of wounds and cracks. When ripe, the key quality indices include absence of diseases, physical disorders and physical damage (Allan *et al.,* 2003). Many of these quality factors are cultivar dependent and consumer preference for size, shape and colour vary from one region to another. Furthermore, there are significant differences in size, texture and flavour between the different cultivars.

Colour is an important food quality parameter that affect consumer acceptance and could evoke emotional feelings in human (Crisosto *et al*., 2003; Bayarri *et al*., 2001). It is also used to indicate the stages of ripening for the avocado industry and consumers (Arzate-Vazquez *et al*., 2011). Generally, mature avocado fruits are harvested green although 'Hass' cultivars that are harvested late in the season may have skin darkening at harvest. 'Hass' avocado fruit changes colour during ripening from green to purple and eventually black when ripe (Cox *et al*., 2004; Arzate-Vazquez *et al*., 2011). However, green skin cultivars such as 'Reed', 'Fuerte' and 'Sharwil' do not darken with maturity nor ripening, therefore, objective colour measurements commonly using a chroma-meter or colorimeter are utilised to distinguish the various stages of ripening (Maftoonazad and Ramaswamy, 2008).

Fruit size is also an important component of yield with premium prices often paid for larger fruit (Cowan, 1997). In South Africa, fruit are weight sized into the following counts: 10 (346-462g), 12 (300-371g), 14 (258-313g), 16 (227-274g), 18 (203-243g), 20 (184-217g), 22 (165-196g), 24 (151-175g), 26 (144-157g), 28 (134-137g), 30 (123-137g), small (100-123g) (NDA, 2013). According to Graham and Wolstenholme (1991), fruit should be harvested at a more profitable count 16 and 14 for South African avocados.

The chemical quality parameters are closely related to both the physical and sensory appeal of avocado (Obenland *et al*., 2012). Some of the chemical parameters include pH, total titratable acid, moisture content, oil content, dry matter content and total soluble sugars. The degree of change of these parameters depends on the harvest time, maturity, cultivar and storage conditions (Maftoonazad and Ramaswamy, 2008).

2.2.4 Maturity and harvest indices

Generally, fruit maturity involves the comprehension of developmental stages of fruit growth from flowering to maturity and end with senescence. Wills *et al*. (2008) defined maturity as the fundamental component of fruit quality, particularly horticultural maturity. However, Lee *et al*. (1983), defined physiological maturity as the phase of development when ultimate growth has been achieved so that the following phase of development could be accomplished. Commercial or horticultural maturity, however, is concerned with the time of harvest as related to a particular end-use that could be translated into market requirement which often bears little relation to physiological maturity (Wills *et al*., 2008). Therefore, it is important to allow horticultural products such as avocado fruit to reach their optimum maturity before harvesting and delivering to the market.

Maturity at harvest plays an important role on post-harvest life and eating quality, in particular for climacteric fruits, where ripening is regulated by ethylene (Lelièvre, 1997; Dhatt and Mahajan, 2007). Harvest date, a parameter going along with maturity, contributes to quality and maturity of fruit. Fruit harvested at an immature stage might not achieve normal ripening characteristics (Léchaudel and Joas, 2006). On the other hand, an over mature fruit might deteriorate quickly after harvest (Tefera *et al*., 2007).

Fruit maturity requires determination of some characteristics known to change as fruit develop (Wills *et al*., 2008). Avocado is a climacteric fruit which does not ripen on the tree, thus it should be harvested at the suitable physiological maturity stage to achieve an edible characteristics of taste and firmness (Gamble *et al*., 2010). In avocados, maturation stage could be extended by delaying harvest and allowing the fruit to accumulate more oil. A popular method for determining oil content of avocado fruit is the Soxhlet method (Lee, 1981). However, it is slow, expensive and difficult to perform, hence indirect method were developed as an indication of maturity. In

South Africa, moisture content is the preferred indicator of maturity with recommended moisture content on the range of 69 to 80% depending on the cultivar (Mans *et al*., 1995). This method is based on the high correlation between a fruit increase in dry matter and oil content (Lee *et al.*, 1983; Woolf *et al.*, 2004).

Avocado yields are strongly influenced by the interaction of fruit load and duration of the crop on the tree. Although little information is available on ‘Reed’ cultivar, delayed fruit harvest was studied on ‘Hass’ and ‘Fuerte’ cultivar in South Africa. Kaizer and Wolstenholme (1993) found an increase in fruit size, which increased from count 18 (211-235g) to count 14 (266-305g) when fruit were hanged from July to November (4 months). However, when harvest is delayed, the trees would be required to maintain a double fruit load until harvesting or natural abscission takes place. Since these fruit are known to accumulate oil with development, it is expected that on-tree-storage of mature fruit would exert a continuing demand for assimilate supply, with pronounced effects on tree phenology, subsequently productivity.

2.2.5 Avocado ripening physiology

Ripening is the result of a number of complex physiological and physical changes with distinct anabolic and catabolic processes that require large amounts of energy and prolonged membrane integrity (Bower and Cutting, 1988; Wills *et al*., 1998). For most fruits, ripening correspond with number of coordinated biochemical properties that result in changes in colour, texture, flavour and aroma, making the fruit desirable for consumption (Moing *et al*., 1998). Generally, avocado is one of the most rapidly ripening fruit and often complete ripening within 5-7 days after harvest (Seymour and Tucker, 1993). Being a climacteric fruit, it completes maturation after harvest (Viégas, 1994), with high production of carbon dioxide and ethylene, which triggers the ripening process (Martínez-Romero *et al*., 2003). Due to these characteristics, ripening control is essential to increase the shelf-life after harvest (Kluge, 2002).

Recently, market research has shown that high volumes were sold if the fruit were offered to the consumer in a ready to eat condition. This has led to a system of pre-ripening the fruit prior to stocking. Temperature is increased and controlled after cold storage period to reactivate enzyme activities and initiate the ripening process. Ripening temperature is a highly important factor affecting fruit quality (Blakey *et al*.,

2012) with optimal ripening temperatures depending on the growing and storage temperature. Ripening temperature affects the metabolic rate of the fruit, especially ripening enzymes activity. At relatively lower temperatures (15°C), there are fewer occurrences of fungal rots. Under these conditions, fruit spend less time in a vulnerable state of having highly permeable cell membranes suitable for the pathogen development. However, high ripening temperatures increases the risk of fungal decay, vascular browning and mixed ripening (Hopkirk *et al*., 1994). A temperature of approximately 18-20°C is most suitable for ripening fruit (Hopkirk *et al*., 1994).

2.3 Work not yet done on the problem statement

Avocado maturity studies in South Africa have focused mainly on ‘Fuerte’ and ‘Hass’ as the major cultivars. Thus, the impact of increasing maturity on nutritional content of ‘Reed’ and other minor cultivars has not been established. Furthermore, maturity has shown to influence the response of avocado fruit to the post-harvest treatments. The impact of increasing avocado fruit maturity in response to ripening temperature has not been fully defined for South African avocados.

2.4 Addressing the identified gaps

Avocado fruit demand increases the necessity for searching methods to extend the South African avocado harvest season. Delayed fruit harvest presents a potential to stretch the production season over summer months. However, this would increase the duration of fruit on the tree: therefore, increased fruit maturity. Avocado fruit growth is accompanied by alterations in chemical properties and physical properties as it matures. It is therefore expected to respond differently to different treatments depending on the stage of maturity. In this study, two areas had been identified as still having gaps with respect to delaying harvest of the late maturing ‘Reed’ cultivar as a strategy to extend the South African harvest season viz: (i) influence of fruit maturity on the nutritional status and tree productivity of ‘Reed’ avocado and (ii) impact of fruit maturity in response to ripening temperature. Closing of the identified gaps will assist in increasing the avocado production during the summer months and also increase the South African export volumes during the second export season.

Refinement of ripening temperatures appropriate for a specific fruit maturity would also improve the quality of ripe and ready avocado markets.

CHAPTER 3

IMPACT OF DELAYED HARVEST ON NUTRITION AND TREE PRODUCTIVITY OF ‘REED’ AVOCADO (*Persia americana - Mill*)

3.1 Introduction

Delayed fruit harvest (DFH) or on-tree-fruit storage has been studied in detail only on ‘Hass’ and ‘Fuerte’ avocado cultivars (Kaizer and Wolstenholme, 1993; Whiley *et al*., 1996). However, the strategy has not been studied on several minor and new avocado cultivars including ‘Reed’ avocado. Delay fruit harvest is an alternative strategy being considered to take advantage of November to January late prices on the local market. Furthermore, DFH of late maturing cultivars such as ‘Reed’ can also assist in increasing the market volume during the second half of the South African export season. The South African avocado harvest season extends from February to October, with very low to no fruit during November to January (FPEF, 2015). This avocado fruit availability gap has long been identified (Graham and Wolstenholme, 1991) to potentially increase the avocado industry export market competitiveness.

‘Reed’ is a late Guatemalan race cultivar developed in California. It is an important minor cultivar in California and now produced in South Africa due to its late maturity, high flesh quality, and ability to crop under tropical conditions. Delayed harvest potentially brings a significant fruit size increase (Kaizer and Wolstenholme, 1994) due to the continued cell division and growth in avocado fruit when attached to the tree. On-tree-fruit storage increased both yield and fruit size of ‘Hass’ from count 18 (211-235 g) to count 14 (266-305 g) when fruit were hanged from July to November (4 months) (Kaizer and Wolstenholme, 1993). Whiley *et al.* (1996) also found that fruit size and yield of ‘Fuerte’ was not reduced by late harvesting. However, Kassim *et al*. (2013) showed that late fruit growth beyond physiological maturity was accompanied by alterations in certain chemical properties of the fruit. Generally, it is important to understand these compositional changes, especially late in the growing season to ensure that fruit are harvested at their optimal mineral levels (Ferguson and Boyd, 2002). Therefore, the objective of this study was to assess effects of delayed harvest on mineral, proximate composition, fruit size and yield of late maturing ‘Reed’ avocado fruit.

3.2 Materials and methods

3.2.1 Study site and growth conditions

Avocado fruit were harvested from 10 year old ‘Reed’ trees at ZZ2-Bertie van Zyl farms (Olyfberg) in Limpopo, South Africa (23°42’00”S, 29°54’00”E). The site is characterized by an annual rainfall of >1000 mm, with a monthly maximum and minimum temperatures of 21 and 12°C, respectively. Soil is classified as clay soil (45% clay).

3.2.2 Experimental design, treatments and procedures

Mature ‘Reed’ avocado fruit were harvested at three different maturities *viz*, early (commercial maturity in October), middle (November) and late (December) during 2013 and 2014 harvest seasons. Treatments were laid out in a randomized complete block design (RCBD), with five replications. Treatments were applied to a single tree plot and same trees were used during both seasons. Orchard cultural practices such as fertilization, irrigation, pruning, and pest and disease control followed the ZZ2 management standards.

3.2.3 Data collection

During each harvest time, fruit samples were harvested and sent to Nvirotek Labs (North West Province, South Africa) for proximate and mineral analysis. During each harvest, 2 sets of 5 fruit samples were collected and the first set was used for proximate and mineral analysis while the second set was used for moisture content analysis. Proximate analysis included; protein, crude fiber, ash, starch, oil and moisture content while the minerals analysed included phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), and zinc (Zn). Fruit moisture content was performed at ZZ2 avocado commercial packhouse within the same day of harvest.

Protein determination

Protein content was determined using Dumas combustion method (AOAC, 1990). Approximately 2.0 g of dry pulp samples were combusted at 900°C chamber in the presence of oxygen to release carbon dioxide, water and nitrogen. Thereafter, the gases were passed over potassium hydroxide aqueous solution that absorbs the carbon dioxide and water. A column containing a thermal conductivity detector at the end was then used to separate nitrogen from any residual carbon dioxide and water, and the remaining nitrogen content was measured. The measured signal from the thermal conductivity detector for the samples was converted into nitrogen content. Protein content was calculated as percentage crude protein = N X 6.25, where N= nitrogen content.

Crude fiber determination

Crude fiber was determined using the method of Van Soet and Jeraci (1990). Five grams of dry pulp samples were boiled in 150 ml of 1.25% H_2SO_4 solution for 30 minutes under reflux to remove fats and carbohydrates. The boiled samples were washed in several portions of hot water using a two-fold cloth to trap the particles. Afterwards, samples were returned again to the flask, re-boiled in 15 ml of 1.25% NaOH for another 30 minutes under reflux in order to remove all remaining minerals and complex carbohydrates. Thereafter, the samples were transferred to an ashing dish and dried at 130°C to a constant weight. Afterwards, samples were cooled in a desiccator and weighed. Crude fiber was determined by the difference and calculated as percentage of the weight analysed, thus: Crude fiber (%) = Wt. of residue + ash X 100/ Initial Wt. of sample.

Ash content determination

To determine ash content, dried pulp samples (5.0 g) were weighed into porcelain crucible and placed in a temperature controlled furnace at 600°C for 4 hours until properly ashed. The crucible was then cooled off in a desiccator and immediately weighed. Ash content was calculated as:

$$\text{Ash content} = \frac{\text{Wt. of dry sample}}{\text{Wt. of ash content}} \text{X100}$$

Starch content determination

Starch content was determined using Ewers polarimetric method (AOAC, 1990). Five grams of dried pulp was ground and mixed with 25 ml of hydrochloric acid and immersed in a boiling water bath while shaking to prevent the formation of agglomerates. The samples were cooled to 20°C by mixing with cold water, filtrated and clarified with Carrez solutions. Optical rotation was then measured in a 200 mm tube with a polarimeter (P1). Afterwards, samples were extracted with 40% ethanol before being acidified, filtered and clarified again with hydrochloric acid. A second optical rotation (P2) was measured and starch content determined as the difference between the two measurements multiplied by the optical rotation of pure starch (185.4) using the formula below: Starch content (%) = 2000 (p1-p2)/185.4.

Oil extraction

Oil was extracted by mixing 200 g of the dried pulp samples with petroleum ether (100 ml) as a solvent. The samples were evaporated in a steam bath to remove most of the petroleum ether, thereafter, filtered through a dry, folded paper and dried at 100°C. The weight of oil extract was determined by the difference and calculated as percentage of the weight of sample analyzed, thus:

$$\text{Oil} = \frac{\text{Wt. of sample} - \text{Wt. of residue}}{\text{Wt. of sample}} \text{ X100}$$

Moisture content determination

Moisture content was determined by longitudinally cutting 10 fruit with the seed and seed coat adhering to the flesh removed. The peeled halves were triturated with a kitchen grater consisting of five cutters per square centimeter to get approximately 1 mm thick shreds. Ten (10) grams of the grated avocado pulp samples were thoroughly mixed and weighed in a tarred petri-dish. Moisture content was determined by drying the fresh pulp sample in an oven (400L EcoTherm Digital, Japan) at 30°C for 48 hours. The petri-dish was cooled off in a desiccator and dry weight was measured. The moisture content was calculated by difference using the formula: $\text{Moisture \%} = \frac{\text{Wt. of fresh sample}}{\text{Wt. of dry sample}} \text{ X100}$

Determination of mineral elements

Minerals were determined by dry ashing the dry pulp samples at 65°C to constant weight. The ash was dissolved in a volumetric flask using distilled and deionized water with a few drops of concentrated hydrochloric acid. Potassium was determined with a 410 dual channel flame photometer using NaCI and KCI to prepare the standards, while other minerals (Ca, Mg, P and Zn) were determined with an Atomic

Absorption Spectro-photometer (AAS, 210VGB, Melbourne, Australia). Zinc was reported in mg/kg while other elements were reported in percentages.

