Investigating the Origin and Extent of Variation in Apple Fruit Quality

By

Andrew Frank William van Lingen

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Supervisor: Dr E.D. Louw
Co-supervisor: Prof W. J. Steyn

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DECLARATION

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This thesis is dedicated to my Lord and Saviour, Jesus Christ.

SUMMARY

Title: Investigation the origin and extent of variation in apple fruit quality

Developed countries with abundant food sources, have become more discerning of apple fruit quality. In addition to specific qualities that consumers may find desirable, variation in quality within a batch is perceived negatively, regardless if that fruit would have been acceptable when presented individually. Variation in apple fruit quality and maturity present a challenge for producers and complicate the postharvest handling and marketing of fruit. Understanding the origin and consequence of variation can lead the industry towards mitigating such issues.

The variance components: region, orchard, tree, canopy position (top vs bottom fruit, sun exposed vs shaded fruit) and bearing wood (shoots vs spurs) were, investigated. Variation in 'Golden Delicious' fruit quality and maturity was extensive at harvest and in some cases even greater after storage. The largest contributors to total variation in both seasons was orchard-to-orchard and tree-to-tree differences. Region contributed less to overall variation than expected and variation in starch conversion percentage (SC%) was prevalent in both Elgin and the Koue Bokkeveld with the bottom branches having a higher SC% than top branches. Position within a tree interacting with light exposure influenced fruit firmness with sun exposed fruit and fruit from the top branches having a higher firmness. The effect persisted through to fruit after storage showing greater differences between the positions. Fruit peel lightness values and hue angles showed moderate and strong correlations from at harvest to after storage but correlated poorly with other variables.

Relative bloom date (RBD) was investigated as an additional source of variation but accounted for less of the variation than anticipated. RBD had a stronger influence on maturity in the milder winter region than the colder winter region, with early clusters producing fruit with the higher starch conversion percentage than late cluster in Elgin, while there was no difference in the Koue Bokkeveld. The effect of RBD on fruit mass was commercially consequential in both regions with late blooming clusters producing the smallest fruit.

Flower quality was also evaluated in warm and cold site, with number of flowers per cluster, receptacle diameter, pedicel length and dry weight of flower clusters being measured across bloom time (early and late), canopy position (top and bottom) and spur age (2-year-old spurs, 3-year-old spurs, old spurs, and shoots). Greater dry weight was observed for early clusters in the colder site than for late clusters, but there was no significant difference in the warmer site. The effect of bearing wood on flower cluster dry mass showed that flowers on shoots in the warmer site had the greatest dry mass while those on old spurs in the colder site had the highest dry mass. This translated through to fruit mass at harvest, where the heaviest fruit in the warmer site were found on shoots, and old spurs produced the heaviest fruit in the colder site.

Results in this study confirm, that separate harvesting of the bottom and top halves of trees and sorting fruit on colour before storage, would increase the level of uniformity in batches of fruit. It should be noted though that colour sorting would not decrease the variance in maturity within batches of fruit but the fruit colour within a box or bag would be more consistent. By judicious pruning, summer pruning and branch removal, light distribution would be improved in existing orchards and possible result in less overall variation. The use of ethylene inhibitors such as aminoethoxyvinylglycine or 1- methyl-cyclopropene could also reduce variation in maturity at harvest. For the planting of new apple orchards, higher tree densities with thinner canopies are recommended and shade netting should be considered. Rest breaking programmes should be aimed at reducing the variance in flowering time between the bottom and top halves of trees.

OPSOMMING

Titel: Ondersoek na die oorsprong en omvang van variasie in appelvrugkwaliteit.

Ontwikkelde lande, met ŉ oorvloed voedselbronne, het meer oordeelkundig geword rakende die gehalte van appels. Benewens spesifieke eienskappe wat verbruikers wenslik mag vind, word variasie in kwaliteit binne 'n karton of sak appels as negatief ervaar, ongeag of dieselfde vrugte aanvaarbaar sou wees as dit individueel aangebied was. Variasie in appelvrugkwaliteit en rypheid bied 'n uitdaging vir produsente en bemoeilik die na-oes hantering en bemarking van vrugte. Die verstaan van die oorsprong en gevolge van hierdie variasie, kan die appelbedryf lei om hierdie probleme op te los.

Die variansie komponente: produksie streek, boord, boom, blaredak posisie (bo of onder en sonblootgestelde teenoor skadu vrugte) en drahout (lote of spore) is ondersoek. Variasie in ‘Golden Delicious’ vrugkwaliteit en rypheid was verrassend groot tydens oes en in sommige gevalle selfs groter na opberging. Die grootste bydraers tot die totale variasie in beide seisoene was boord-tot-boord en boom-tot-boom verskille. Produksie streek het minder bygedra tot algehele variasie as wat verwag was en variasie in styselomsettingspersentasie (SC%) was teenwoordig in beide Elgin en die Koue Bokkeveld, waar die onderste takke 'n hoër SC% as die boonste takke getoon het. Posisie binne 'n boom, wat met ligblootstelling in wisselwerking was, het 'n effek op die vrugfermheid gehad. Sonblootgestelde vrugte en vrugte van die boonste takke het 'n hoër fermheid gehad. Die effek het voortgeduur tydens opberging, waar groter verskille tussen die posisies aangetoon was. Ligheidswaardes van die skil en kleurhoeke het matige en sterk korrelasies getoon vanaf oes tot na opberging, maar was swak gekorreleerd met ander veranderlikes.

Relatiewe blomdatum (RBD) is ondersoek om die oorsprong van die variasie te probeer aanspreek, en daar is gevind dat dit minder tot die variasie bygedra het as wat verwag was. RBD het 'n sterker invloed op rypheid van vrugte in die warmer winterstreek as die koeler winterstreek gehad. In Elgin het vroeë trosse se vrugte ŉ hoër styselomsettingspersentasie as laat trosse getoon, terwyl daar geen verskille in die Koue Bokkeveld was nie. Die effek van RBD op vrugmassa was kommersieel betekenisvol in beide streke, waar laatbloeiende trosse die kleinste vrugte geproduseer het.

Blomgehalte was ook in beide ŉ kouer en ŉ warmer proefperseel geëvalueer, met aantal blomme per tros, deursnee van die blombodem, steellengte en droë gewig van blomtrosse bepaal tydens blomtyd (vroeg en laat), blaredakposisie (bo en onder) en drahoutouderdom (2-jarige spore, 3-jarige spore, ou spore, en lote). Vroeë trosse was swaarder in die kouer perseel as laat trosse, maar daar was geen betekenisvolle verskil in droë gewig in die warmer perseel nie. Die effek van drahout op die blomtrosse se droë massa het getoon dat blomme op lote in die warmer perseel die grootste droë massa gehad het terwyl dié op ou spore in die kouer perseel die hoogste droë massa

gehad het. Dit het deurgevoer na vrugmassa tydens oes, waar die swaarste vrugte in Elgin (warmer area) op lote gevind is, en ou spore die swaarste vrugte in die Koue Bokkeveld (kouer area) opgelewer het.

Resultate in hierdie studie bevestig dat die eenvormigheid in vrugte kwaliteit verhoog kan word indien die onderste en boonste helftes van bome apart geoes word, asook deur die sortering van vrugte volgens hul kleur voor opberging plaasvind. Daar moet egter gelet word dat kleursortering nie die variasie in rypheid binne ‘n besending vrugte sal verminder nie, maar eerder sal verseker dat die vrugkleur binne 'n boks of sak meer uniform sal wees. Deur oordeelkundige snoei, somersnoei en takverwydering sal ligverspreiding in bestaande boorde verbeter kan word, en moontlik minder algehele vrugvariasie tot gevolg hê. Die gebruik van etileen-inhibeerders soos aminoetoksivinielglisien of 1-metiel-siklopropeen kan ook variasie in rypheid tydens oes verminder. Vir die vestiging van nuwe appelboorde, word hoër boomdigthede met dunner blaredakke aanbeveel en skadunet kan ook oorweeg word. Rusbreekprogramme moet daarop gemik wees om die variasie in blomtyd tussen die onderste en boonste helftes van bome te verminder.

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GENERAL INTRODUCTION

The apple originates from Central Asia and the majority of commercial apple fruit production occurs in the latitude range of 25° to 52° (Forsline *et al.,* 2003; Palmer *et al.,* 2003). In 2019 a total 87 million tons of apples were produced worldwide ranking as the third largest fresh fruit commodity, after watermelons and bananas (FAOSTAT 2019). In 2020, apple production in South Africa totalled 991 252 tons and covered 25 272 hectares of arable land (Hortgro, 2020). The consistently abundant supply of fresh apples over the last few decades had made consumers more discerning of the quality of fresh apples (Richards, 2000).

Adapted for the Kazakhstani winters, which often plunge to -40°C, apple trees have evolved mechanisms to survive these harsh conditions (Horvath *et al*., 2003). Due to these adaptations, most production is at mid to high latitudes; however, with modified practices, production is possible in low latitudes (Jackson, 2000). In South Africa, most apple production takes place where winters are not sufficiently cold, resulting in delayed and reduced bud break that extends over a longer period than is typical of orchards from colder regions (Cook *et al.,* 2017). The resultant branching habit in South Africa is more basitonic resulting in a basal dominant canopy structure (Cook and Jacobs, 2000).

The visual perception of quality is the first hurdle to initiating a purchase (Jaeger *et al.,* 1998), and the metrics consumers focus on are the shape, size, colour, and condition of the fruit (Kays, 1999). The internal quality traits, viz. texture, sweetness, acidity and flavour profile, are central to drive consumers to repeatedly purchase apples (Daillant-Spinnler *et al.,* 1996; Jaeger *et al.,* 1998). Not only does the absolute quality of individual fruit matter to consumers, but also that each apple within a purchased batch (bag or box) be of uniform quality. Variability of fruit quality within a batch, be it size, colour, shape or another distinguishing characteristic, will negatively influence the consumer's perception of total fruit quality (Richards, 2000). In addition, variation in fruit maturity may also affect the marketability of fruit by increasing the risk of postharvest defects developing. For example, diffuse internal browning is a major postharvest defect of 'Pink Lady'™ apples and whole consignments are downgraded in the EU market if the disorder is present. Over maturity is a major risk factor in the development of diffuse internal browning (Crouch *et al.,* 2014).

Variation in fruit quality can be introduced in many ways. Besides the influence of genetics on quality, environmental conditions such as light and temperature are the strongest effectors of yield and quality (Palmer *et al.,* 1989). Due to large differences in light exposure and therefore in radiant heating, fruit position within the canopy also has a significant impact on fruit quality. Fouché *et al.* (2010) showed that partially shaded 'Granny Smith' apples were the darkest green in colour while fully exposed fruit were more subject to sunburn and red blush development. Hamadziripi *et al.* (2014) found that apples from the outer canopy were juicier, sweeter, and more flavoursome than apples from the inner canopy and were preferred by consumers in terms of taste. Aggelopoulou *et al.* (2010) showed significant variation in apple fruit quality was present within an orchard, chiefly due to the effect of topography and slope aspect on microclimate within the orchard. Bearing position also has an influence on fruit quality with apple fruit quality differing between shoots and spurs; this difference is genetically demined and differs between cultivars (Hirst and Ferree, 1995). In a study on 'Braeburn' and 'Granny Smith', Volz *et al.* (1993) showed that 2-year-old spurs yielded the largest fruit while 1-year-old laterals and spurs older than 3 years produced the smallest fruit. It is known that insufficient winter chill causes modified branching habits (Cook and Jacobs, 2000), and duly it may be possible for cultivars that typically bear the highest quality fruit on spurs to behave otherwise under these mild winter conditions.

The presence of variation in quality may already be present at blossom. Flower quality is important in determining fruit set and final fruit quality (Lauri *et al*., 1996). The best quality flowers are typically the first too open (Denne, 1963). Again, the effect of insufficient winter chill in causing delayed bud break may increase the variation in flower quality and in turn the final fruit quality and maturity.

The work presented in this thesis was intended as a descriptive study to quantify variation in fruit quality and maturity, both at harvest and after storage, as well as to determine the sources of said variation. Emphasis was placed on the effect of insufficient winter cold, with a mild winter production region being compared to a colder winter region. The preceding literature review examined the factors that influence fruit quality and maturity, emphasizing the physiological interaction between the tree and the environment (with specific focus on light and temperature).

Paper 1 determined the relative contribution of several factors to variation in 'Golden Delicious' apple fruit maturity and quality, namely: region, orchard, tree, and position (top and bottom; sun and shade; shoot and spur). It was also determined how these factors influenced specific internal and external fruit quality traits at harvest and after storage.

In Paper 2 the influence of relative bloom date on fruit quality and maturity of 'Golden Delicious' was evaluated in conjunction to the factors of Paper 1. It also determined the influence of region, orchard, tree, and position (top and bottom; sun and shade; shoot and spur) on relative blossom date.

The purpose of Paper 3 was to determine if the variation observed in the fruit was already present at bloom time, by evaluating flower quality across bloom time (early and late), position within a tree (top and bottom) and bearing position (shoots, 2-year-old spurs, 3-year-old spurs, and spurs older than 3 years). This study was conducted in both a mild winter region and a colder winter region.

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LITERATURE REVIEW: Variation in apple fruit quality and maturity: a South African perspective.