Determination of yield and fruit size

Trees were harvested individually and yield was recorded during each harvest time (kg /tree). Fruit were sent through the commercial pack line to determine fruit size distribution (Table 3.1)

Table 3.1: Avocado fruit size distribution codes (NDA Gazette, 2013)

Class	**Fruit weight**
Class 1	576 - 462 g
Class 2	461 - 247 g
Class 3	246 - 174 g
Class 4	173 - 80 g

3.2.4 Data analysis

Data was subjected to analysis of variance (ANOVA) using Statistix 10.0 (Statistical Analytical Software, 1985-2012). Sum of squares were partitioned, while treatment mean separation was achieved using Least Significant Difference (LSD) test at 5% probability level.

3.3 Results

Harvest time significantly affected ($P \leq 0.05$) fruit Potassium (K), Calcium (Ca) and Magnesium (Mg) content on both harvesting seasons (Table 3.2). However, there was no significant difference on Zinc (Zn) content during 2013 when compared with 2014 season, whereby, treatments showed significant differences. Similarly, harvest time affected phosphorus (P) content during 2013 while no significant differences were observed during 2014 season. The concentration of P and K remained constant during early and mid-harvest time, but declined at the late harvest during 2013 season. However, a different trend was observed during 2014, whereby, the levels of both minerals fluctuated with different harvest time. Phosphorus and K were low for the early harvest time, peaked at mid-harvest; and slightly, declined for the late harvest time. Phosphorus content ranged from 0.15 to 0.09 and 0.13 to 0.10% while K content ranged from 2.09 to 1.54 and 1.86 to 1.48% for both 2013 and 2014 seasons, respectively (Table 3.2). Calcium content declined consistently with harvest time during 2013. The levels of Ca declined from a high of 0.05 to 0.02% from early to late harvest, respectively. However, mid-harvest Ca content (0.12%) significantly increased when compared with the early (0.05%) and late (0.02%) harvest time during 2014. Magnesium content decreased steadily with harvesting time from early (0.14%) to late (0.06%) during 2013 season in contrast to 2014, where Mg level remained constant during early and mid-harvest, and thereafter, decreased during late harvest. Zinc content remained constant over the three harvesting times during 2013, while mid- harvest fruit had high zinc content while early and late harvest times showed lower but not significantly different Zn content in 2014.

Table 3.2: Effects of delayed fruit harvest on mineral composition of 'Reed' avocado fruit

Season	Harvest time	Phosphorus (%)	Potassium (%)	Calcium (%)	Magnesium (%)	Zinc (mg/kg)
2013	Early	0.14a	1.98a	0.05a	0.14a	28.00a
	Mid	0.15a	2.09a	0.02b	0.11b	29.60a
	Late	0.09b	1.54b	0.03b	0.06c	29.80a
P-value at 0.05		0.02*	0.02*	0.01*	0.00*	0.90*
2014	Early	0.10b	1.48b	0.05b	0.10a	13.20b
	Mid	0.13a	1.86a	0.12ab	0.10a	23.00a
	Late	0.12ab	1.71ab	0.26a	0.9b	14.60b
P-value at 0.05		0.06 ns	0.01*	0.00*	0.01*	0.00*

Values with same letters were not significantly different at 0.5 probability level

* Significantly different at P=0.05, ns Not significant at P≤ 0.05

Delayed fruit harvest significantly (P≤0.05) affected the entire proximate analysis variables measured during both seasons (Table 3.3). Protein content was constant during early and mid-harvest time, but decreased during late harvest from high of 5.34 to 3.58% and 5.43 to 3.56% during 2013 and 2014 seasons, respectively. While, moisture content decreased with increasing fruit maturity during both seasons. Moisture content dropped from 74.0 to 65.0% and 75.6 to 66.0% from early to late harvest times during both seasons, respectively. Furthermore, there was a progressive significant increase in oil and starch content as the fruit hanged longer on the trees. Oil content increased steadily from 40 to 64% and maintained the same trend for both seasons. However, starch content increased from early to mid-harvest, and thereafter, levelled-off through late harvest time. Starch content increased from 3.2 to 5.5 % and 3.1 to 4.59% from the early to mid-harvest during 2013 and 2014 seasons, respectively. High crude fibre content was recorded on early and late harvest times while mid-harvest time resulted in the least fibre content during both seasons. Ash content was low during early harvest, increasing sharply during mid-harvest time, and thereafter, decreased during late harvest time.

Table 3.3: Effects of delayed fruit harvest on proximate composition of 'Reed' avocado fruit

Season	Harvest time	Protein (%)	Crude fiber (%)	Ash (%)	Starch (%)	Oil (%)	Moisture (%)
2013	Early	5.26a	28.62a	3.22b	3.24b	39.96c	74.88a
	Mid	5.34a	21.02b	3.72a	5.02a	44.24b	72.32a
	Late	3.58b	30.38a	3.50ab	5.20a	63.86a	65.06b
P-value at 0.05		0.04*	0.00*	0.03*	0.01*	0.00*	0.00*
2014	Early	5.43a	29.11a	3.12b	3.10b	38.19c	75.60a
	Mid	5.28a	20.73b	3.65a	4.92a	43.51b	71.00b
	Late	3.56b	30.23a	3.45a	4.59a	64.74a	66.00c
P-value at 0.05		0.00*	0.00*	0.00*	0.00*	0.00*	0.00*

Values with same letters were not significantly different at 0.5 probability level

* Significantly different at P=0.05, ns Not significant at P≤ 0.05

Treatments showed no significant difference (P≤0.05) in yield during 2013, while, significant differences were observed during 2014 (Table 3.4). Yield dropped from 42.21 to 12.26 kg^{-1} tree from early to late harvest time while higher yields were observed during early and late harvest times in 2014 season. Treatments showed no significant difference (P ≤ 0.05) on class 1, 3 and 4 fruit during both 2013 and 2014 harvesting season (Table 4), while the class 2 fruit differed significantly during 2013. The percentage of class 1, 3 and 4 fruit remained the same during both seasons while the percentage of class 2 fruit decreased with harvest time during 2013 season. The percentage of class 2 fruit dropped from 62.6 to 31.8% from early to late harvest time.

Table 3.4: Effects of delayed fruit harvest on yield and fruit size distribution of 'Reed' avocado fruit

Season	Harvest time	Yield (kg/tree)	Class 1 (%)	Class 2 (%)	Class 3 (%)	Class 4 (%)
2013	Early	42.21ab	20.60a	62.60a	15.40a	1.20a
	Mid	59.11a	42.00a	35.40b	16.00a	2.60a
	Late	12.26b	49.60a	31.80b	15.40a	0.60a
P-value at 0.05		0.09^{ns}	0.15^{ns}	0.01*	0.99^{ns}	0.46^{ns}
2014	Early	54.68a	35.60a	50.20a	11.20a	1.20a
	Mid	24.16b	43.00a	36.40a	13.60a	3.00a
	Late	52.20b	40.40a	42.00a	12.80a	2.40a
P-value at 0.05		0.04*	0.93^{ns}	0.54^{ns}	0.92^{ns}	0.62^{ns}

Values with same letters were not significantly different at 0.5 probability level

* Significantly different at P=0.05, ns Not significant at P≤ 0.05

3.4 Discussion

Our study hypothesised that delaying fruit harvest by either one or two months after physiological maturity under South African growing conditions would not affect the nutritional value and yield of 'Reed' cultivar. However, significant differences in mineral content, proximate composition and yield were found between the various harvest times.

Phosphorus (P) and potassium (K) levels were significantly low for the late harvest (December) during both seasons (Table 3.2). Such results were in agreement with findings of Rosecrance *et al.* (2012), whereby, early harvested 'Hass' fruit were associated with higher levels of P and K, but decreased later as the season progressed. The decline in both P and K was attributed to their uptake pattern during fruit development. The movement and accumulation of P and K into the fruit is closely related to fruit dry matter, with a reduction or possibly ceasing of accumulation after reaching maturity (Clark and Smith, 1988). These results suggested that delaying fruit harvest results in lower mineral levels due to the dilution of minerals during the increasing fruit growth. During 2013, levels of both P and K were low at 0.09% and 1.54%, respectively. However, these values were still higher than the standard mineral recommended dietary allowance values of P (0.04%) and K (1%) (Ortega *et al*., 2013)

Fruit calcium (Ca) content decreased with advancement in fruit maturity. Results were in line with the findings of Cutting *et al*. (1992), who reported calcium decline on 'Fuerte' from a high of 0.018% to 0.012% from first to latest harvest date, respectively. However, the current study recorded a higher resultant Ca reduction of 60% between the early and late harvest time during both seasons. Calcium is an immobile element and its accumulation in the fruit slows down with increasing fruit maturity (Knee, 2002). Furthermore, high available soil K and Mg often result in lower fruit Ca through a reduction in Ca root uptake (Coates *et al.,* 1997). The importance of Ca as a determinant of post-harvest fruit quality has been recognised for many years (Koen *et al.*, 1990). According to Thorp *et al*. (2010), harvesting fruit too late resulted in fruit that were quick to ripen, highly susceptible to rots and physiological damage, especially with low Ca levels. In this study, higher Ca levels were observed than the established threshold Ca content of 25 to 28 mg/100g dw for South African avocados that was found to practically have no vascular or flesh browning (Cutting and Bower, 1992).

An accumulation of Magnesium (Mg) decreased systematically during each harvest time. Similarly, Cutting *et al*. (1992) reported Mg content decrease from a high of 0.08% at the first harvest date to a low of 0.06% at late harvest date. Fruit Mg accumulation is xylem based and highly active during the early phase of fruit development. Later in the season, with an increased fruit dry matter accumulation, phloem tends to provide a predominant flow, resulting in low or no supply of xylem based elements to the fruit (Knee, 2002). Zinc (Zn) content remained constant throughout the season during 2013 season, but a significant increase was recorded during mid-harvest time of 2014 season. Interestingly, this significant Zn content increase coincided with Ca content increase. In fruit trees, Zn content increases Ca levels by reducing levels of oxalate bound Ca and improve the strength of water retention by the fruit tissues (Shear, 1980).

Protein content remained constant during early and mid-harvest but declined during the late harvest (Table 3.3). On the contrary, Blakey *et al*. (2009) and Graham and Wolstenholme (1991) found 'Hass' fruit protein content to remain constant throughout the season despite the increasing fruit maturity. Reduction in protein content observed in this study could be attributed to an indirect calculation from nitrogen concentration using a conversion constant of 6.25. Therefore, the differences in protein concentration might merely reflect differences in nitrogen content in the fruit which was also low on the late harvested fruits (data not included).

In relation to crude fibre, levels ranged from 20 to 30%, with the lower levels recorded during the mid-harvest time. However, there was no significant difference in crude fibre between the early and late harvest time. Avocado fruit have high fibre content among other fruits, comprising 75% insoluble and 25% soluble fibre (Oluwole *et al*., 2013). The observed fruit fibre content increase during the late harvest time might have been due to increased biosynthesis and accumulation of fibre components (pectin, cellulose and hemicelluloses) as the fruit hanged longer on the trees (Msogoya *et al*., 2004). A similar trend was also reported on citrus, whereby, dietary fibre contents increased with advanced fruit maturity (Xiuli and Guanlun, 2007).

Ash content didn't differ significantly between the three harvests on both seasons. However, the ash content was three times higher when compared with those reported for 'Fuerte' (Pearson, 1975). Starch content increased with increasing fruit maturity. These starch trends were consistent with the work of Burdon *et al*. (2010) who reported starch content to increase steadily as the season progressed. According to Whiley *et al*. (1988), while fruit are attached to the tree and increasing in maturity concomitantly, the tree continues with its normal phenological cycle. This implies periods of strong competition for limited carbohydrates during new vegetative and flowering growth flushes, intensifying during late maturity when flowering and fruit set occurs for the subsequent season. However, Burdon *et al*. (2010) elucidated that fruit starch content is not simply reduced by direct competition with vegetative or reproductive growth but more related to the overall availability of carbohydrates within the tree. In this study, the increasing fruit starch content observed suggested that the trees had high carbohydrates reserves to maintain the normal tree cycle while carrying the hanging fruit from the previous season.

The high oil content observed suggested that ‘Reed’ could be an important cultivar for oil production. Oil ranged from 39 to 64% from early to late harvest times. According to Yousef and Hasseine (2010), oil content increases in the mesocarp after fruit set and could be correlated with the fruit age. Oil increased by 4.28% from early to mid-harvest and 19.62% from mid to late harvest. These trends were in line with findings of Hofman *et al*. (2000). Blakey *et al*. (2012) reported increased oil content to be higher between mid and late harvest when compared with early and mid-harvest. This was also in agreement with Lizana *et al*. (1992) who found increased oil content to be more pronounced late in the season when compared with early to mid-season fruit. Mostert *et al*. (2007) suggested that increased oil content was due to increased cell wall degradation, resulting in improved accumulation of oil on the fruit mesocarp.

Moisture content decreased from 74.8 to 65.0% and 75.0 to 66.0% between the early and late harvest during 2013 and 2014. These results lend further support to the reduction in moisture content observed in previous studies (Osuna-Garcia *et al*., 2010; Yousef and Hasseine, 2010). In South African avocado industry, moisture content is the preferred indicator of maturity with the recommended moisture content in the range of 69 to 75% depending on the cultivar (Mans *et al*., 1995). Currently, ‘Reed’ cultivar is classified as “other cultivars” under the South African standards and requirements for control of the export of avocados with a specified limit of 75% moisture content at harvest (NDA, 2013). The observed moisture reduction was due to increased oil content as the fruit maturity advanced. Moisture content has an inverse relationship with oil content (Kruger *et al*., 1995) and as the oil content increase with maturity, moisture content decrease by the same amount.

Delaying fruit harvest by two months reduced the fruit yield during 2013 season (Table 3.4). Yield showed a reduction of 30% from the early to late harvest times. However, Newett *et al.* (2002) reported that 'Reed' fruit could be hanged on tree for one month longer than 'Hass' cultivar under California avocado growing conditions without affecting the subsequent crop. In this study, the variation observed could be attributed to tree to tree variation and nature of the seedling trees used. The yield did not differ significantly across the harvesting times during the second season with an average of 55 kg/tree. Kaiser and Wolstenholme (1993), found no significant differences on 'Hass' yield during the first two seasons but significant decline was obtained in the third season. This justified further investigation into the potential long term effects of delaying fruit harvest consecutively on tree productivity and possibility of alternate bearing.

Class 1 and 2 fruit dominated during both seasons with each class accounting an average of 37 and 39, 43 and 42% during 2013 and 2014, respectively. However, the percentage of class 2 fruit was significantly lower during the mid and late harvesting times when compared with early harvest time during 2013 season. Generally, avocado fruit size was highly variable. Avocado fruit set over an extended period resulting in mixed fruit size classes on the tree. However, in the study conducted by Graham and Wolstenholme, (1991), there was an increase in larger fruit, notably, class 2 fruit, corresponding with decreased proportion of smaller fruit.

3.5 Conclusions

The present investigation revealed that the mineral and nutritional value of 'Reed' avocado fruit varies with fruit maturity. 'Reed' fruit mineral content didn't differ significantly between the early and mid-season, but showed to undergo mineral degradation late in the season. Fruit harvested two months after reaching the minimum picking maturity had low fruit P, K, Ca, Mg and Zn content compared with early and mid-season fruit. However, oil, starch and ash content increased steadily with increasing maturity, and resulted in fruit of high nutritional value during late season. Delaying fruit harvest reduced yield during the first season, but showed no treatment effect on the following season. Furthermore, there was no treatment effect on fruit size distribution.