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1. Introduction

The apple is a temperate fruit crop originating from central Asia, specifically Kazakhstan (Forsline *et al*., 2003). It is adapted to cold winters and duly the majority of production is at mid to high latitudes, however with modified practices, production is possible in low latitudes (Jackson, 2000). Most commercial apple production occurs in the latitude range of 35 to 52° and culminated in 87 million tons of apples produced worldwide in 2019; this ranks apple fruit production as third largest fresh fruit commodity, after watermelons and bananas (Palmer *et al*., 2003; FAOSTAT 2019). In 2020, South African apple production totalled at 991 252 tons cultivated across 25 272 hectares (Hortgro, 2020).

Globally, apple producers have sustained high levels of production for decades. The abundant supply of apples has driven consumers to be more discerning of apple fruit quality (Richards, 2000). The demand for high quality and consistency, prompted producers to focus on the production of superior quality apples (Sansavini *et al*., 2004). Musacchi and Serra (2018) outlined the subjectivity of the term “fruit quality” as it is dependent on a consumer’s desire or expectation, but in essence, it can be divided into internal and external traits that collectively fulfil their expectations. External fruit traits such as shape, size, colour and visual appearance of the fruit, are contributing factors to perceived quality (Kays, 1999). Internal quality traits of the fruit, specifically texture, sweetness, acidity and flavour profile are important for consumer satisfaction (Daillant-Spinnler *et al*., 1996). For consumers, the appearance of fruit is central to driving initial purchases and upon consumption, the internal fruit quality encourages the consumer to make repeated purchases (Jaeger *et al*., 1998; Vanoli and Buccheri, 2012). It is seen that consumers presented with a batch of fruit are perceptive of, not only the quality, but also the uniformity of the fruit within the batch, and when consumers recognise a variation in size, colour, or some other distinguishing characteristic, their perception of the overall fruit quality within the batch is negatively influenced (Richards and Patterson, 2000; Richards 2000). This behaviour places a commercial importance on maintaining uniformity in both the internal and external quality traits of the fruit and drives many pre- and postharvest practices to align the produce specification to the consumers’ expectations. For the purpose of this review, more emphasis is placed on traits that are simple to measure and part of the South African export criteria, such as size, colour, firmness, and starch breakdown.

The environment has a profound effect on yield, quality, and variation in apple production (Palmer *et al*., 1989). Respiration rates and certain signals within a plant are temperature dependent, and because very little temperature control is possible in the open field, understanding how trees interact with its surrounding temperature is important (Lakso *et al*., 1999; Bastías and Corelli-Grappadelli, 2012). Light interception and distribution, with regard to availability and quality, influences fruit size, colour and dry matter content (Corelli-Grappadelli and Lakso, 2004). Conversely, light in excess causes photobleaching, and when coupled with elevated temperatures sunburn can occur (Hengari *et al*., 2014a). Much less is known about how the extremes of light and temperatures that fruit at different positions within the canopy affect fruit internal quality, storability and eating quality. Hamadziripi *et al.* (2014) showed that inner and outer canopy fruit differed in visual appeal for consumers, with inner canopy fruit from ‘Golden Delicious’ and ‘Granny Smith’ being preferred over the outer canopy fruit. Existing production is threatened by shifts in environmental conditions brought about by climate change. The predicted warming is believed to affect Mediterranean type climates in particular, and in the case of temperate fruit crops a warmer winter spells disaster for dormancy physiology (Baldocchi and Wong, 2006; Midgley and Lötze, 2008). Changes have already been noted in the length of the growing season, as well as in the relative fruit quality (DaMatta *et al*., 2010; Grab and Craparo, 2011). These changes can have a direct effect on final fruit size as detailed by Reginato *et al.* (2019) where they showed that an increased season length produced larger fruit, albeit second to the effect of adequate winter chilling. The review by Moretti *et al.* (2010) looked at the effect of climate change on postharvest quality of various fruit and vegetable crops, where most of the literature showed that under elevated carbon dioxide levels, apple fruit quality remained unchanged but for temperature extremes, fruit quality deteriorated due to sunburn and heat damage.

Climate change is slow-moving, and it would be more pertinent to look at the microclimate within orchards and trees, which plays a leading role when it comes to the variation in quality and storability of fruit (Sperling *et al*., 2017). Microclimate, in this review, mainly refers to the humidity and temperature conditions within an orchard or tree canopy. It can easily be manipulated by production practises such as tree architecture, pruning, and tree spacing in orchards (Dallabetta *et al*., 2012) making it a factor that can be managed using cultivar specific pre-harvest practices. Severe shading within a tree results in an imbalance of assimilates available to the fruit, and simpler tree structures have notably better light distribution (Corelli-Grappadelli and Lakso, 2004). Producers are thus tasked with maintaining uniform orchards of a desirable structure (Bastías and Corelli-Grappadelli, 2012). Differences in fruit quality within a location are observed due to the interaction of scion/rootstock combination and the environmental conditions (Corelli-Grappadelli and Lakso, 2004). Moreover, planting systems and tree architecture have an influence on the quality and uniformity of a batch of fruit. Furthermore, the genetic makeup of a cultivar is a major determinant driving fruit quality and growth habits such as increased red colour development in recent 'Fuji' mutations (Iglesias *et al*., 2012; Musacchi and Serra, 2018).

In this review, the factors influencing fruit quality and maturity are examined with emphasis placed on tree physiology and its interaction with the environment. Light and temperature are paramount in determining final fruit quality and these are looked at from the context of South African production regions, which are typified by suboptimal winter chilling (Cook *et al*., 2017).

2. Understanding fruit quality

In developed countries with abundant food sources, food quality has become increasingly important. Consumers are presented with a wide range of choice and are willing to pay a premium to obtain fruit of superior quality and increased food safety standards (Grunert, 2005). In recent years, research has been directed at improving fruit quality, not only to satisfy the stringent demands of customer, but also because higher quality fruit have greater storage potential. Defining fruit quality is challenging, because it is not hinged to any one specific trait, but a myriad of traits. The internal and external quality traits are also not additive, but only one notable deviation from the norm, would negatively affect the perceived quality (Harker *et al*., 2003). Aggregating these traits into acceptability would be fitting but makes quantifying specific quality near impossible. To complicate matters even further, what consumers deem acceptable is quite variable and fruit that would satisfy one, does not necessarily satisfy another (Jaeger *et al*., 1998). This is illustrated by a consumer preference study done by Idun *et al*. (2016) on nine different apple cultivars in South Africa, that found that apples with a higher acidity (sourness) were better tolerated by Indian and white consumers while black South Africans generally preferred the sweeter apples.

To understand how quality interacts with acceptability, it can be split into external and internal quality attributes. The external quality is what appeals visually to consumers. The appearance of the fruit is compared to what they perceive as desirable, which is typically fruit size, colour, a lack of defect or blemish and an overall healthy appearance. External quality is considered a first purchase driving trait, and if the fruit is visually appealing, the consumer is likely to purchase it; after purchase, the importance of internal quality is raised, and eating the fruit needs to satisfy the expectation that was built on its initial appearance (Vanoli and Buccheri, 2012). The satisfaction gained from consumption, based on the internal quality traits, such as sweetness, acidity and firmness, is paramount to driving consumers to repeat their initial purchases (Jaeger *et al*., 1998). The intended purpose of the fruit should also be considered when defining quality, as for the strawberry market, the ease of calyx removal is important for the processing industry, but is irrelevant to the fresh strawberry market (Tromp, 2005a). The distinction here between the fresh market and the processing industry (typically juice processing), is that the intended use of the product needs to be established so that relevant quality characteristics can be assessed. The processing industry for apples, where any visual characteristic is of no importance, is relatively insignificant compared to the fresh market, and only takes produce that does not satisfy the benchmark of quality for the fresh market, duly this review focuses on quality parameters relevant to the fresh market. The export of fresh apples is especially important for South African growers as the price for exported apples is often over twice the amount received for fruit marketed locally (Hortgro, 2020). In 2020, 45% of apple produced in south Africa were exported to the following destinations: Africa (27%), Far East and Asia (27%), United Kingdom (18%), Russia (9%), the Middle East (8%), Europe (7%), Indian Ocean Islands (2%), and North America (<1%) (Hortgro, 2020).

3. Apple fruit quality attributes

Fruit quality can be complex and is underpinned by individual characteristics. As discussed above, these characteristics often do not act alone in determining overall quality. The next section aims to discuss some of the more important external and internal traits relating to the South African apple export market as both external and internal fruit quality are important and can influence the monetary value of the fruit when packed to the specific quality standards associated with the export market.

3.1 External quality traits

Apple peel colour, which is easily noticed, is one of the most important attributes consumers consider when purchasing apples (Kays, 1999). The desirability of a colour is greatly influenced by cultivar, and to a lesser extent the origin of consumers (Harker *et al*., 2003). Bicolour apple fruit, such as ‘Cripps Pink’, ‘Braeburn’, ‘Gala’ and ‘Nicoter’ develop a partial pink/red colouring on the peel. The greater the degree of blush, the more desirable the fruit, which are then priced at a premium (Seppä *et al*., 2015; Hamadziripi *et al*., 2014). A red blush is not always favourable, as with ‘Granny Smith’ a red blush is regarded as a blemish, reducing its acceptance. Apple peel colour is predominantly determined by environmental conditions. Fortunately, these can be manipulated by production practices and is discussed further below. In the case of ‘Golden Delicious’, European countries tolerate a more yellow apple, where they associate it with higher sweetness, while African countries prefer a greener apple, which they associate with freshness (I. Adams 2021, personal communication, 22 November).

Fruit size is easily perceived by consumers and apples of “good size” fetch premium prices in the fresh market, but larger apples are not necessarily preferred. Different countries vary in their size preference. Most European countries, as well as Canada, prefer larger apples (Hampson and Quamme, 2000), while Scandinavian countries show preference for smaller apples (Redalen, 1987). For most cultivars larger apples fetch a higher price, but for South African ‘Gala’ types exported to the UK, smaller fruit can earn more money (I. Adams 2021, personal communication, 22 November). For ‘Golden Delicious’ larger sizes are in greater demand (I. Adams 2021, personal communication, 22 November).

Unlike size, fruit shape does not have any premium consideration, but when fruit shape deviates from what consumers perceive to be the norm, it is considered a negative quality. Apple fruit aspect ratio (length:diameter ratio) differs depending on the winter conditions the buds were exposed to and Petri and Leite (2003) showed that with warmer winters, apple fruit were shorter and had a smaller aspect ratio. The fruit aspect deviations they showed, are likely of little consequence to consumer preference as long as the fruit within a batch (carton or shelf) were of uniform dimensions and colour. Work on litchi by Rahman *et al*. (2010) and on watermelon by Sadrnia *et al.* (2007) both affirm that uniformity and a lack of visible blemishes being most important to consumers than specific dimensions.

Fruit peel blemishes (sunburn, bitter-pit, russeting, sunburn, etc.) significantly reduce acceptability of apple fruit when present (Harker *et al*., 2003). The prevalence of blemishes is chiefly due to environmental stresses such as nutrient deficiency or harsh climatic conditions, but because of genotypic variation, some cultivars are more sensitive to stresses, making defects more prevalent in certain cultivars (Ferguson *et al*., 1999).

The environment alters gene expression and influence tree growth and fruit production. In-depth reviews exist for the genetic component associated with quality and variation (Kumar *et al*., 2012), but is beyond the scope of this review. One of the most perceptible blemishes on ‘Golden Delicious’ is sunburn. Excess solar radiation and high fruit peel temperatures result in damage that ranges from bleaching to browning and dark necrotic lesions. Sunburn in apple fruit can cause significant economic loss, and duly has been well researched (Piskolczi, *et al*., 2004; Racsko and Schrader, 2012).

3.2 Internal quality traits

Looking at internal attributes, taste and texture are important, but their definition is a challenge. Upon consumption, taste is perceived in the mouth as sweet, salt, sour, and bitter; this is complimented by the aroma of volatile compounds in the fruit. The texture of the fruit contributes to "mouthfeel" and eating satisfaction. Sugar content and acidity can be measured as an indication of taste, with both their absolute amount and their ratio contributing to taste (Harker *et al*., 2002). A specific ratio cannot be pinned as ideal, and not only does it vary significantly between cultivars, but consumer preference also varies greatly. Here, consistency within a cultivar is the more important measure, as the variability of eating quality is perceived negatively (Richards and Patterson, 2000). 'Golden Delicious' is predominantly sweet with moderate acidity and relatively firm at harvest, but the firmness decreases markedly during storage (Abbott *et al.,* 2004).

Fruit firmness contributes to the texture of the fruit and is used as export criterion for quality from harvest, to storage, and right through to shelf display. E.g., the South African export minimum for firmness of 'Golden Delicious' fruit at harvest is 6.8 kg (Department of Agriculture, Forestry and Fisheries, 2013). Because of the extensive adoption of this measurement, attempts have been made to determine the relationship between measured firmness and sensory evaluation of texture by a panel (Harker *et al*., 2003). Wills *et al.* (1980) showed that fruit firmness correlated well with eating satisfaction.