CHAPTER 4
EFFECT OF DELAYED HARVEST AND RIPENING TEMPERATURE ON THE POST-HARVEST QUALITY OF REED AVOCADO

4.1 Introduction

Fruit quality is the function of physiological status of the commodity and its environment (Kaiser *et al*., 1995). Avocado is a highly perishable fruit with high metabolic rate, and therefore, has short shelf-life (4 weeks) when stored at optimum temperatures and humidity (Yahia and Gonzalez-Aguilar, 1998). The climacteric nature of avocado fruit prevents it from being preserved for longer periods after harvest at room temperatures. Therefore, temperature management becomes a critical component in the post-harvest life of avocado fruit (Lee and Kader, 2000). Low storage temperature is used to reduce the metabolic activity, particularly ripening enzymes and ethylene production occurring only at certain temperature range to extend the fruit shelf-life (Donkin, 1995). According to Medicott *et al*. (2006), ripening temperature has a great influence on the shelf-life, colour, flavour and texture development of the fruit. Temperature also affects the metabolic rate of the fruit, especially the activity of the ripening enzymes. Moreover, post-harvest diseases and disorders have been shown to increase with the increasing temperature due to the ethylene production during ripening (Hopkirk *et al*., 1994). It is therefore essential to maintain a balance between optimal ethylene production and limiting the post-harvest physiological disorders and pathological diseases.

Fruit maturity at harvest is an important factor determining storage-life and the final fruit quality (Kader, 1999). Maturity has shown to influence the response of avocado fruit to the post-harvest treatments with the early season fruit at high moisture content being more susceptible to cold storage disorders when compared with more mature fruit (Ernest, 2007). However, the impact of increasing avocado fruit maturity in response to ripening temperature has not been fully defined for South African avocados. Thus, the objective of this study was to evaluate the effect of delayed harvest and ripening temperature on the post-harvest quality of ‘Reed’ avocado fruit.

4.2. Materials and methods

4.2.1 Experimental sites

Fruit were harvested from 10 years old trees at ZZ2-Bertie van Zyl farms (Olyfberg) in Limpopo, South Africa (23°59’10”S, 29°44’15”E). The site is characterised by an annual rainfall of >1000 mm, with a monthly maximum and minimum temperatures of 22.5 and 14°C, respectively. Soil is classified as clay (45 % clay). After each harvest, fruit were transported to the Agricultural Research Council-Institute of Tropical and Subtropical Crops (ARC-ITSC) post-harvest laboratory for storage and post-harvest quality assessments.

4.2.2 Experimental design, treatments and procedures

A 3 x 3 factorial experiment arranged in randomized complete block design (RCBD) with five replicates was used. Fruit were harvested at three different maturities *viz*, early (commercial maturity October), middle (November) and late (December) during 2013 and 2014 seasons. Fruit moisture content was at 75, 72 and 65 % and 75, 71 and 66 % during 2013 and 2014 for early, mid and late harvest times, respectively. After each harvest, fruit were carefully graded into uniform size and packed into 4 kg perforated cartoon boxes and stored at 5.5°C (80 ± 5 % relative humidity) in forced air cooling room for 28 days. After cold storage, fruit were divided into three lots of 5 replicates consisting of six fruit and ripened at three different temperatures *viz*, 16, 21 and 25°C (Table 4.1).

Table 4.1: Treatment combinations for the experiments

Harvesting time	**Ripening temperature**		
	16°C	**21°C**	**25°C**
Early (E)	E-16°C	E-21°C	E-25°C
Middle (M)	M-21°C	M-21°C	M-25°C
Late (L)	L-25°C	L-21°C	L-25°C

4.2.3 Data collection

Determination of disorders on ripe fruit

Fruit quality was assessed after ripening at 16, 21 and 25°C. Days to ripening were recorded as number of days after withdrawal from cold storage (5.5°C) to fully ripening. Ripe fruit were cut longitudinally and evaluated for incidence of chilling injury (diffuse areas of discoloured flesh with poorly defined margins), vascular browning (browning of the vascular strands running longitudinally through the fruit tissue), anthracnose (rounded, dark-coloured, sunken lesions that expand rapidly on the fruit skin and into the pulp causing rot) and stem-end rot (a dark rot that develops from the stem end as fruit ripen after harvest and produces dark streaking of the water-conducting tissues) incidence (number of fruits showing symptoms) according to White *et al*. (2004).

CO_2 determination

Fruit from each ripening temperature were individually weighed, placed in 1.5 L jars and sealed for 1 hour with a cap and a rubber septum. After an hour, a gas sensor (250 -Dual Gas Analyser, Osaka, Japan) was connected to the chamber to monitor CO_2 levels. The levels of CO_2 were evaluated on daily basis and expressed as respiration in µmol CO_2 /kg/hr.

Fruit colour determination

Colour characteristics were assessed using a Chromameter (CR-400, Konica Minolta, Osaka, Japan). Values were obtained on the basis of CIELAB colour system (L, a*, b*, C* and h^0) where L describe the lightness or brightness [L= 0 (black) and L = 180 (white)], a^* specify the greenness or redness (where - a* indicates greenness whereas +a* means redness) and *b** indicates yellowness or blueness (where - b* indicates blueness whereas +b* means yellowness). Chroma (C*) value indicates the degree of saturation of colour and was proportional to the strength of the colour whereas, Hue angle (h^0) was the basic unit of colour (0^0 = red; 90^0 = yellow; 180^0 = bluish-green and 270^0 = blue). Measurements were taken from five samples and the average of *L, a** and *b** values were obtained. Hue angle (h^0) and Chroma (C*) values were calculated based on a^* and *b** according to the following formulas: Hue angle (h^0) = 180^0 + $[\tan(a*/b*)]$ and Chroma (C*) =$\sqrt{(a*)2+(b*)2}$, (McGuire, 1992). Colour was measured on daily basis until the fruit were declared fully ripe.

Determination of firmness

Fruit firmness was measured using a non-destructive Sinclair firmometer (Sinclair IQ™ International, Norwich, United Kingdom) on daily basis during ripening period. Firmness of 6 fruit from each ripening temperature was measured objectively by averaging the four measurements taken on the equatorial region of the fruit. Fruit were considered to be fully ripe at 25 Sinclair units (SU).

Determination of fruit weight loss

Fruit weight loss was measured from 6 fruit per replicate of each ripening temperature. Fruit weight was measured on daily basis until the fruit reached designated ripening stage. Weight loss was determined by measuring the sample on a digital balance scale (SBA 61, Scale Tec, Goettingen, Germany) and was reported as percentage of weight loss based on the original mass.

4.2.4 Data analysis

Data were subjected to analysis of variance (ANOVA) using GenStat 12.1 (VSN International, Hemel Hempsted, UK) 2011. Means were separated at the 5% level of significance. Treatments discussed under section 4.3.1 were significant at 5% level of probability while the interactive trends under section 4.3.2 were discussed regardless of the treatment effect.

4.3 Results

4.3.1 Effect of harvest time and ripening temperature on the post-harvest quality of 'Reed' avocado fruit.

Harvest time and ripening temperature had a significant effect ($P \leq 0.05$), on chilling injury of 'Reed' avocado fruit while their interaction had no significant effect ($P \leq 0.05$) during 2013 harvest season. Fruit harvested late showed higher incidences of chilling injury (48 %) compared with early (31%) and mid (21%) harvest time (Figure 4.1A). Similarly, the incidence of chilling injury increased with increasing ripening temperature. Fruit ripened at 16°C showed low chilling injury incidences followed by 21 and 25°C (Figure 4.1B).

The main effects of harvesting time and ripening temperature had no significant (P ≤ 0.05) effect on chilling injury during 2014 season, while their interactive effect was highly significant (P ≤ 0.05). The incidence of chilling injury increased with ripening temperature during early and mid-harvest time. The incidences of chilling injury during early harvest were 3, 7 and 77 % for fruit ripened at 16, 21 and 25°C, respectively (Figure 4.1C). Similar trend was observed during the mid-harvest with values of 13, 20 and 33 % for fruit ripened at 16, 21 and 25°C, respectively. However, a different trend was observed on late harvest time, whereby, chilling injury decreased with increased ripening temperatures. Furthermore, fruit ripened at 16°C showed higher chilling injury incidences of 36 %, while, there was no significant difference between fruit ripened at 21 (20 %) and 25°C (10 %).

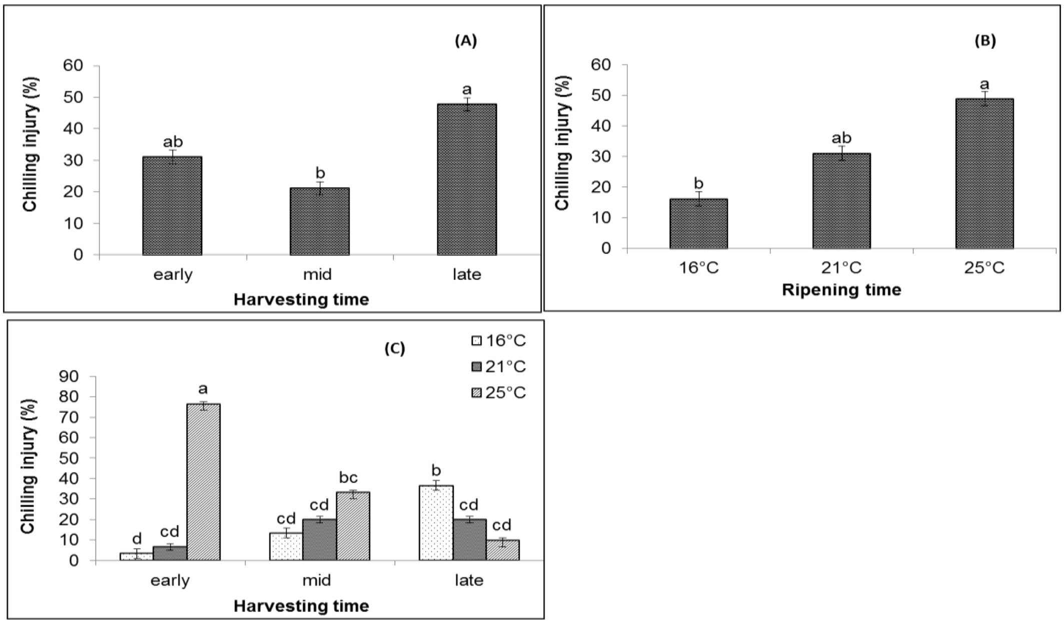

Figure 4.1: Effects of harvest time (a) and ripening temperature (b) in 2013 and their interactive effect (c) on chilling injury in 2014

The incidence of anthracnose was significantly (P ≤ 0.05) affected by the interaction of harvest time and ripening temperature during both 2013 and 2014 seasons. In 2013, the incidence of anthracnose was significantly higher at 21 (40%) and 25°C (37%) when compared with 16°C (30%) ripening temperatures during early harvest time, while there was no significant difference among the ripening temperatures at the mid-harvest time (Figure 4.2A). However, fruit ripened at 21°C showed a higher incidence of 63% compared fruit ripened at 16 (60%) and 25°C (50%) at the late harvest time.

Similar trend was observed during the 2014 season, whereby, fruit ripened at 25°C had higher incidences (56%) of anthracnose when compared with fruits ripened at 21°C (40%) and 16°C (33%) during the early harvest time (Figure 4.2B). There was no significant treatment effect among the ripening temperatures during mid-harvest time. However, fruit ripened at 21 (64%) and 16 (63%) showed significantly higher incidences of anthracnose during the late harvest time when compared with fruit ripened at 25°C (36%).

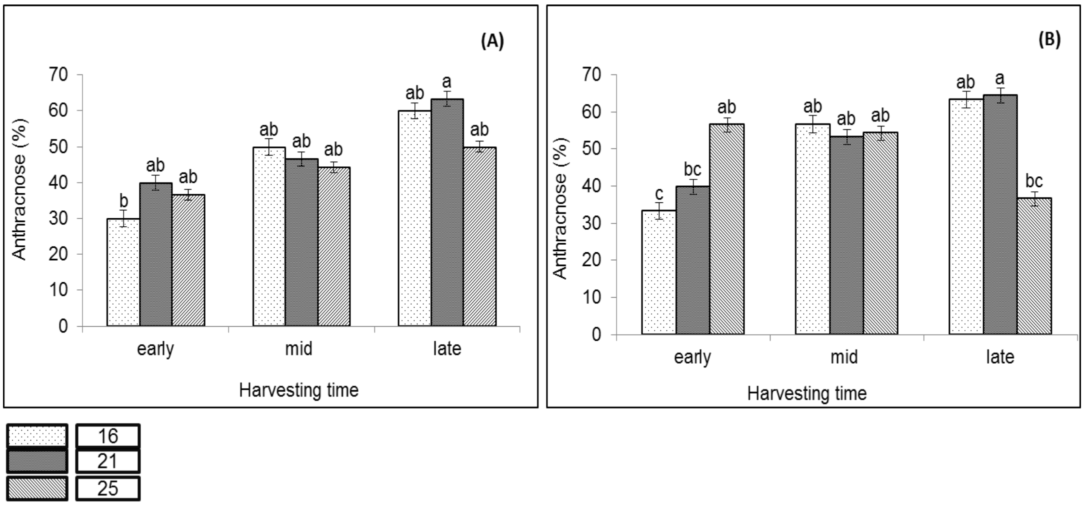

Figure 4.2: Interactive effect of harvest time and ripening temperature on the incidence of anthracnose during (a) 2013 and (b) 2014 seasons

Interaction effect of harvest time and ripening temperature on vascular browning showed significant ($P \leq 0.05$) difference during 2013 season. Vascular browning was significantly higher on fruit ripened at 16°C when compared with 21 and 25°C during the early harvest time. The incidence of vascular browning was 40, 17 and 10% for fruit ripened at 16, 21 and 25°C, respectively (Figure 4.3A). Similarly, vascular browning decreased with increasing temperature during the mid-harvest time. However, a different trend was observed during the late harvest time with fruit ripened at 25°C, whereby, a significantly higher incidences of vascular browning were observed when compared to fruit ripened at 16°C and 21°C. The highest incidences of vascular browning were recorded on fruit ripened at 25°C during the late harvest, while the lowest incidences were recorded during mid harvest time under similar temperature regime.

The main effect of ripening temperature significantly influenced the incidence of vascular browning, while the interaction of harvest time and ripening temperature had no significant ($P \leq 0.05$) effect on vascular browning during 2014. Fruit ripened at lower temperature (16°C) had significantly higher incidences of vascular browning, while there was no significant difference between fruit ripened at 21 and 25°C (Figure 4.3B).

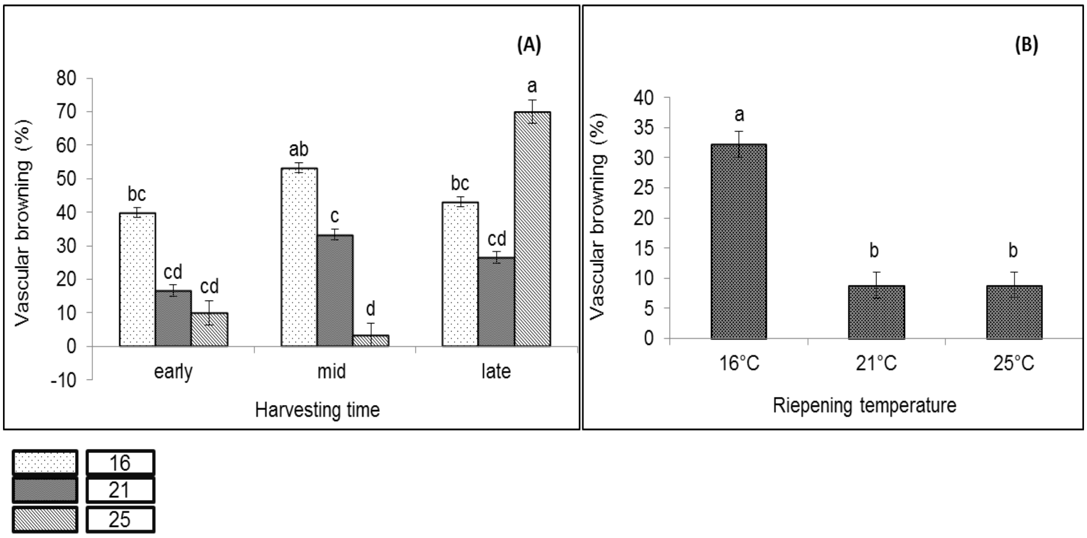

Figure 4.3: (a) Interactive effect of harvest time and during 2013 and (b) main effect of ripening temperature on incidence of vascular browning during 2014 season

Significant effect of ripening temperature on stem-end rot was observed during 2013 season, while harvest time and interaction effects were not significant ($P \leq 0.05$). Stem-end rot was significantly higher on fruit ripened at 16 (19%) and 21°C (27%), while the lowest incidence was recorded on fruit ripened at 25°C (7%). There was no significant effect of the main factors or interaction during 2014 season.