Starch conversion percentage, as is the case with firmness, is used as an indication of fruit maturity. As the apple ripens and nears maturation, starch that has accumulated since development began is hydrolysed to sugar (Krotkoc and Helson, 1946). Starch conversion is measured by cutting an apple in two across the equator, and treating the exposed flesh with iodine solution. Where starch is still present, the flesh stains black while that parts where starch has broken down to sugars does not stain and remains pale. This has led to the development of starch maturity indices, but Blanpied and Silsby (1992) highlighted potential drawbacks in using this metric, where if trees bore an unusually heavy crop, fruit would not stain black even if they had not reached maturity yet. In South Africa, 'Golden Delicious' fruit need to have a starch conversion percentage above 15% to qualify for export (Department of Agriculture, Forestry and Fisheries, 2013). The starch conversion percentage and firmness are used in conjunction to correctly time harvest dates.

4. Factors influencing quality traits

There is a myriad of factors (genetic, agronomic and environmental) that contribute to the final fruit quality at harvest. Musacchi and Serra (2018) produced a thorough review on environmental (brown), genetic (red) and agronomic (blue) factors, as shown in Fig. 1. For the purpose of this review, the environmental factors of temperature and light will be discussed in more detail in the context of genetic homogeneity. Agronomic factors will be highlighted where they can be used to manipulate the tree's interaction with the environment.

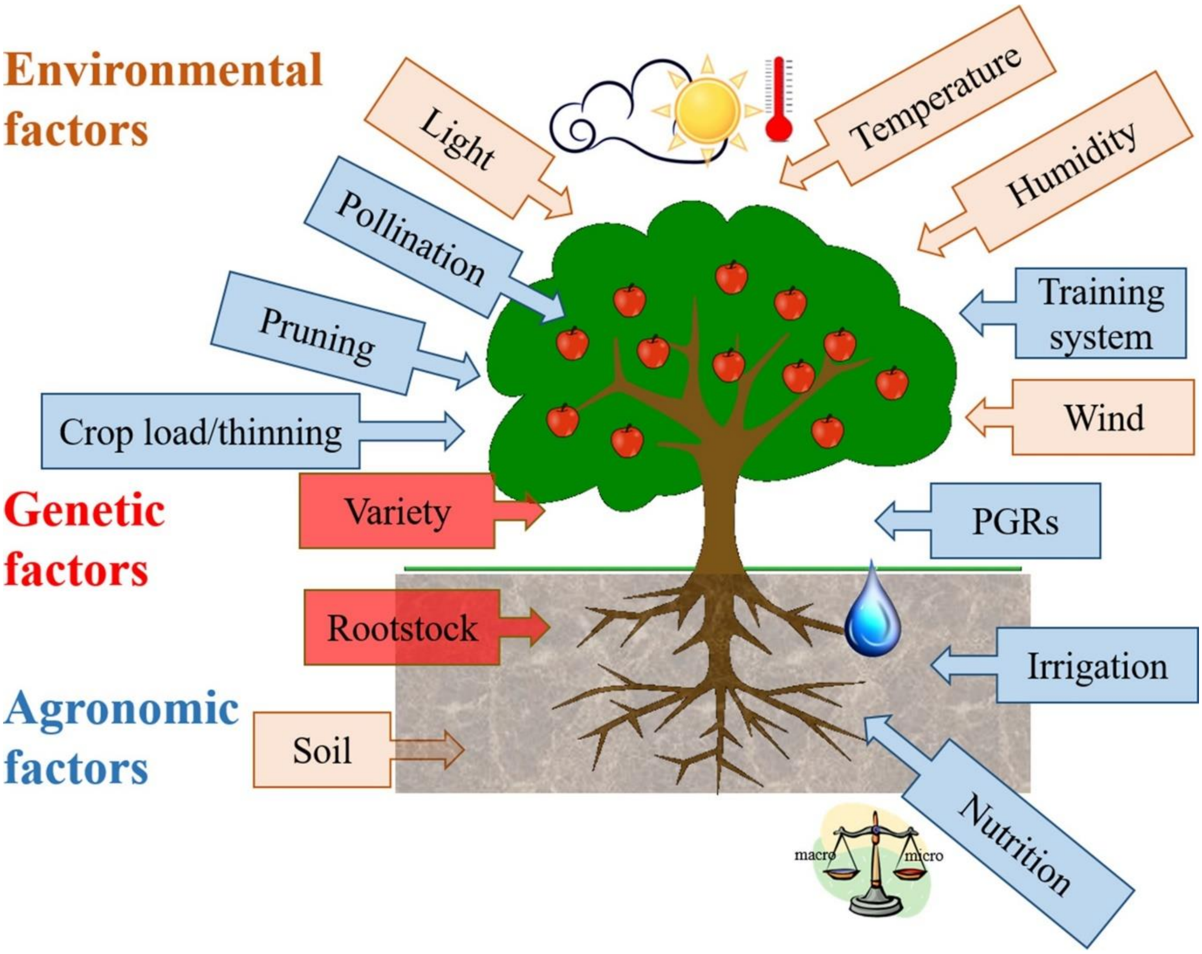

Figure 1. Factors affecting apple quality grouped in genetic (red), environmental (brown) and agronomic (blue) (with permission from Musacchi and Serra, 2018).
https://www.sciencedirect.com/journal/scientia-horticulturae

4.1. Temperature and climate

Fruit production needs to take place in a suitable environment, whose conditions influence the development of any plant organ (Jackson, 2000). Water and nutrient availability, soil quality, light availability and temperature regimes, all interact to produce varying growth habits, fruit quantity, quality and variation (Palmer *et al*., 1989). Apple trees can bear fruit from tropical climates to the furthest temperate zones, but what defines a specific environment as suitable, is not merely whether the tree will be able to bear fruit, but if production in said area would be of economic gain (Jackson, 2000). An environment similar to which the plant has adapted for, is expected to be the most feasible for production (Sansavini *et al*., 2004).

The apple is naturally adapted to cold winters, typical of temperate zones in higher latitudes. It evolved for the -40°C winters of Kazakhstan, from which most genic material originates (Forsline *et al*., 2003). Although naturally adapted for these conditions, it has not prevented the cultivation of apples in various suboptimal climates.

Faust (2000) describes four climatic categories, regarding their influence on the physiology of temperate fruit trees. Firstly, climates to which the temperate trees are well suited; typically, these have cool days and cool nights. Examples of areas with this climate type are Northern Europe and high-altitude regions surrounding the Alps. The relatively low temperatures result in low to moderate growth and dwarfing rootstocks are seen to be very effective at reducing vigour in said climate type.

The second climate is typified by winters cold enough to supplement the chilling requirement of the trees, but with a growing period, markedly warmer than the first category. This, sometimes arid environment, has cool nights and warm days, allowing for high assimilation of photosynthetic products during the day and little loss during the cool night. This climate type is considered highly productive, and is apparent in central Italy, Washington State, New Zealand and Tasmania.

The third climate type described has a short winter period of cooler to cold conditions, followed by a hot, dry summer. The amount of cold in winter is insufficient for the trees to overcome dormancy and the high summer temperatures have a detrimental effect on development of the trees. Here there are typically hot days and moderate night temperatures. This climate is typical of Mediterranean climates, and Mediterranean-type climates such as California, southern Australia, and central Chile. The Western Cape of South Africa would also form part of this category (Midgley and Lötze, 2008). This review will chiefly discuss scenarios within this climatic category.

A continuous cycle of warm days and warm nights typifies the fourth climate. Production areas within these subtropical climate types are found in Brazil, around the mid-Atlantic coast of the USA, southern France, and some parts of Japan and China (most apple production in Japan and China occur in the first climate type described). Under these conditions, a substantial portion of carbohydrates produced (40 to 50%) is lost at night due to respiration, making this the least productive of the four climates. Production in this climate is particularly challenging and the complete lack of winter cold necessitates different production methods, where after harvest, the trees are forced to regrow, which prevents the tree from entering a dormant state (Faust, 2000).

4.1.2 Dormant season temperatures

Temperature has a significant effect on apple cultivation (Palmer *et al*., 2003). A certain measure of cold is required in winter, because as a temperate crop, it has adapted to require a long winter dormancy period (Forsline *et al*., 2003). To fulfil the chilling requirement, the environment needs a sufficiently cold winter, which would allow for adequate bud break (Cook and Jacobs, 2000). Temperature also has an effect during the growing season, where warmer temperatures are favourable, but excessively hot temperatures can be detrimental to both fruit development and final fruit quality (Palmer *et al*., 2003). Temperature dictates the rate of physiological processes, for both the induction and the rate, of these reactions (Lakso *et al*., 1999).

Existing production is threatened by the environmental changes brought about by global warming. The predicted warming is believed to effect Mediterranean-type climates in particular, and in the case of temperate fruit crops, a warmer winter spells disaster for dormancy physiology (Baldocchi and Wong, 2006; Midgley and Lötze, 2008). In these climates, winters have become warmer, and the length of the growing season has behaved erratically, with some areas starting their season earlier and others markedly later (DaMatta *et al*., 2010; Grab and Craparo, 2011). Moretti *et* *al.* (2010) investigated the effect of climate change on the postharvest quality of various fruit and vegetable crops, but from their work, apples appear to be minimally affected (besides the obvious increase in sunburn or heat damage during the growing season). In a study on ‘Fuji’ apple quality spanning 40 years of climate data, Sugiura *et al*. (2013) showed that fruit firmness and acid concentration decreased as the climate warmed, while sugar levels increased.

Deciduous fruit trees cease growth in late summer or early autumn. Soon after growth cessation, a cold spell or physiological changes, cause the canopy to begin defoliation. The tree will remain leafless and in a dormant state during winter, and in spring, bud break occurs and growth resumes (Dennis, 1994). To standardise nomenclature, Lang (1987) defined the temporary suspension of visible growth, in plant structures containing a meristem, as dormancy. To further refine definitions, he presented three subclasses of dormancy, namely: endodormancy, paradormancy and ecodormancy. These subclasses were defined, according to the conditions of each, which can independently sustain dormancy.

Endodormancy: dormancy is sustained, because of a response to physiological signals from within the specifically affected structure.

Paradormancy: dormancy is sustained, because of an inhibitory signal from a structure, separate to which, is currently exhibiting a dormant state.

Ecodormancy: dormancy is sustained by a limitation in the growth environment of the specific structure, which inhibits development.

In summer and early autumn, vegetative growth stops, and buds on the trees enter a dormant state, which at this point is primarily upheld by paradormancy. This state results from apical dominance and correlative inhibition. At this time, bud development can be stimulated by the removal of the plant structures that are maintaining dormancy. Towards late autumn and early winter, the buds move into a deeper state of dormancy, endodormancy, where the inhibition of growth is maintained by signals within the buds themselves (Lang, 1987). During this state, even conditions favourable for bud development will not induce bud break. As winter progresses, periods of low temperatures induce physiological changes within the bud, reducing the extent to which the bud remains dormant (Cook and Jacobs, 2000). While the extent of endodormancy is being reduced, bud break is inhibited by ecodormancy, due to unfavourable environmental conditions. Low temperatures, inadequate nutrients, or other limiting factors are considered part of ecodormancy, and once the tree is exposed to warmer spring temperatures, with no other limiting factors present, ecodormancy no longer inhibits growth, and bud break ensues (Lang, 1987).

In winter, a certain measure of cold is required to break, or alleviate, the endodormant state of the buds; this is referred to as the chilling requirement (Fishman *et al*., 1987). A model to quantify the amount of winter cold such as the Utah model, measures “chilling units”, and considers the efficacy of chilling at various temperature intervals; this model also take into account the negation of previously accumulated units, by subsequent warm temperatures (Cook *et al*., 2017). If the winter temperatures are too warm, the buds will not receive sufficient chill units to exit endodormancy. This inadequately chilled state, results in buds that will remain partially dormant, and at the onset of spring, normal bud development will be impeded (Cook and Jacobs, 2000). This effect, of insufficient chilling, is exacerbated by cold spring weather (Lang, 1987). Trees that received a greater degree of winter chill, will respond with bud break at lower spring temperatures, compared to the case of insufficiently chilled trees, which require more heat during spring to initiate bud break (Atkinson *et al*., 2013). With regard to dormancy alleviation, spring temperatures have an important role to play. The issues caused by insufficient cold in winter can be exacerbated by a cool spring. Trees that received greater degree of winter chill will respond with bud break at lower temperatures compared to the case of insufficiently chilled trees, which require more heat during spring to initiate bud break (Atkinson *et al*., 2013).

Symptoms of warm winter conditions are extended bud break and a delay in the onset of bud break. This protracted bud break means that once buds start breaking in spring, the bloom period will be extended and flowering will be staggered. This can result in poor cross pollination, low fruit set and variable maturity in fruit ripening; which in turn, affects the timing of chemical sprays and forces producers to perform multiple picks at harvest (Erez, 2000). The total percentage of final bud break is also reduced, decreasing the number of flowers and shoots which develop, and resulting in lower yields and poor foliage development (Cook and Jacobs, 2000). In the case of apple trees, branching is modified toward basal dominance, with few distal auxiliary buds bursting, in relation towards increased proximal lateral buds bursting (Cook and Jacobs, 1999). This negatively effects the morphology of the tree and is detrimental to the establishment of new orchards where acrotonic branching is necessary for filling the space allocated to each tree (Cook and Jacobs, 1999). The lack of winter cold has also been shown to reduce flower quality. Studying the effect of winter chill on sweet cherry flower quality, Mahmood *et al*. (2000) showed that increased winter chill produced larger flowers with longer pedicels and led to a greater proportion of fruit set. Flower quality is an important factor in fruit set and final fruit quality in apples (Lauri *et al*., 1995).