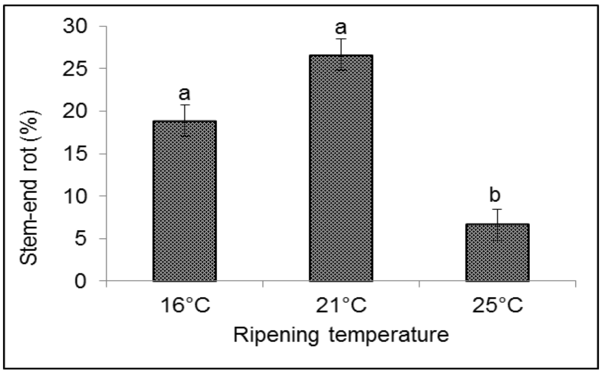

Figure 4.4: Effect of ripening temperature on the incidence of stem-end rot during 2013 season

4.3.2 Effects of harvest time and ripening temperature on skin colour, firmness, respiration rate and fruit weight loss of 'Reed' avocado fruit during ripening

Fruit lightness (*L*) gradually increased throughout the ripening period during early and mid-harvest times, regardless of ripening temperature during 2013 season (Figure 4.5A). Lightness increased from values of 30 to 47, 30 to 40 and 31 to 43 on day 0 to fully ripening day when held at 16, 21 and 25°C, respectively. However, a different trend was observed with late harvest time, whereby, fruit held at 16 and 25°C only increased by 6 and 21% from day 0 to day 2, respectively, and remained constant until fruit ripened fully. Lightness of fruit ripened at 21°C showed an insignificant increase from day 0 to day 2, afterwards, decreased until fruit ripe fully. A corresponding increasing trend was observed with early harvest time during 2014 (Figure 4.5B). However, fruit ripened at a lower temperature showed lower L values throughout the ripening period when compared with fruit ripened at higher temperatures (21 and 25°C). Mid-harvest fruit ripened at 25°C showed significantly higher L values which increased from a value of 40 to 49 from day 1 to 4. While mid-harvest fruit ripened at 16 and 21°C only increased non-significantly in lightness from day 0 to 2, and remained constant until fruit were fully ripe. Fruit lightness was similar across the three ripening temperatures during the late harvest time. Furthermore, fruit lightness did not differ between the early and mid-harvesting times but was significantly lower during the late harvest. Moreover, lightness showed least changes during ripening time across all the ripening temperatures.

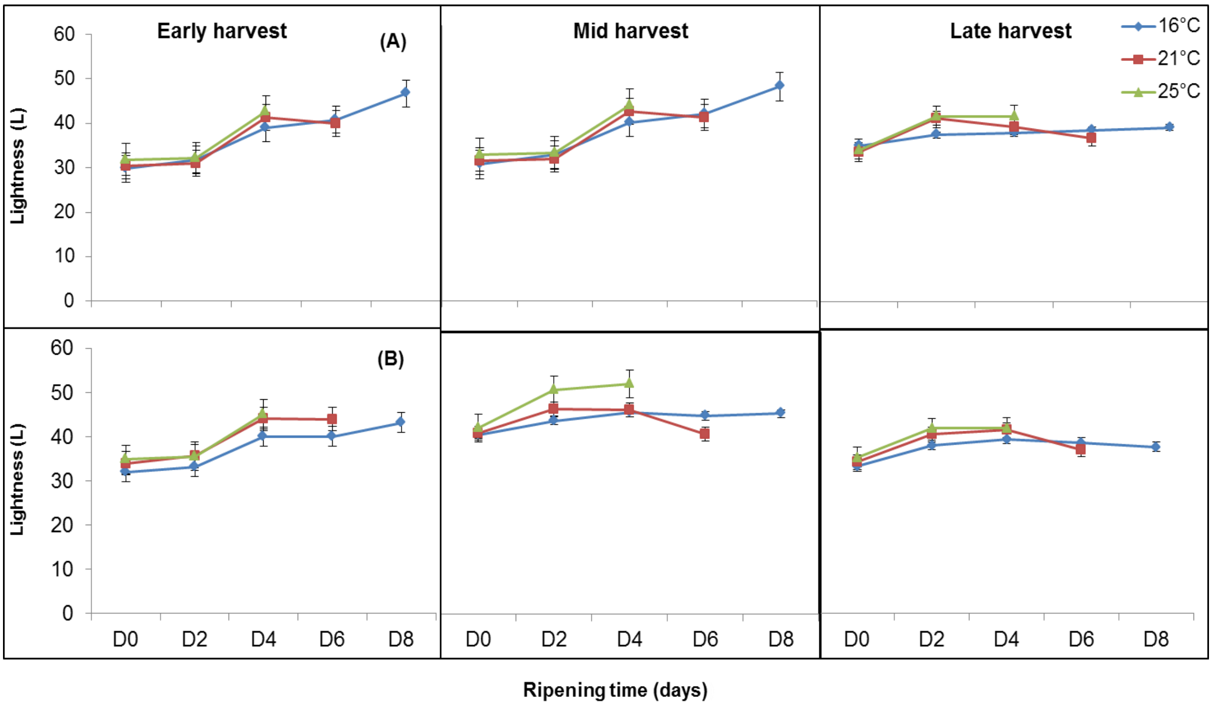

Figure 4.5: Changes in lightness (*L**) values of fruit harvested at different maturities during ripening at different temperatures in 2013 (a) and 2014 (b)

Hue angle values decreased with ripening time at each ripening temperature during the early harvest of 2013 season. Hue angle remained constant from day 0 to day 2, but decreased gradually until ripening. However, mid and late harvest fruit maintained similar hue angle throughout ripening with no difference observed between the ripening temperatures (Figure 4.6A). A similar trend was observed for early harvest, whereby, hue angle values decreased after day 2 of ripening during 2014. However, there was a difference among the ripening temperatures in 2014 (Figure 4.6B). Fruit ripened at 16°C had higher hue angle values (145) and showed higher magnitude of decrease (33 %) between day 4 and 6 followed by fruit ripened at 16 (141) and 21°C (139), respectively.

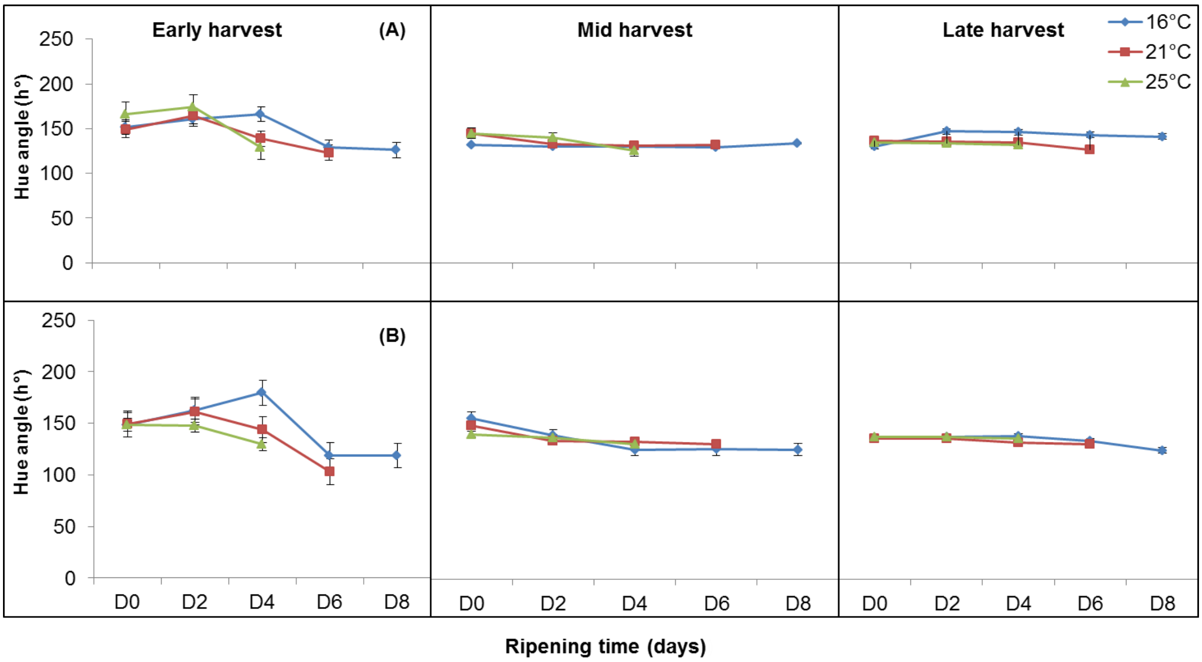

Figure 4.6: Changes in hue angle (h^0) values of fruit harvested at different maturities during ripening at different temperatures in 2013 (a) and 2014 (b)

Chroma values were similar at the beginning of the shelf-life (day 0) for early and mid-harvest times, however, significantly decreased for late season fruit during 2013 (Figure 4.7A). Chroma values decreased gradually with ripening time for fruit held at 16 and 25°C, while fruit held at 21°C showed a rapid decrease between day 0 and day 2, and levelled off until fully ripened at day 6 for early and mid-harvest times. Fruit ripened at 21 and 25°C showed similar trends on the late harvesting time with a higher magnitude of decrease in chroma between day 0 and day 2, but increased slightly as ripening progressed. In 2014, fruit harvested early and ripened at 16°C showed the same trend as during 2013. However, fruit ripened at 21 and 25°C showed a decrease in chroma at day 2, afterwards, increased as ripening progressed (Figure 4.7B). Mid-harvest fruit maintained a decreasing trend when fruit were ripened at 21 and 25°C, while lower ripening temperature showed an increase at day 4, afterwards, decreased until fully ripe. Late harvest fruit maintained the same trend across all the ripening temperatures as during 2013.

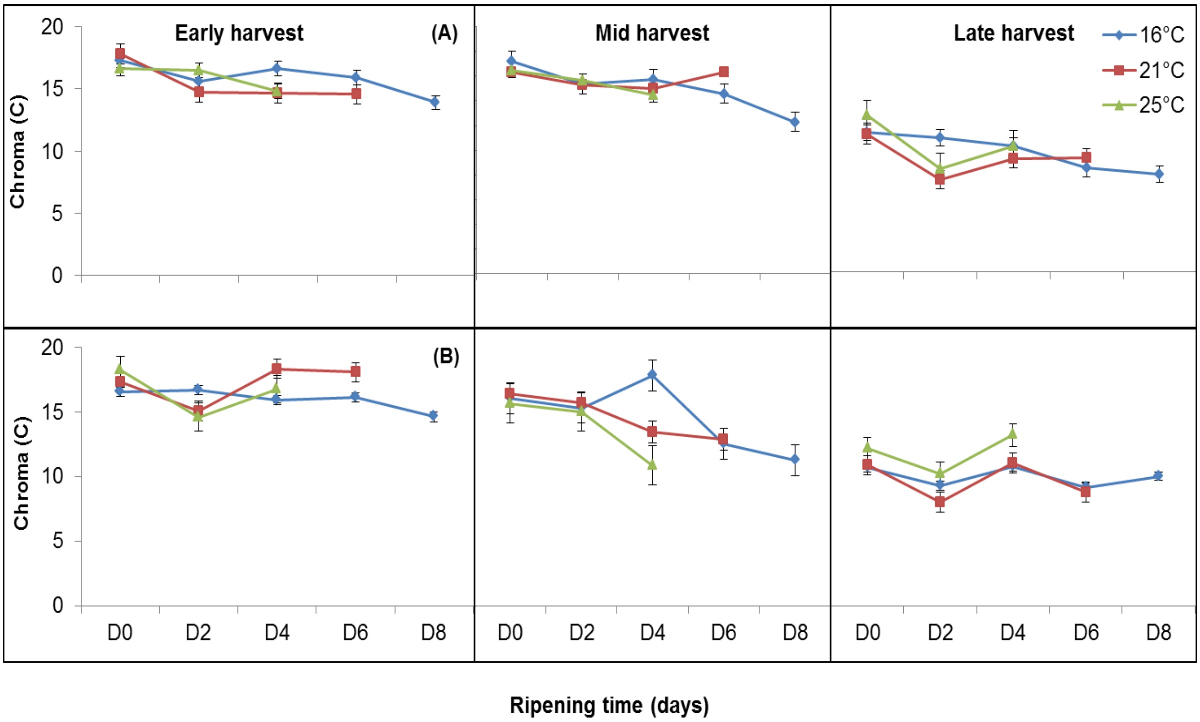

Figure 4.7: Changes in chroma values (C) of fruit harvested at different maturities during ripening at different temperatures in 2013 (a) and 2014 (b)

Avocado fruit showed a respiratory pattern characteristic of climacteric fruits under each ripening temperature during both harvest seasons. At all harvesting times, respiration rate was higher on fruit ripened at 25°C (Figure 4.8A). In 2013, fruit reached the respiratory climacteric peak of 5139, 5095, 5737 µm CO_2/kg/hr on day 2 during early, mid and late harvesting times, respectively. Fruit held at 21°C showed a similar respiratory pattern with a respiratory peak observed on day 2 of ripening. The fruit reached a respiratory peak of 4689, 3680 and 5788 µm CO_2/kg/hr during early, mid and late harvest times, respectively. However, the start-up values of fruit ripened at 16°C increased gradually during ripening time to a peak in day 4, afterwards, the trend decreased until the fruit were fully ripe. The respiration rate was lower at 16°C regardless of harvesting time. Corresponding trend was observed on early and late harvest times during 2014. However, mid-harvest fruit ripened at 21°C showed similar respiration trend as those ripened at 25°C with respiratory peak at day 4 (Figure 4.8B).