High variation in fruit maturity makes the determination of a harvest date challenging, and when the average maturity of an orchard is optimal for harvest, there will be a portion of fruit both above and below the desired maturity parameters (Erez, 2000). Aside from a lack of flavour development, the storage of immature fruit can result in physiological disorders such as superficial scald, internal browning, or shrivelling (Fellman *et al*., 2003). The storage of over mature fruit is less effective than with optimally ripe fruit and it has a significantly reduced shelf life (Costamagna *et al*., 2012; Peirs *et al*., 2005). Similarly, Lau (1998) showed that later harvested 'Breaburn' apples had a higher incidence of internal browning. Having a large amount of variation in maturity at harvest could reduce the storage potential of the entire batch since immature and overmature fruit are combined. It also makes it difficult to implement and optimally operate newer storage technology such as dynamic controlled atmosphere (DCA) storage by means of chlorophyll fluorescence. DCA entails assessing the fluorescence signals of a small sample of fruit which in the case of a high level in variation between fruit, might not be representative of the whole cold room and may result in suboptimal storage conditions for some fruit.

4.1.2 Growing season temperatures

The growing season starts at bud break, typically mid-October for ‘Golden Delicious’ in South Africa, and continues until leaf drop and the start of the dormant period at the end of May or early June.

After bud break, the reproductive buds give rise to flower clusters. Pollination is integral to apple production and is the transmission of pollen from the anther to the style. Apple inflorescences are generally self-incompatible for fertilization, and the crosspollination from a compatible cultivar, is necessary to stimulate growth and development of the ovary, and to set fruit (Ramírez and Davenport, 2013). For successful crosspollination to take place, the bloom period of the chosen cultivar and the pollinizers, need to overlap, to ensure that pollen is available at the required time (Dennis, 2003). Most apple cultivars readily bloom together under ideal climates, but when faced with warmer winters, bloom periods are erratic, and more care needs to be taken in choosing a suitable pollinizer (Dennis, 2000a).

Honeybees act as pollinators, and in commercial orchards hives are deliberately placed to increase pollination. Temperature has a considerable influence on insect activity. Bee activity is inhibited at temperatures below 13°C and an incremental increase in bee activity is noted for each 1°C increase until 19°C, where further temperature increases show negligible effects on bee activity (Ramírez and Davenport, 2013). Once pollen has been transferred and germinated, pollen tube growth needs to take place. Pollen tube growth is temperature related and requires 2 days to reach the ovules at 15°C, 4 days at 13°C and 8 days at 9°C (Williams and Maier, 1977). Warm temperatures are desirable during bloom, but extraordinarily high or low temperatures are detrimental to fruit set (Dennis, 2003). Williams and Maier (1977) determined that at 20°C effective pollination was maximized. If successful pollination takes place, many of the flowers will set fruit. In most case this number of fruit will be too high, which increases the effort required for fruit thinning. If too many fruitlets are left on the tree, it could suffer excessive reserve depletion, reduced cold hardiness, and breaking of branches. Furthermore, excessive crop load reduces the following year’s bloom, because of resource depletion and an inhibition of flower bud initiation (Tromp, 2005b). Excessive crop loads also reduce fruit size and quality (Dennis, 2000b).

The respiration rate and carbon assimilation in the leaf are both responsive to changes in temperature. Leaf assimilation behave quite differently and shows a parabolic response to temperature with a peak centred around 30°C; it is a broad-shouldered curve within the 15-35°C range but at temperatures above 35°C a sharp decrease in leaf assimilation rates is observed (Lakso *et al*., 1999). From 10-30°C leaf dark respiration increases exponentially by a factor of 2.5 for each 10°C incremental increase of temperature. The other plant parts, such as stems, trunks and roots, also show this exponential increase in respiration, but their temperature fluctuates less, due to buffering by a greater volume to surface area ratio (Lakso *et al*., 1999). These other plant parts thus see only small fluctuations in respiration.

The time of fruit thinning is integral to minimizing the mentioned negative consequences of high crop loads. Timing plays a key role because of the different growth phase of an apple. For the first three to four weeks, growth takes place in the form of cell division and thereafter growth only takes place as cell enlargement (Bain and Robertson, 1951). The final number of cells at harvest can range from 27 to 67 million cells and thinning before the cell division phase has ended has shown high final cell numbers (Dennis, 2000b; Smith, 1950). Warm spring temperatures (19-25 °C), during the aforementioned first three to four weeks, are favourable for cell division, and not only does this result in larger fruit, but an increase in fruit dry matter has been observed (Warrington *et al*., 1999). Apple fruit size increases at an exponential rate until 7-20 days after cell division has stopped, after which fruit growth is linear. This linear growth continues until near harvest where it slows to an almost complete stop. By measuring heat units over a number of years from four European apple growing regions, Kronenberg (1988) showed that fruit growth during the exponential phase, as well as towards growth cessation near harvest, were more sensitive to temperature and responded positively to relatively warmer temperatures. During the linear growth phase temperature did not appear to have an influence on final fruit size (Kronenberg, 1988).

Cooler mid-season temperatures have shown a delay in maturity with their harvest dates being later than from apples grown in climates with warmer midseason temperatures (Warrington *et al*., 1999). A disparity arises 1-2 weeks before harvest, where high late season temperatures inhibit starch breakdown in fruit and cold temperatures hasten it (Watkins *et al*., 1982).

4.2. Light

Light provides energy for the growth and development of the plant, and it has a profound effect on tree growth, biomass production, and produce quality (Corelli-Grappadelli and Lakso, 2004). The leaves of the tree convert light energy into chemical energy by means of photosynthesis, and for this to take place, the leaves need to be exposed to light in the visible spectrum (Tromp, 2005b). Thus, it is desirable for photosynthetic rates to be maximized, to supply an adequate amount of carbon assimilates for the different plant parts (Corelli-Grappadelli and Lakso, 2004). For optimal carbon assimilation by photosynthesis, a maximum amount of incident light radiation needs to be intercepted, while maintaining adequate distribution of said light throughout the leaf canopy.

There is a direct relationship between the amount of light intercepted by a tree, and the total amount of dry matter fixed (Wünsche *et al*., 1996). A maximum light interception (100%) is realized when no incident light radiation reaches the soil, and while this is easily achieved with cereals or other densely planted arable crops, maximizing light interception of perennial tree crops is challenging (Tromp, 2005b). Tree crops pose more of a challenge, which need to develop a branch canopy over years, and requires alleyways, to grant access for machinery and tractors (Bastías and Corelli-Grappadelli, 2012). Because of this, achieving 100% light interception in apples orchards is not beneficial to production; the distribution of light would need to be suboptimal to make 100% light interception possible. Wertheim *et al*. (1986) confirmed this and showed that while apple orchards show a linear increase in productivity up to 70% interception, further increases would cause too much shading to the lower branches and inner canopy and productivity would decline (Wertheim *et al*., 1986). Fouché *et al*. (2010) demonstrated in a study on 'Granny Smith' that fruit quality differed from the inner and outer canopy, in terms of green colour intensity, with fruit in the inner canopy being pale. The fruit being pale is considered a negative quality trait but they also found that in the outer canopy there was a higher incidence of sunburnt fruit (Fouché *et al.,* 2010).