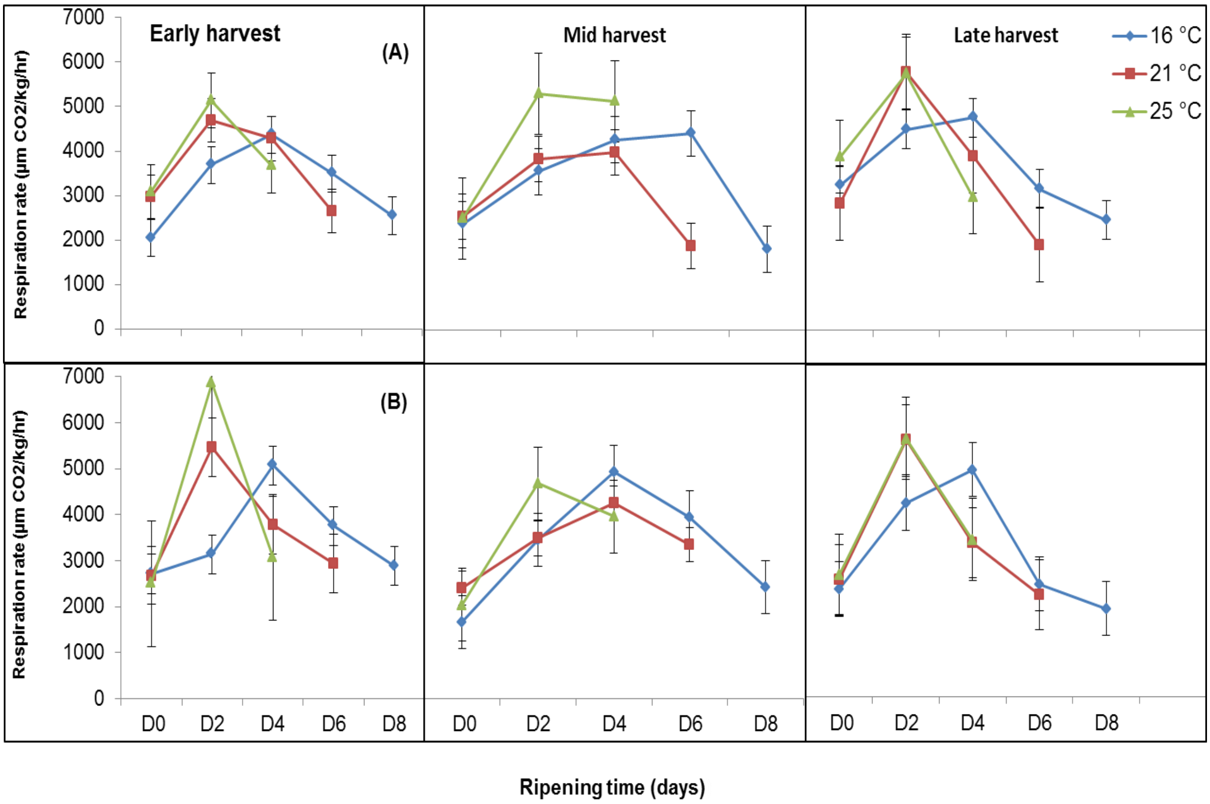

Figure 4.8: Respiration rate of fruit harvested at different maturities during ripening at different temperatures in 2013 (a) and 2014 (b)

Fruit firmness was similar at the beginning of shelf-life (day 0) for all harvesting times during 2013. The response of ‘Reed’ avocado fruit to harvest time was consistent, however, decreased with ripening at different ripening temperatures. In 2013, firmness was reduced by 72, 67 and 54 % at 16, 21 and 25°C, respectively. Firmness decreased rapidly at 25°C followed by 21 and 16°C (Figure 4.9A). Fruit ripened within 8, 6 and 4 days at 16, 21 and 25°C, respectively. Similar results were observed during 2014 season (Figure 4.9B).

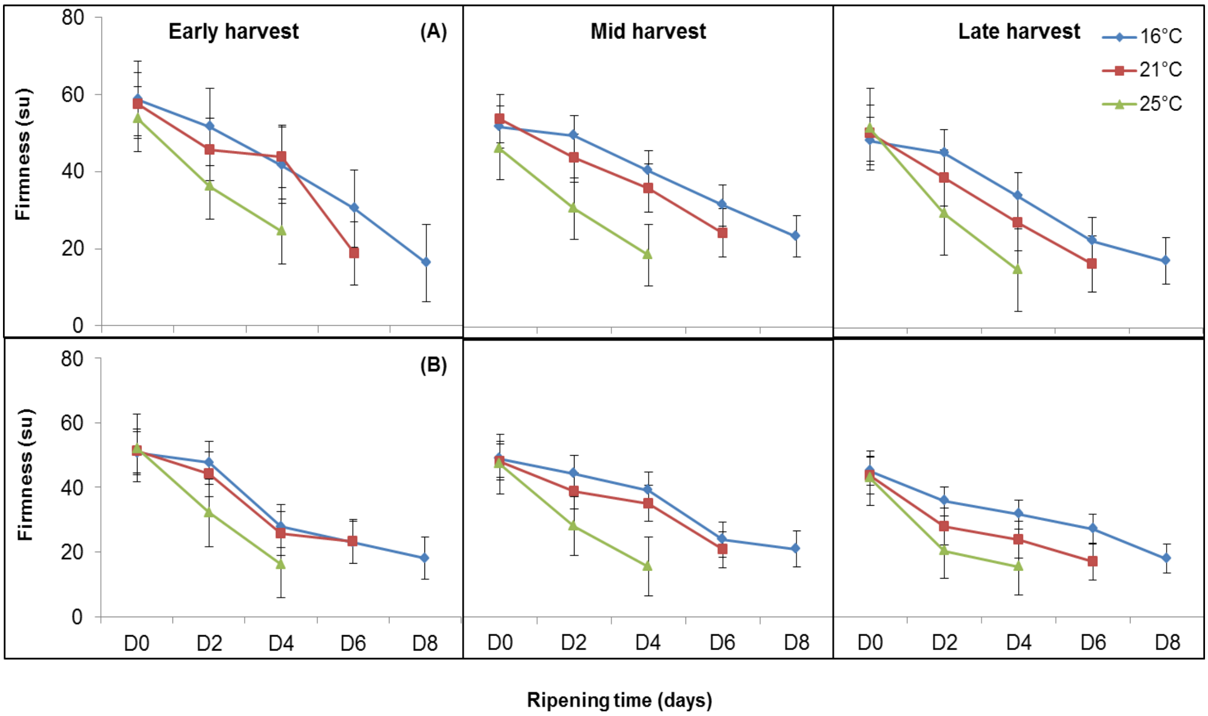

Figure 4.9: Firmness of fruit harvested at different maturities during ripening at different temperatures in 2013 (a) and 2014 (b)

In general, fruit weight loss showed an increasing trend for all ripening temperatures and harvest times for both harvest seasons. Through all harvest times, fruit ripened at 25°C had higher percentage of weight loss followed by 21 and 16°C (Figure 4.10A). Fruit lost 7, 9 , and 7 % early in the season; 5, 5, and 9 % in the mid harvesting time 6, 7 and 9 % of their initial weight late in the season when ripened at 16, 21 and 25°C, respectively. Fruit held at 25°C had higher fruit weight loss percentage at mid-harvest when compared with early and late harvest times.

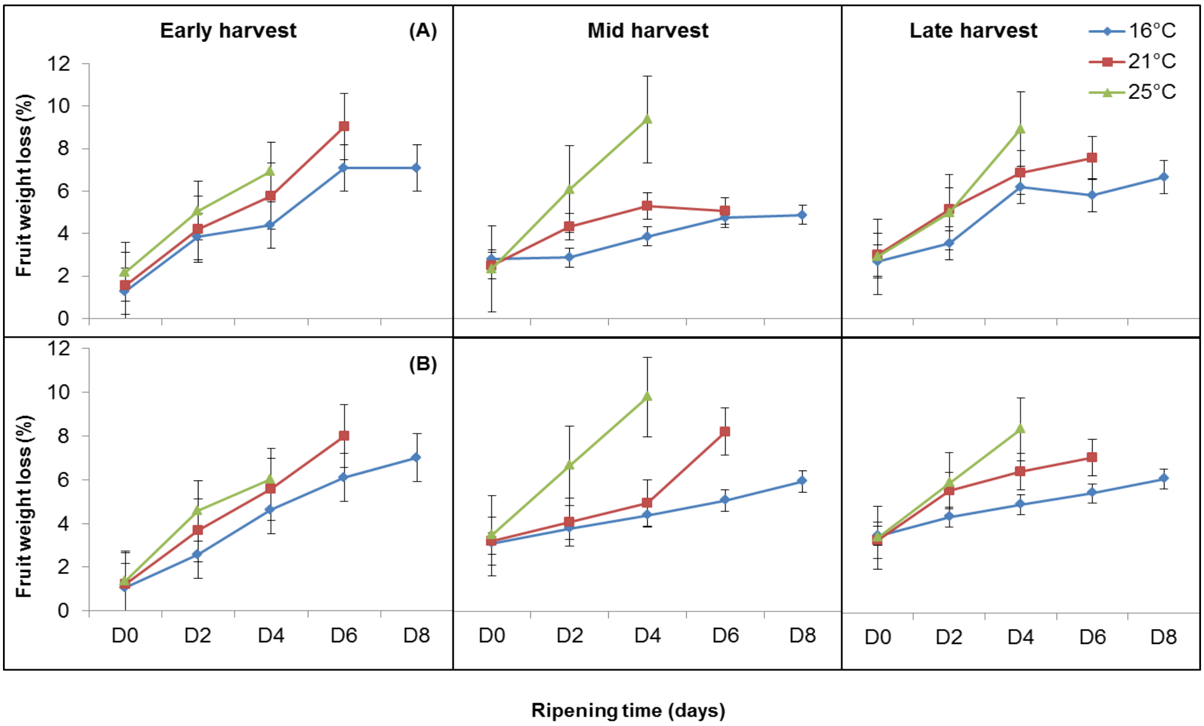

Figure 4.10: Weight loss percentage of fruit harvested at different maturities during ripening at different temperatures in 2013 (a) and 2014 (b)

4.4 Discussion

In this study, it was hypothesised that the interaction of harvest time and ripening temperature after cold storage would not affect post-harvest quality of 'Reed' avocado fruit. Fruit maturity and ripening temperature had a differential effect on quality of 'Reed' avocado fruit. Generally, good potential avocado fruit quality could only be achieved by reducing pre-harvest stress and controlling post-harvest environmental conditions (Bower and Cutting, 1987).

4.4.1 Effect of harvest time and ripening temperature on the post-harvest quality of 'Reed' avocado fruit

Delayed fruit harvest and higher ripening temperatures increased the susceptibility of 'Reed' avocado fruit to chilling injury (Figure 4.1). Results showed that late harvest fruit had the highest levels of chilling injury, while the lower levels were recorded in the mid-harvest time. This observation was consistent with the findings of Dixon *et al*., (2003), who showed fruit harvested early and late in the export season to have high levels of chilling injury when stored at 5°C for 28 days. However, these results contradict with the general consensus that early harvest fruit are more susceptible to chilling injury than late harvest fruit. Some recent evidence emerged from the work of Bower and Magwaza (2004), which suggested that late season fruit could also be more susceptible to chilling injury due to lower moisture content at time of fruit harvest. Ripening fruit at 25°C after cold storage increased the incidence of chilling injury compared with fruit ripened at a lower temperature (16°C). Similar results were reported by Hopkirk *et al*. (1994) who found fruit ripened at higher temperatures (25 to 30°C) after cold storage to have high incidences and severity of chilling injury. According to Hedley *et al*. (1987), high fruit respiration and ethylene production were closely associated with the degree of chilling injury. Leungwalai and Beckles (2013) elucidated that higher chilling injury incidences result from a burst of ethylene that occurs when cool stored fruits are reconditioned to higher temperature. Furthermore, chilling injury increased with ripening temperature during early and mid-harvest 'Reed' avocado fruit during 2014 season. However, ripening temperature showed to override maturity during the late harvest time with chilling injury decreasing as the ripening temperature increased.

Ripening fruit at 16°C reduced the incidence of anthracnose for early season fruit, while the disease was high on the late harvesting time under higher ripening temperatures. According to Sarananda *et al*. (2004), the rate of infection increase with increasing time of exposure to the inoculum. This was also observed on the current study, whereby, the incidence was high on late harvest fruit due to the increased time of exposure to inoculum while hanged on the trees. Nelson (2008) reported the incidence of anthracnose to increase as the season progress. Moreover, the development of anthracnose was almost completely inhibited at 5°C, while temperatures between 20 to 24°C enhanced the development of the disease. The low incidences recorded on early harvest fruit ripened at 16°C could be attributed to the small time of exposure to infection early in the season coupled with the low temperatures that supressed the development of the disease.

Lower ripening temperature induced vascular browning compared with fruit ripened at higher temperatures. This differed with findings of Hopkirk *et al.* (1994), who found vascular browning to be higher on 'Hass' fruit when ripened at 20 and 25°C after cold storage. According to Swarts (1984), vascular browning was a storage disorder associated with poor temperature management. Furthermore, vascular browning is intensified by the presence of astringency early in the season when fruit are still maturing. However, higher temperatures were reported to induce loss of astringency; and therefore, resulted in reduced vascular browning (Hedley, 1987). Fruit ripened at 25°C showed high incidences of vascular browning during the late harvest season. These results suggested that fruit maturity might have dominated the influence of ripening temperature on late season fruit. Cutting *et al.* (1992) also reported vascular browning increase with advanced fruit maturity.

Temperature had a significant effect on the incidence of stem-end rot. According to Kulkarni (2012), the establishment and progress of post-harvest fruit diseases is dependent upon the availability of suitable temperature. The incidence of stem-end rot was high on fruit ripened at 16°C and 21°C than fruit ripened at 25°C. This contradicts with results observed by Jadeja (2000), that stem-end rot developed optimally at 25°C. However, Dixon *et al*. (2003) stated that the rate of fruit ripening was dependent on temperature with higher temperatures favouring faster fruit ripening while reducing time required for disease development. Truter and Eksteen, (1987) reported that fruit that ripen more rapidly have low stem-end rot incidences. Fruit ripened at 16°C and 21°C required 8 and 6 days to ripen, respectively and showed higher incidences of stem-end rot than fruit ripened at and 25°C which ripened within 4 days after cold storage.

4.4.2 Effects of harvest time and ripening temperature on skin colour, firmness, respiration rate and fruit weight loss of 'Reed' avocado fruit during ripening

Skin lightness increased moderately throughout the ripening period regardless of ripening temperature during early and mid-harvest time; but, remained fairly stable for the late harvest fruit. These results were in agreement with those obtained by Chen *at al*. (2009) on 'Sharwil' cultivar who found L* values to slightly increase during ripening. According to Senthilkumar and Vijayakumar (2014), green skin cultivars retain their lightness during storage and ripening. The lower L* values early during ripening could be attributed to the loss of lightness as result of cold storage but decreased as the fruit defrost and gain their lightness when reconditioned to higher temperatures. Ripening temperature significantly influenced the L* values during the early harvest time in 2014 with higher L* values recorded on fruit ripened at 25°C than 21°C and 16°C. Similar behaviour of L* have been reported during ripening of 'Hass' avocado, whereby, fruit ripened at 15°C showed a loss in brightness than fruit ripened at 20°C (Maftoonaz and Ramaswamy, 2008). Furthermore, fruit that were held at 15°C required more days to ripen, were therefore, exposed to lower temperatures for longer period than fruit that were ripened at 21°C and 25°C, and resulted in loss of skin lightness (Abou-Aziz *et al.*, 2005).

Hue angle values decreased with ripening at each ripening temperature during the early harvest for both seasons. The results suggested that fruit harvested early in the season changed from a more green and took a darker shade green colour during ripening. Corresponding results were obtained by Abou-Aziz *et al.* (2005) who found green skin cultivar 'Fuerte' to decrease in h^0 values when fruit were ripened at 20°C after cold storage. However, fruit harvested during mid and late harvest times showed no change in h^0 values during ripening regardless of the ripening temperature.

Skin colour intensity (chroma (C*)) decreased gradually from day 0 to 2 of ripening regardless of ripening temperature and remained fairly stable until fruit were fully ripe. Some studies (Barreiro *et al*., 1997; Lee and Coates, 1999) have reported similar observations. In 2014, a similar Chroma pattern was obtained during early and late harvest times. However, mid harvest fruit showed a sharp rise in Chroma values at day 4 when ripened at 16°C.

Avocado fruit showed a respiratory pattern characteristic of climacteric fruits under each ripening temperature regime during both harvest seasons. Respiration rate was higher on fruit ripened at 25°C, followed by 21 and 16°C at all the harvest times. According to Jobling (2012), temperature has a direct effect on the respiration rate of a product. Maftoonazad and Ramaswamy (2008) reported that ripening 'Hass' avocados under lower temperatures reduces the respiration rate. Fruit ripened at 25 and 21°C reached a climacteric peak in day 2 of ripening than fruit ripened at 16°C which reached their climacteric peak in day 4. Higher and rapid respiration rates recorded at higher temperatures result from increased chemical reaction rate, mainly conversion of sugars and oxygen to carbon dioxide, water and heat during respiration (Brian and Fricke, 1996).

The response of fruit firmness to harvest time was consistent but depended on the different ripening temperatures. Fruit held at 25°C lost firmness more rapidly than fruit that were held at 16 and 21°C regardless of harvest time during both seasons. These results were in agreement with Swarts (1980), who showed that the avocado fruit firmness declined rapidly while held at higher temperatures. Fruit softening involves cell wall modifications by cell wall degrading enzymes (Fischer and Bennett, 1991), which function at a higher rate under higher temperatures, resulting in a rapid firmness loss. Moreover, fruit reached their eating ripe stage within 8, 6 and 4 days while held at 16, 21 and 25°C, respectively. The delay in ripening when fruit were held in 16°C was associated with reduction in loss of firmness compared with higher temperatures.

Fruit held at 25°C showed significantly greater weight loss than fruit ripened at lower temperatures. Similar results were reported by Ahmed *et al*. (2001), who found fruit weight loss per day to decrease with increasing temperature. The high weight losses at higher temperatures could be related to the higher respiration rate at higher temperatures (Lebibet *et al.*, 1995). According to Maguire *et al*. (2001), fruit weight loss occurred mainly as a result of transfer of water vapour from the fruit to atmosphere and to a lesser extent through carbon dioxide loss. High temperature resulted in higher respiration within the fruit, thus raising the vapour pressure at the surface and increasing transpiration (Gaffney *et al*., 1985).

4.5 Conclusion

The response of fruit to different ripening temperatures varied with harvest time, should therefore, be adjusted based on fruit maturity. Late harvest fruit were more sensitive to chilling injury when subjected to high ripening temperatures. Furthermore, high temperature was conducive for development of anthracnose disease, while the stem-end rot and vascular browning could be associated with the lower ripening temperatures. Fruit skin colour parameters were slightly reduced during ripening at all ripening temperatures, though this was not significant. Temperature was the main factor that influenced respiration rate, fruit firmness and fruit weight loss. High temperatures resulted in high and rapid respiration rate, reduced firmness and fruit weight loss.

CHAPTER 5
SUMMARY, RECOMMENDATIONS AND CONCLUSIONS

5.1 Introduction

Delayed fruit harvest on late maturing cultivars has a potential to extend the South African avocado harvest season. This would allow the industry to access the profitable late season market price for both export and local market. However, harvest time has potential to influence the response of avocado fruit to post-harvest treatments. Ripening temperature has an influence on the ripening behaviour of the fruit. This study was carried out to investigate the effects of delayed harvest on yield, fruit nutritional content and to evaluate the fruit quality response to varying ripening temperature regimes.

5.2 Summary

Fruit harvested two months after physiological maturity had low fruit P, K, Ca, Mg and Zn content compared with fruit harvested at physiological maturity and one month after reaching maturity (Chapter 3). However, the levels of these mineral elements were still higher than those recommended as the standard dietary allowance values. Fruit protein content decreased with increasing fruit maturity while starch and ash content increased with increasing fruit maturity. Fruit moisture content decreased with harvest time and was associated with a decrease in oil content.

The response of ‘Reed’ avocado fruit to post storage ripening temperatures was also influenced by fruit maturity (Chapter 4). Chilling injury increased with both increasing maturity and ripening temperature. Higher ripening temperatures induced higher incidence of anthracnose during early and mid-harvest, but were reduced on the late harvest time. Incidence of vascular browning and stem-end rot was high on fruit ripened at 16°C than 21 and 25°C. Fruit lightness increased steadily with ripening, while the fruit maintained the intensity of the colour at all ripening temperatures. Firmness, respiration and weight loss were high and rapid at 25°C resulting in a short fruit shelf-life, followed by 21 and 16°C regardless of the harvest time.

5.3 Recommendations

The data showed that:

- ‘Reed’ avocado fruit could be hanged on tree for one month post physiological maturity without compromising the subsequent yield and nutritional value of the fruit.
- Ripening temperatures should be adjusted for each harvest time to keep the post-harvest disorders and diseases under control.

Although data of two seasons for master degree purpose was collected during this study, further work on the following aspects would still be recommended:

- Due to the tree to tree variability and alternate bearing nature of avocado trees (Graham and Wolstenholme, 1991), repetition of this study for at least five seasons is recommended for conclusive results.
- Avocado cropping is dependent on adequate carbohydrate reserves (Graham and Wolstenholme, 1991), therefore an investigation on the effect of delayed harvest on tree starch cycle of ‘Reed’ cultivar is recommended.

5.4 Conclusions

Delaying fruit harvest showed no negative effect on tree productivity of ‘Reed’ avocado tree. Generally, delaying fruit harvest by two months improved the nutritional status of the avocado fruit. However, this was accompanied by a decrease in mineral elements of fruit. Moreover, harvest time influenced the response of the fruit to the post storage ripening temperature. Late harvested fruit were more sensitive to cold storage disorders. Higher ripening temperatures were highly conducive for development of post-harvest diseases during early harvest time, than on late harvested fruit. Ripening temperature was the main factor affecting the physico-chemical properties of fruit. Fruit firmness, respiration and weight loss were high and rapid under 25°C ripening temperature. However, delaying fruit harvest showed no effect on colour parameters of fruit.

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APPENDICES

Appendix 3.1 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit phosphorus content during 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	0.005	27.77		
Treatments	2	0.007	38.88	5.85	0.02
Error	8	0.005	27.77		
Total	14	0.018	100		

Appendix 3.2 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit phosphorus content during 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	3.427	36.87		
Treatments	2	2.893	31.10	3.89	0.06
Error	8	2.973	31.99		
Total	14	9.293	100		

Appendix 3.3 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit potassium content during 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	0.174	10.96		
Treatments	2	0.844	53.18	5.94	0.02
Error	8	0.568	35.79		
Total	14	1.587	100		

Appendix 3.4 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit potassium content during 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	0.631	53.11		
Treatments	2	0.354	29.79	7.01	0.01
Error	8	0.202	17.00		
Total	14	1.188	100		

Appendix 3.5 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit calcium content during 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	0.333	35.69		
Treatments	2	0.413	44.26	8.13	0.01
Error	8	0.187	20.04		
Total	14	0.933	100		

Appendix 3.6 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit calcium content during 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	0.000	0.00		
Treatments	2	0.022	84.61	20.33	0.00
Error	8	0.004	15.38		
Total	14	0.026	100		

Appendix 3.7 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit magnesium content during 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	0.001	4.76		
Treatments	2	0.018	85.74	48.24	0.00
Error	8	0.001	4.761		
Total	14	0.021	100		

Appendix 3.8 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit magnesium content during 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	0.400	54.57		
Treatments	2	0.333	45.42	6.67	0.01
Error	8	0.000	0.00		
Total	14	0.733	100		

Appendix 3.9 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit zinc content during 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	163.73	29.56		
Treatments	2	9.733	1.75	0.10	0.90
Error	8	380.26	68.67		
Total	14	553.73	100		

Appendix 3.10 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit zinc content during 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	82.267	21.15		
Treatments	2	280.93	72.23	43.67	0.00
Error	8	25.733	6.61		
Total	14	388.93	100		

Appendix 3.11 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit protein content during 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	2.489	11.85		
Treatments	2	9.877	47.01	4.57	0.04
Error	8	8.642	41.13		
Total	14	21.009	100		

Appendix 3.12 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit protein content during 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	8.332	37.15		
Treatments	2	10.817	48.23	13.20	0.00
Error	8	3.278	14.62		
Total	14	22.429	100		

Appendix 3.13 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit crude fibre content during 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	18.723	6.69		
Treatments	2	247.44	88.42	72.35	0.00
Error	8	13.681	4.88		
Total	14	279.84	100		

Appendix 3.14 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit crude fibre content during 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	1.665	0.58		
Treatments	2	286.35	93.95	68.71	0.00
Error	8	15.681	5.47		
Total	14	296.70	100		

Appendix 3.15 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit ash content during 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	1.417	56.14		
Treatments	2	0.628	24.88	5.25	0.03
Error	8	0.478	18.94		
Total	14	2.524	100		

Appendix 3.16 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit ash content during 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	1.403	58.00		
Treatments	2	0.730	30.18	10.20	0.00
Error	8	0.286	11.82		
Total	14	2.419	100		

Appendix 3.17 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit starch content during 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	0.624	3.47		
Treatments	2	11.737	65.36	8.39	0.01
Error	8	5.596	31.16		
Total	14	17.957	100		

Appendix 3.18 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit starch content during 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	1.128	9.45		
Treatments	2	9.402	78.76	26.74	0.00
Error	8	1.406	11.78		
Total	14	11.937	100		

Appendix 3.19 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit oil content during 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	13.68	0.81		
Treatments	2	1624.1	96.6	151.72	0.00
Error	8	42.82	2.54		
Total	14	1680.8	100		

Appendix 3.20 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit oil content during 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	9.45	0.46		
Treatments	2	1973.0	96.07	111.04	0.00
Error	8	71.08	3.46		
Total	14	2053.6	100		

Appendix 3.21 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit moisture content during 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	12.6	4		
Treatments	2	259.4	87	40.40	0.00
Error	8	25.6	8.5		
Total	14	297.8	100		

Appendix 3.22 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on fruit moisture content during 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	17.73	5.47		
Treatments	2	230.56	71.21	12.22	0.00
Error	8	75.46	23.16		
Total	14	323.73	100		

Appendix 3.23 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on yield during 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	14776.5	53.87		
Treatments	2	5630.4	20.53	3.21	0.09
Error	8	7022.0	25.60		
Total	14	27428.9	100		

Appendix 3.24 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on yield during 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	605.00	10.24		
Treatments	2	2873.32	48.64	4.73	0.04
Error	8	2428.45	41.11		
Total	14	5906.77	100		

Appendix 3.25 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on percentage of class 1 fruit during 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	1570.2	20.77		
Treatments	2	2261.2	29.91	2.43	0.15
Error	8	3728.1	49.32		
Total	14	7559.6	100		

Appendix 3.26 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on percentage of class 1 fruit during 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	4015.3	36.16		
Treatments	2	140.9	1.27	0.08	0.92
Error	8	6947.1	62.57		
Total	14	11103.3	100		

Appendix 3.27 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on percentage of class 2 fruit during 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	646.93	12.98		
Treatments	2	2835.7	56.91	7.56	0.07
Error	8	1500.2	30.11		
Total	14	4982.9	100		

Appendix 3.28 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on percentage of class 2 fruit during 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	2091.7	37.62		
Treatments	2	481.73	8.66	0.65	0.54
Error	8	2986.2	53.71		
Total	14	5559.7	100		

Appendix 3.29 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on percentage of class 3 fruit during 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	384.27	31.05		
Treatments	2	1.20	0.10	0.01	0.99
Error	8	852.13	68.85		
Total	14	1237.60	100		

Appendix 3.30 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on percentage of class 3 fruit during 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	442.40	33.27		
Treatments	2	14.93	1.12	0.07	0.93
Error	8	872.40	65.61		
Total	14	1329.73	100		

Appendix 3.31 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado on percentage of class 4 fruit during 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	13.733	18.63		
Treatments	2	10.533	14.29	2.90	0.13
Error	8	49.466	67.09		
Total	14	73.733	100		

Appendix 3.32 Analysis of variance (ANOVA) for three harvesting times of 'Reed' avocado percentage of class 4 fruit during 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	12.400	14.03		
Treatments	2	8.400	9.50	0.50	0.62
Error	8	67.600	76.47		
Total	14	88.400	100		

Appendix 4.1 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on chilling injury of 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	3210.5	8.76		
Harvest time (A)	2	5444.0	14.85	4.40	0.02
Ripening temperature (B)	2	6363.9	17.35	5.14	0.01
A x B	4	1850.8	5.05	0.75	0.56
Error	32	19800.9	54.00		
Total	44	36670.0	100		

Appendix 4.2 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on chilling injury of 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	824.3	2.33		
Harvest time (A)	2	444.4	1.26	0.51	0.60
Ripening temperature (B)	2	5483.0	15.52	6.24	0.00
A x B	4	14514.6	41.10	8.26	0.00
Error	32	14051.5	39.79		
Total	44	35317.8	100		

Appendix 4.3 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on vascular browning of 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	1378.1	2.77		
Harvest time (A)	2	4654.9	9.36	2.48	0.10
Ripening temperature (B)	2	3579.0	7.19	1.90	0.16
A x B	4	10016.8	20.14	2.66	0.05
Error	32	30086.4	60.51		
Total	44	49715.2	100		

Appendix 4.4 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on vascular browning of 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	1168.6	5.84		
Harvest time (A)	2	37.0	0.18	0.05	0.95
Ripening temperature (B)	2	5439.8	27.20	7.03	0.00
A x B	4	964.1	4.82	0.62	0.64
Error	32	12383.8	61.93		
Total	44	19993.4	100		

Appendix 4.5 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on anthracnose of 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	2688.1	7.22		
Harvest time (A)	2	6788.0	18.23	7.41	0.00
Ripening temperature (B)	2	2650.1	7.11	2.89	0.69
A x B	4	10455.0	28.08	5.71	0.0014
Error	32	14649.4	39.34		
Total	44	37230.6	100		

Appendix 4.6 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on anthracnose of 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	1758.2	6.94		
Harvest time (A)	2	729.3	2.88	0.91	0.4135
Ripening temperature (B)	2	4178.2	16.50	5.20	0.0111
A x B	4	5767.3	22.78	3.59	0.0158
Error	32	12853.5	50.77		
Total	44	25313.4	100		

Appendix 4.7 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on stem-end rot in 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	4359.8	27.10		
Harvest time (A)	2	641.6	3.98	1.31	0.28
Ripening temperature (B)	2	3050.5	18.96	6.21	0.00
A x B	4	175.0	1.08	0.18	0.94
Error	32	7858.5	48.85		
Total	44	16084.4	100		

Appendix 4.8 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on stem-end rot in 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	1394.9	8.12		
Harvest time (A)	2	455.5	2.65	0.55	0.58
Ripening temperature (B)	2	1271.8	7.40	1.54	0.23
A x B	4	800.6	4.66	0.48	0.74
Error	32	13244.5	77.14		
Total	44	17167.3	100		

Appendix 4.9 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on skin colour lightness (l*) values during ripening in 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	1527.1	0.30	1.21	
Harvest time (A)	2	14067.5	2.83	22.32	0.00
Ripening temperature (B)	2	6079.6	1.22	9.64	0.00
Ripening day (C)	4	78857.7	15.90	62.55	0.00
A x B	4	1623.0	0.32	1.29	0.27
A x C	8	72333.8	14.58	10.34	0.00
B x C	8	4908.1	0.98	28.69	0.00
A x B x C	16	55474.1	11.18	0.97	0.48
Error	176	260954.9	52.63		

Total	224	495825.8	100

Appendix 4.10 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on skin colour lightness (l*) values during ripening in 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	279.40	3.99	3.75	
Harvest time (A)	2	623.32	8.91	16.73	0.00
Ripening temperature (B)	2	177.80	2.54	4.77	0.01
Ripening day (C)	4	1569.86	22.46	21.07	0.00
A x B	4	32.14	0.45	0.43	0.78
A x C	8	1451.14	20.76	9.74	0.00
B x C	8	150.12	2.14	1.61	0.16
A x B x C	16	96.57	1.38	0.52	0.87
Error	176	2608.21	37.32		
Total	224	6988.56	100		

Appendix 4.11 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on hue angle (h^0) values during ripening in 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	7272.7	8.17	4.58	
Harvest time (A)	2	1966.0	8.21	2.48	0.08
Ripening temperature (B)	2	580.2	0.65	0.73	0.48
Ripening day (C)	4	9452.8	10.62	5.95	0.00
A x B	4	2027.7	2.27	1.28	0.28
A x C	8	11966.9	13.45	3.77	0.00
B x C	8	3632.6	4.08	1.83	0.11
A x B x C	16	2391.1	2.68	0.60	
Error	176	55587.3	62.49		
Total	224	88946.5	100		

Appendix 4.12 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on hue angle (h^0) values during ripening in 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	1497.1		1.17	
Harvest time (A)	2	307.0		0.48	0.62
Ripening temperature (B)	2	1874.7		2.93	0.05
Ripening day (C)	4	22234.9		17.35	0.00
A x B	4	3092.7		2.41	0.05
A x C	8	15784.6		6.16	0.00
B x C	8	1038.6		0.65	0.66
A x B x C	16	4047.9		1.26	0.25
Error	176	44857.9			
Total	224	79299.5	100		

Appendix 4.13 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on chroma (c*) values during ripening in 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	74.12	2.65	3.29	
Harvest time (A)	2	1488.20	53.28	132.27	0.00
Ripening temperature (B)	2	8.58	0.30	0.76	0.46
Ripening day (C)	4	306.97	10.99	13.64	0.00
A x B	4	12.27	0.43	0.55	0.70
A x C	8	38.01	1.36	0.84	0.56
B x C	8	24.18	0.86	0.86	0.51
A x B x C	16	53.18	1.90	0.95	0.49
Error	176	787.59	28.19		
Total	224	2793.1	100		

Appendix 4.14 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on chroma (c*) values during ripening in 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	90.26	2.68	2.25	
Harvest time (A)	2	1198.12	35.60	59.82	0.00
Ripening temperature (B)	2	0.07	0.00	0.00	0.99
Ripening day (C)	4	199.46	5.92	4.98	0.00
A x B	4	115.95	3.44	2.89	0.02
A x C	8	222.31	6.60	2.77	0.00
B x C	8	28.07	0.83	0.56	0.73
A x B x C	16	108.33	3.21	1.08	0.38
Error	176	1402.00	41.66		
Total	224	88946.5	100		

Appendix 4.15 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on respiration during ripening in 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	286065413.00	5.33	4.86	
Harvest time (A)	2	2001954.0	0.37	0.68	0.50
Ripening temperature (B)	2	7909858.0	1.47	2.69	0.07
Ripening day (C)	4	1361935630.0	25.39	23.13	0.00
A x B	4	20327602.0	3.79	3.45	0.01
A x C	8	99188917.0	18.49	8.42	0.00
B x C	8	7959942.0	1.48	1.08	0.37
A x B x C	16	27920853.0	5.20	1.90	0.05
Error	176	206119609.0	38.43		
Total	224	536227678	100		

Appendix 4.16 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on respiration during ripening in 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	13316888.0	2.34	2.39	
Harvest time (A)	2	22700670.0	3.99	8.16	0.01
Ripening temperature (B)	2	27449196.0	4.83	9.87	0.01
Ripening day (C)	4	159347084.0	28.04	28.64	0.01
A x B	4	14348099.0	2.52	2.58	0.04
A x C	8	108736280	19.13	9.77	0.00
B x C	8	10084609	1.77	1.45	0.21
A x B x C	16	17468369	3.07	1.26	0.26
Error	176	194758092.0	34.27		
Total	224	519361689	100		

Appendix 4.17 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on fruit firmness during ripening in 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	229.76	0.45	2.71	
Harvest time (A)	2	1042.41	2.04	24.55	0.00
Ripening temperature (B)	2	5664.16	11.12	133.38	0.00
Ripening day (C)	4	37448.33	73.54	440.92	0.00
A x B	4	249.70	0.49	2.94	0.02
A x C	8	974.13	1.91	5.73	0.00
B x C	8	2090.84	4.10	19.69	0.00
A x B x C	16	244.98	0.48	1.15	0.32
Error	176	2972.61	5.83		
Total	224	50916.92	100		

Appendix 4.18 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on fruit firmness during ripening in 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	81.94	0.21	1.79	
Harvest time (A)	2	1128.83	3.02	49.42	0.00
Ripening temperature (B)	2	3793.88	10.17	166.09	0.00
Ripening day (C)	4	27154.62	72.84	594.41	0.00
A x B	4	358.05	0.96	7.84	0.00
A x C	8	1307.17	3.50	14.31	0.00
B x C	8	1650.22	4.42	28.90	0.00
A x B x C	16	202.34	0.52	1.77	0.07
Error	176	1598.92	4.28		
Total	224	37275.97	100		

Appendix 4.19 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on fruit weight loss during ripening in 2013

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	6.85	6.62	1.89	
Harvest time (A)	2	29.67	2.71	16.35	0.00
Ripening temperature (B)	2	112.44	10.28	61.94	0.00
Ripening day (C)	4	647.47	59.24	178.34	0.00
A x B	4	14.78	1.35	4.07	0.04
A x C	8	82.30	7.53	11.33	0.00
B x C	8	50.64	4.63	11.15	0.00
A x B x C	16	21.67	1.98	2.42	0.01
Error	176	127.07	1.62		
Total	224	1092.8	100		

Appendix 4.20 Analysis of variance (ANOVA) for three harvesting times and ripening temperatures on fruit weight loss during ripening in 2014

Source	Df	SS	Percent	F-value	P≤ 0.05
Replication	4	6.73	0.65	1.64	
Harvest time (A)	2	41.69	4.06	20.28	0.00
Ripening temperature (B)	2	116.29	11.34	56.58	0.00
Ripening day (C)	4	559.21	54.54	136.04	0.00
A x B	4	20.19	1.96	4.91	0.00
A x C	8	66.48	6.48	8.09	0.00
B x C	8	54.48	5.31	10.60	0.00
A x B x C	16	16.31	1.59	1.59	0.11
Error	176	143.869	14.03		
Total	224	1025.24	100		

Appendix 5.1 Abstract submitted and presented as the oral at the 3rd all Africa Horticultural Congress, 7 -12 August 2016

Mafeo, T.P., Novela, P., Mathaba, N. and B.F. Nzanza. 2016. Delayed Fruit Harvest on Tree Productivity and Nutritional Composition of Reed Avocado (*Persia americana*). III All Africa Horticultural Congress.

Abstract

Delayed fruit harvest has not been studied on several minor avocado cultivars including Reed. Thus, a two year field study was conducted to evaluate the effect of delayed harvest on fruit mineral and proximate composition, yield and fruit size of Reed avocado. Fruit were harvested at three different maturity stages (early, mid and late) on a monthly interval. Phosphorus content decreased by 40 and 23% from early to late harvest time during 2013 and 2014 seasons, respectively. Similar decreasing patterns were observed for K, Ca and Mg content. Zinc content remained constant during 2013, however, mid-season fruit maintained higher zinc levels during 2014 season when compared to early and late season fruit. Protein content was constant on early and mid-season fruit but decreased during the late harvest. Moisture content decreased from 74.0 to 65.0% from early to late harvest during both seasons. Starch, oil and ash content were high during late harvest time while crude fiber fluctuated throughout the season. Yield dropped from 42 to 12 kg^{-1} tree from early to late harvest during 2013, while there was no treatment effect during 2014. Treatments had no effect on fruit size during both seasons. Generally, mineral content of avocado fruits deteriorates with maturity which might lead to poor fruit quality. However, delayed harvest has potential to improve nutritional value of fruit.

Keywords: Delayed fruit harvest, Fruit yield, nutritional composition, Reed avocado

Appendix 5.2 Article submitted for publication at the International Journal of Agriculture and Biology, 2016

DELAYED HARVEST ON NUTRITION AND TREE PRODUCTIVITY OF AVOCADO

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Novelty statement

Avocado maturity studies have focused mainly on ‘Fuerte’ and ‘Hass’ as the major cultivars. These findings on Reed avocado as minor avocado cultivar are the first and showed that fruit can be hanged on tree for a month after reaching physiological maturity without compromising the nutritional value of the fruit and yield.

Abstract

Delayed fruit harvest has not been studied on several minor avocado cultivars including Reed. Thus, a two year field study was conducted to evaluate the effect of delayed harvest on fruit mineral and proximate composition, yield and fruit size of Reed avocado. Fruit were harvested at three different maturity stages (early, mid and late) on a monthly interval. Phosphorus content decreased by 40 and 23% from early to late harvest time during 2013 and 2014 seasons, respectively. Similar decreasing patterns were observed for K, Ca and Mg content. Zinc content remained constant during 2013, however, mid-season fruit maintained higher zinc levels during 2014 season when compared to early and late season fruit. Protein content was constant on early and mid-season fruit but decreased during the late harvest. Moisture content decreased from 74.0 to 65.0% from early to late harvest during both seasons. Starch, oil and ash content were high during late harvest time while crude fiber fluctuated throughout the season. Yield dropped from 42 to 12 kg^{-1} tree from early to late harvest during 2013, while there was no treatment effect during 2014. Treatments had no effect on fruit size during both seasons. Generally, mineral content of avocado fruits deteriorates with maturity which might lead to poor fruit quality. However, delayed harvest has potential to improve nutritional value of fruit.

Keywords: Delayed fruit harvest, Fruit yield, nutritional composition, Reed avocado

Introduction

Delay harvest or on-tree-fruit storage has been studied in detail only on 'Hass' and 'Fuerte' avocado cultivars (Kaizer and Wolstenholme, 1993; Whiley *et al.*, 1996). However, the strategy has not been studied on several minor and new avocado cultivars including 'Reed' avocado. Delayed fruit harvest (DFH) is an alternative strategy being considered to take advantage towards the October to January late prices on the local market. Furthermore, DFH of late maturing cultivars such as 'Reed' can also assist in increasing the market volume during the second half of the South African export season. The South African avocado harvesting season extends from March to September, with less to no fruits during October to February. This avocado fruits availability gap has long been identified (Graham and Wolstenholme, 1991) to increase the avocado industry export market competitiveness.

‘Reed’ is a late Guatemalan race cultivar which originates from California. It is an important minor cultivar in California and now produced in South Africa due to its late maturity, high flesh quality, and ability to crop under tropical conditions. Delayed harvest has the potential to bring a significant increase in fruit size (Kaizer and Wolstenholme, 1994) due to the continued cell division in avocado fruits when attached to the tree. On tree-fruit storage increased both yield and fruit size of ‘Hass’ from count 18 (211-235 g) to count 14 (266-305 g) when fruit were hanged from July to November (4 months) (Kaizer and Wolstenholme, 1993). Whiley *et al.* (1996) also found that fruit size and yield of ‘Fuerte’ was not reduced by late harvesting. However, Kassim *et al.* (2013) showed that late fruit growth beyond physiological maturity was accompanied by alterations in certain chemical properties of the fruit. Generally, it is important to understand these compositional changes, especially late in the growing season to ensure that fruit are harvested at their optimal mineral levels (Ferguson and Boyd, 2002). Therefore, the aim of this study was to assess effects of delayed harvest on mineral, proximate composition, fruit size and yield of late maturing Reed avocado fruit.

Methodology

Study sites, design, treatments and procedures

Avocado fruit were harvested from 10 year old Reed trees at ZZ2-Bertie van Zyl farms (Olyfberg) in Limpopo, South Africa (23°42’00”S, 29°54’00”E). The site is characterized by an annual rainfall of >1000 mm, with a maximum and minimum temperatures of 21 and 12°C, respectively, and soil being classified as clay soil (45% clay). Mature ‘Reed’ avocado fruit were harvested at three different maturities *viz,* early (commercial maturity in October), middle (November) and late (December) during 2013 and 2014 harvest seasons. Treatments were laid out in a randomized complete block design (RCBD), with five replications. Treatments were applied to a single tree plot and same trees were used during both seasons. Orchard cultural practices such as fertilization, irrigation, pruning, and pest and disease control followed the ZZ2 management standards. During each harvesting time, fruit samples were harvested and sent to Nvirotek Labs (North West Province, South Africa) for proximate and mineral analysis. Moisture content was performed at ZZ2 avocado commercial packhouse within the same day of harvest. During each harvest, 2 sets of 5 fruit samples were collected and the first set was used for proximate and mineral analysis while the second set was used for moisture content analysis. For both proximate and mineral analysis, fruit pulp was dried at 30°C until constant dry weight was obtained and dry pulp for determining other proximate parameters; then, moisture content was derived from these samples. Proximate analysis included the protein, crude fiber, ash, starch, oil and moisture content while the minerals analysed included phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), and zinc (Zn).

Protein content was determined using Dumas combustion method. Approximately 2.0 g of dry pulp sample was combusted at 900°C chamber in the presence of oxygen to release carbon dioxide, water and nitrogen. Thereafter, the gases were then passed over potassium hydroxide aqueous solution that absorbs the carbon dioxide and water. A column containing a thermal conductivity detector at the end was then used to separate the nitrogen from any residual carbon dioxide and water, and the remaining nitrogen content was measured. The measured signal from the thermal conductivity detector for the sample was converted into nitrogen content. Protein content was calculated as percentage crude protein = N X 6.25.

Crude fiber was determined using Van Soet (1990) method. Fats and carbohydrates were removed from a 5 g dry pulp sample which was then boiled in 150 ml of 1.25% H_2SO_4 solution for 30 minutes under reflux. The boiled sample was washed in several portions of hot water using a two-fold cloth to trap the particles. The sample was returned again to the flask and boiled in 15 ml of 1.25% NaOH for another 30 minutes under the same conditions on order to remove all remaining minerals and complex carbohydrates. Thereafter, the sample was transferred to an ashing dish and dried at 130°C to a constant weight. Afterwards, samples were cooled in a desiccator and weighed. Crude fiber was determined by the difference and calculated as percentage of the weight analyzed, thus: Crude fiber (%) = Wt. of residue + ash X 100/ Initial Wt. of sample.

To determine the ash content, dried pulp sample (5.0 g) was weighed into porcelain crucible and placed in a temperature controlled furnace at 600°C for 4 hours until proper ashing. The crucible was then cooled off in a desiccator and immediately weighed. Ash content was calculated as: $\text{Ash content} = \frac{\text{Wt. of dry sample}}{\text{Wt. of ash content}}$ X100

Starch content was determined using Ewers polarimetric method. Five grams of dried pulp was ground and mixed with 25 ml of diluted hydrochloric acid and immersed in a boiling water bath while shaking to prevent the formation of agglomerates. The samples were cooled by mixing with cold water to 20°C, filtrated and clarified with

Carrez solutions. Optical rotation was then measured in a 200 mm tube with a polarimeter (P1). The samples were extracted with 40% ethanol before being acidified, filtered and clarified again with hydrochloric acid. A second optical rotation (P2) was measured and starch content determined as the difference between the two measurements multiplied by the optical rotation of pure starch (185.4) using the formula below: Starch content (%) = 2000 (p1-p2)/185.4

Oil was extracted by mixing 200 g of the dried pulp sample with petroleum ether (100 ml) as a solvent. The sample was evaporated in a steam bath to remove most of the ether, thereafter, filtered through a dry, folded paper and dried at 100°C. The weight of oil extract was determined by the difference and calculated as percentage of the weight of sample analyzed, thus: $\text{Oil} = \frac{\text{Wt. of sample} - \text{Wt. of residue}}{\text{Wt. of sample}}.\ X100$

Moisture content was determined by longitudinally cutting ten fruits with the seed and seed coat adhering to the flesh removed. The peeled halves were triturated with a kitchen grater consisting of five cutters per square centimeter to get approximately 1 mm thick shreds. Ten (10) grams of the grated avocado pulp sample was thoroughly mixed and weighed in a tarred petri-dish. Moisture content was determined by drying the fresh pulp sample in an oven (400L EcoTherm Digital, Japan) at 30°C for 48 hours. The petri-dish was cooled off in a desiccator and dry weight was measured. The moisture content was calculated by difference using the formula:

$$\text{Moisture \%} = \frac{\text{Wt. of fresh sample}}{\text{Wt. of dry sample}}.\ X100$$

Minerals were determined by dry ashing the dry pulp sample at 65°C to constant weight. The ash was dissolved in a volumetric flask using distilled and deionized water with a few drops of concentrated hydrochloric acid. Potassium was determined with a 410 dual channel flame photometer using NaCI and KCI to prepare the standards, while other minerals (Ca, Mg, P and Zn) were determined with an Atomic Absorption Spectro-photometer (AAS, 210VGB, Melbourne, Australia). Zinc was reported in mg/kg while other elements were reported in percentages.

Trees were harvested individually and yield was recorded during each harvesting time (kg tree^{-1}). Fruit were sent through the commercial pack line to determine fruit size distribution.

Table 1: Fruit size distribution codes (NDA Gazette, 2013)

Class	Fruit weight
Class 1	576 - 462 g
Class 2	461 - 247 g
Class 3	246 - 174 g
Class 4	173 - 80 g

Data analysis

Data was subjected to analysis of variance (ANOVA) through Statistix 10.0 (2012). Sum of squares were partitioned, while treatment mean separation was achieved using Least Significant Difference (LSD) test at 5% probability level.

Results

Fruit nutritional content

Harvesting time significantly affected ($P \leq 0.05$) fruit Potassium (K), Calcium (Ca) and Magnesium (Mg) content on both harvesting seasons (Table 2). However, there was no significant difference on Zinc (Zn) content during 2013 in contrast to 2014 results where the treatments differed significantly. Similarly, harvesting time affected phosphorus (P) content during 2013 while it had no treatment effect during 2014 season. The concentration of P and K remained constant during early and mid-harvesting times, but declined at the late harvest during 2013 season. However, a different trend was observed during 2014 whereby the levels of both mineral elements fluctuated with different harvest time. Phosphorus and K were low for the early season harvest time, peaked at mid-harvest, and slightly, declined for the late harvest time. Phosphorus content ranged from 0.15 to 0.09 and 0.13 to 0.10% while K content ranged from 2.09 to 1.54 and 1.86 to 1.48% for both 2013 and 2014 seasons, respectively (Table 2). Calcium content declined consistently with harvesting time during 2013. The concentration of Ca declined from a high of 0.05 to 0.02% from early to late- harvest, respectively. However, during 2014 mid-harvest time, Ca content (0.12%) significantly increased when compared with the early (0.05%) and late (0.02%) harvest times. During 2013 season, magnesium content decreased steadily with harvesting time from 0.14 to 0.06% from early to late harvest times, respectively. However, during the 2014 harvesting season, Mg levels remained constant during early and mid-harvest and decreased during late harvesting time. Zinc content remained constant over the three harvesting times during 2013. However, mid- harvesting time had high zinc content while early and late harvest times didn't differ significantly during 2014.

Table 2: Effects of delayed fruit harvest on mineral composition of Reed avocado

Season	Harvest time	Phosphorus (%)	Potassium (%)	Calcium (%)	Magnesium (%)	Zinc (mg/kg)
	Early	0.14a	1.98a	0.05a	0.14a	28.00a
	Mid	0.15a	2.09a	0.02b	0.11b	29.60a
	Late	0.09b	1.54b	0.03b	0.06c	29.80a
P-value at 0.05		0.04*	0.02*	0.02*	0.01*	0.00*
2014	Early	0.10b	1.48b	0.05b	0.10a	13.20b
	Mid	0.13a	1.86a	0.12a	0.10a	23.00a
	Late	0.12ab	1.71ab	0.26b	0.9b	14.60b
P-value at 0.05		0.00*	0.06^{ns}	0.01*	0.00*	0.01*

Delayed fruit harvest significantly affected the entire proximate analysis variables measured during both harvesting seasons (P≤0.05) (Table 3). Protein content was constant during early- and mid-harvest time, but decreased during late-harvest from high of 5.34 to 3.58% and 5.43 to 3.56% during 2013 and 2014 seasons, respectively. While, moisture content decreased with increasing fruit maturity during both seasons. The moisture content dropped from 74.0 to 65.0% and 75.6 to 66.0% from early- to late-harvesting times during both seasons, respectively. Furthermore, there was a progressive significant increase in oil and starch content as the fruit hanged longer on the trees. Oil content increased steadily from 40 to 64% and maintained the same trend for both seasons. However, starch content increased from early-harvest to mid-harvest, and thereafter, levelled-off through late harvest time. While, starch content increased from 3.2 to 5.5 % and 3.1 to 4.59% from the early to mid-harvest during 2013 and 2014 seasons, respectively. High crude fibre content was recorded on early and late harvest times while mid-harvest time resulted in the least fibre content in both seasons. Ash content was low during early harvest, rising sharply during mid-harvest time and thereafter decreased during late harvest time.

Table 3: Effects of delayed fruit harvest on mineral composition of Reed avocado

Season	Harvest time	Protein (%)	Crude fiber (%)	Ash (%)	Starch (%)	Oil (%)	Moisture (%)
	Early	5.26a	28.62a	3.22b	3.24b	39.96c	74.88a
	Mid	5.34a	21.02b	3.72a	5.02a	44.24b	72.32a
	Late	3.58b	30.38a	3.50ab	5.20a	63.86a	65.06b
P-value at 0.05		0.04*	0.00*	0.03*	0.01*	0.00*	0.00*
2014	Early	5.43a	29.11a	3.12b	3.10b	38.19c	75.60a
	Mid	5.28a	20.73b	3.65a	4.92a	43.51b	71.00b
	Late	3.56b	30.23a	3.45a	4.59a	64.74a	66.00c
P-value at 0.05		0.00*	0.00*	0.00*	0.00*	0.00*	0.00*

Values with same letters were not significantly different at 0.5 probability level

Treatments showed no significant difference ($P \leq 0.05$) (Table 4) in yield during 2013, while there was significant effect during 2014. Yield dropped from 42.21 to 12.26 kg^{-1} tree from early to late harvesting time while higher yields were observed during early and late harvest times in 2014 season. Treatments showed no significant difference on class 1, 3 and 4 fruit during both 2013 and 2014 harvesting season ($P \leq 0.05$) (Table 4), while the class 2 fruits differed significantly during 2013. The percentage of class 1, 3 and 4 fruit remained the same during both seasons while the percentage of class 2 fruit decreased with harvesting time during 2013 season. The percentage of class 2 fruit dropped from 62.6 to 31.8% from early to late harvesting time.

Table 4: Effects of delayed fruit harvest on yield and fruit size distribution of Reed avocado

Season	Harvest time	Yield (kg/tree)	Class 1 (%)	Class 2 (%)	Class 3 (%)	Class 4 (%)
2013	Early	42.21ab	20.60a	62.60a	15.40a	1.20a
	Mid	59.11a	42.00a	35.40b	16.00a	2.60a
	late	12.26b	49.60a	31.80b	15.40a	0.60a
P-value at 0.05		0.09^{ns}	0.15^{ns}	0.01*	0.99^{ns}	0.46^{ns}

2014	Early	54.68a	35.60a	50.20a	11.20a	1.20a
	Mid	24.16b	43.00a	36.40a	13.60a	3.00a
	late	52.20b	40.40a	42.00a	12.80a	2.40a
P-value at 0.05		0.4*	0.93^{ns}	0.54^{ns}	0.92^{ns}	0.62^{ns}

Values with same letters were not significantly different at 0.5 probability level

Discussion

Our study hypothesised that delaying fruit harvest by either one or two months after commercial maturity under South African growing conditions would not affect the nutritional value and yield of ‘Reed’ cultivar. However, significant differences in mineral content, proximate composition and yield were found between the various harvesting times.

Phosphorus (P) and potassium (K) levels were significantly low for the late harvest (December) during both seasons. Such results were in agreement with findings of Rosecrance *et al.* (2012), whereby early harvested ‘Hass’ fruit were associated with higher levels of P and K, but decreased later as the season progressed The decline in both P and K was attributed to their uptake pattern during fruit development. The movement and accumulation of P and K into the fruit is closely related to fruit dry matter, and a reduction or possibly ceasing the accumulation after reaching maturity (Clark and Smith, 1988). These results suggested that delaying fruit harvest results in lower mineral levels due to the dilution of minerals during the increasing fruit growth. Though the levels of both P and K were low at 0.09% and 1.54% during 2013 and 2014, respectively, the values were still higher than the standard mineral recommended dietary allowance values of 0.04% and 1% P and K (Ortega *et al.*, 2013)

Fruit calcium (Ca) content decreased with advancement in fruit maturity. Results were in line with the findings of Cutting *et al*. (1992), who showed calcium levels on ‘Fuerte’ to decline from a high of 0.018% one month after first harvest to a low of 0.012% at the latest harvest date, which was 33% decrease. However, the current study recorded a higher resultant Ca reduction of 60% between the early and late harvesting time in both seasons. Calcium is an immobile element and its accumulation in the fruit slows down with increasing fruit maturity (Knee, 2002). Furthermore, high available soil K and Mg often result in lower fruit Ca through a reduction in

Ca root uptake (Coates *et al.*, 1997). The importance of Ca as a determinant of post-harvest fruit quality has been recognised for many years (Koen *et al.*, 1990). According to Thorp *et al*. (2010), harvesting fruits too late resulted in fruit that were quick to ripen, more susceptible to rots and physiological damage, especially with low Ca levels. In this study, Ca levels were observed were higher than the established threshold Ca content of 25 to 28 mg/100g dw for South African avocados and was found to practically have no vascular or flesh browning (Cutting and Bower, 1992).

The accumulation of Magnesium (Mg) decreased systematically during each harvest time. Similarly, Cutting *et al*. (1992), reported a decrease in Mg content from a high of 0.08% at the first harvest date to a low of 0.06% on the late harvest date. The accumulation of Mg in fruits is xylem based and highly active during the early phase of fruit development. Later in the season, with an increased fruit dry matter accumulation, phloem tends to provide the predominant flow, resulting in low or no supply of xylem based elements to the fruits (Knee, 2002). Zinc (Zn) content remained constant throughout the season during 2013 season, but a significant increase was recorded during mid-harvesting time during 2014. Interestingly, this significant increase in Zn content coincided with an increase in Ca content. In fruit trees, Zn increases the Ca levels by reducing levels of oxalate bound Ca and improve the strength of water retention by the fruit tissues (Shear, 1980).

Protein content remained constant during early and mid harvests but declined during the late harvest. On contrary, Blakey *et al*. (2009); Graham and Wolstenholme (1991) found 'Hass' fruit protein content to remain constant throughout the season despite the increasing fruit maturity. Reduction in protein content observed in this study could be attributed to protein indirect calculations from nitrogen concentration using a conversion constant of 6.25. Therefore, the differences in protein concentration might merely reflect differences in nitrogen content in the fruit which was also low on the late harvested fruits (data not included).

In relation to crude fibre, levels ranged from 20 to 30% with the lower levels recorded during the mid-harvesting time while there was no significant difference between the early and late harvest time. Avocado have high fibre content among other fruits, comprising 75% insoluble and 25% soluble fibre (Oluwoles *et al*., 2013). The observed increase in fruit fibre content during the late harvesting time might have been due to the increased biosynthesis and accumulation of fibre components (pectin, cellulose and hemicelluloses) as the fruits hanged longer on the trees (Msogoya *et al*., 2004). Similar trend was also reported on citrus cv. Novel orange which showed an increase in dietary fibre contents with fruit maturity. (Xiuli and Guanlun, 2007).

Ash content didn't differ significantly between the three harvests on both seasons. However, the ash content was three times higher when compared with those reported for Fuerte (±1%) (Pearson, 1975). Starch content increased with increasing fruit maturity. These trends in starch were consistent with the work of Burdon *et al*. (2010) who reported starch content to increase steadily as the season progressed. According to Whiley *et al*. (1988), while fruit are attached to the tree and increasing in maturity; simultaneously, the tree continues with its normal phenological cycle. This implies periods of strong competition for limited carbohydrates during new vegetative and flowering growth flushes, intensifying during late maturity when flowering and fruit set occurs for the subsequent season. However, Burdon *et al*. (2010) elucidated that fruit starch content is not simply reduced by direct competition with vegetative or reproductive growth but more related to the overall availability of carbohydrates within the tree. The increasing fruit starch content observed in this study suggested that the trees had high carbohydrates reserves to maintain the normal tree cycle while carrying the hanging fruit from the previous season.

The high oil content observed in this study suggests that 'Reed' could be an important cultivar for oil production. Oil ranged from 39% to 64% from early to late harvesting times. According to Yousef *et al*. (2010), oil content increases in the mesocarp after fruit set and could be correlated with the fruit age. Oil increased by 4.28% from early to mid-harvest and 19.62% from mid to late harvest. These trends were in line with findings of Hofman *et al*. (2000). Blakey *et al*. (2012) reported that the increase in oil content tend to be higher between the mid and late harvest when compared with early and mid-harvest. This was also in agreement with Lizana *et al*. (1992) who found an increase in oil to be more pronounced late in the season than early to mid-season. Mostert *et al*. (2007) suggested that increased oil content was due to increased cell wall degradation, resulting in an improved accumulation of oil on the fruit mesocarp.

Moisture content ranged from 74.8 to 65.0% and 75.0 to 66.0% between the early and late harvest during 2013 and 2014. These results lend further support to the reduction in moisture content observed in previous studies (Osuna-Garcia *et al*., 2010; Yousef and Hasseine, 2010). Moisture content is the preferred indicator of maturity in South African avocado industry with the recommended moisture content in the range of 75 to 69% depending on the cultivar (Mans *et al*., 1995). Currently, 'Reed' cultivar is classified as "other cultivars" under the South African standards and requirements for control of the export of avocados with a specified limit of 75% moisture content at harvest (Government Gazette, 2013). The observed reduction in moisture content was due to the increase in oil content as the fruit maturity advanced. Moisture content has an inverse relationship with oil content (Kruger *et al*., 1995) and as the oil content increase with maturity, moisture content decrease by the same amount.

Delaying fruit harvest by two months reduced the fruit yield during 2013 season. Yield showed a reduction of 30% from the early to late harvest times. However, Newett *et al.* (2002) reported that 'Reed' fruit could be hanged on tree for one month longer than 'Hass' cultivar under California avocado growing conditions without affecting the subsequent crop. The variation observed in this study could be attributed to tree to tree variation and nature of the seedling trees used in this study. However, there was no effect of delaying harvest on the subsequent crop. The yield didn't differ significantly across the harvesting times during the second season with an average of 55 kg $tree^{-1}$. Kaiser and Wolstenholme (1993) found no significant differences on yield of Hass cultivar during the first two seasons but significant decline was obtained in the third season. This justified further investigation into the potential long term effects of delaying fruit harvest consecutively on tree productivity and possibility of alternate bearing.

Class 1 and 2 fruit dominated during both seasons with each class accounting an average of 37 and 39, 43 and 42% during 2013 and 2014, respectively. However, the percentage of class 2 fruit was significantly lower during the mid and late harvesting times when compared with early harvesting time during 2013 season. Generally, avocado fruit size was highly variable. This was due to that fruits set over an extended period resulting in mixed fruit size classes on the tree. However, in the study conducted by Graham and Wolstenholme, (1991), there was an increase in larger fruit, notably class 2 fruit, corresponding with decreases in the proportion of smaller fruit.

Conclusions

The present investigation revealed that the mineral and nutritional value of avocado fruit varies with fruit maturity. The fruit mineral content didn't differ significantly between the early and mid-season fruit but showed to undergo mineral degradation late in the season. Fruit harvested two months after reaching the minimum picking maturity had low fruit P, K, Ca, Mg and Zn content compared with fruit harvested early and mid-season. However, oil, starch and ash content increased steadily with increasing maturity, and resulted in fruit of high nutritional value during late season. Delaying fruit harvest reduced yield during the first season but showed no treatment effect on the following season. Furthermore, there was no treatment effect on fruit size distribution.

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