When light interception is optimized, the resultant increase of dry matter production does not automatically equate to increases in marketable yields, as the partitioning of resources is not necessarily weighted towards fruit (Tromp, 2005b). The partitioning of resources is influenced by the amount, quality, and distribution of light (Bastías and Corelli-Grappadelli, 2012). Shading can be detrimental and has been shown to reduce the amount of resources available to fruitlets during the cell division phase of growth (Corelli Grappadelli *et al*., 1994; Smith, 1950). In apple trees, the partitioning of carbohydrates between vegetative and reproductive growth responds to changes in light levels (Corelli-Grappadelli and Lakso, 2004). Limited light at the start of the season, has a greater effect on fruit growth than on shoot growth (Tromp, 2005a). Corelli Grappadelli *et al*. (1994) stated that shade during the early part of the season delays the transition of growing shoots from being net importers, to becoming net exporters of carbon. Bepete and Lakso (1998) confirmed this, showing that once light level drops to 75% of full light, fruit growth begins to decrease. Furthermore, a 25% reduction of fruit growth was seen at 45% full sun, and at 15% sunlight, fruit growth was reduced by up to 50%. Shoot growth appeared unaffected, and where the lowest level of light was tested (15% full sun), shoot growth remained unimpeded (Bepete and Lakso, 1998).

A linear relationship exists between light interception by spur leaves, and total yield per tree (Wünsche *et al*., 1996; 2000). These primary spur leaves are the earliest contributors to fruit growth and coincide with the cell division phase of fruit growth (Corelli-Grappadelli and Lakso, 2004). Aside from light's effect on fruit growth, evidence suggests that light is required for flower bud formation (Tromp, 2005b). Flower bud formation in apple is not responsive to photoperiod but rather responds to light level (Hennerty and Forshey, 1971). Proctor and Crowe (1983) found that shading one-year-old potted apple trees reduced flower bud formation, and although the number of flower buds reduced, shading did not appear to affect the number of shoots significantly. Gur (1985) states that at least 30% of full sunlight is necessary for flower bud formation in apples. The ratio of red:far-red light decreases in the shaded parts of the canopy as shorter wavelength light does not penetrate as deeply as far-red light (Tromp, 2005b). Phytochrome sensing within plant organs respond to these changes in light, however, there is very little information regarding this, on apple flower formation. Large fruit size, high soluble solids and good colouration of red or blushed cultivars were positively correlated to high light levels, but the opposite was seen with storage characteristics, where higher light exposure showed poorer storage characteristics (Jackson *et al*., 1977). Furthermore, excessive light, or exposing previously shaded fruit, is known to cause photo-bleaching (Racsko and Schrader, 2012). Previously mentioned, hot temperatures on the apple fruit surfaces, causes tissue damage and result in sunburn. Here, light level plays a significant role, where sun exposed fruit can be 15°C higher than ambient temperature (Chen *et al*., 2008).

5. Variation within orchards and within trees

Micro-climatic variations within an orchard can influence the variation of fruit quality in said orchard. A study by Aggelopoulou *et al*. (2010) on ‘Red Chief’ apples showed that climatic differences within an orchard caused significant variation between the trees in terms of yield and quality. Serra *et al*. (2016) showed that variation in crop load within a ‘Honeycrisp’ orchard also led to significant variation in maturity and final fruit quality.

It is typical for much of the area under apple production in South Africa to be planted at lower densities (<1667 trees per ha). Only recently have producers adopted higher density planting. In South African orchards where each tree has a relatively large canopy, and inadequate light management and pruning is done, there will be poor light distribution and a notable temperature gradient within the tree. These variations in light and temperature within a tree influence carbohydrate allocation to certain organs, and with more complex tree structures a significant variation in fruit quality can arise (Sperling *et al*., 2017; Wünsche and Palmer, 1996). Fruit from the outer and inner canopy can differ in appearance, quality and consumer satisfaction (Hamadziripi *et al*., 2014). Fouché *et al*. (2010) showered for ‘Granny Smith’ that greener fruit were produced in the shaded portion of the canopy compared to the outer canopy while also finding the palest fruit in the most shaded part of the canopy. Evidently, maintaining a well exposed canopy that allows for high light interception and distribution, has driven orchard design and pruning systems toward higher density plantings (Corelli-Grappadelli and Lakso, 2007).

Aside from environmental conditions, shoots and spurs do not necessarily have the same potential in producing high quality apples (Hirst and Ferree, 1995), but genetic makeup determines which bearing position has the highest quality potential (Parisi *et al*., 2014). ‘Golden Delicious’ typically bears the best fruit on short shoots (Lauri *et al*., 1995; Lespinasse and Delort, 1984). The bearing potential of specific ‘Golden Delicious’ wood (shoots vs spurs) is not necessarily consistent across all climates. Segura *et al*. (2007) evaluated the phenotypic expression of a ‘Granny Smith’ x ‘Starkrimson’ cross and highlighted that the environmental factors of the two regions in their study did not influence expression; however, their study did not use a production region with insufficient winter chill which could have caused differences in architectural expression. It would be useful if future tree architecture studies included milder winter climates in their studies.

To determine the causes of within tree variability Volz *et al*. (1993) recorded bloom date, wood age, orientation and cluster size of ‘Royal Gala’ apples. Fruit was harvested multiples times so that each fruit was picked near to optimum maturity, and this date of maturation did correlate well with bloom date. The latest fruit to mature had the poorest quality (low firmness and small size). Bearing position had a greater effect on final fruit quality with those on short shoots being best. Their work suggests that ‘Royal Gala’ bloom date was not important in determining fruit quality or maturation date, but this would not necessarily hold true for other cultivars. To date protracted bloom periods (due to insufficient winter chill) have been studied well (Cook *et al*., 2017; Erez 2000; Funes *et al*., 2016; Gadoury, 2015; Hauagge and Cummins 1991), but work on the influence of bloom period on final fruit quality and maturity in these conditions has not been done.

Both the internal and external quality traits are affected by fruit maturity. In the case of ‘Golden Delicious’, fruit yellow during maturation and continue do so after harvest in storage. Fruit flesh firmness is considered both a quality and maturity parameter and more mature fruit have lower firmness (Wills *et al.,* 1980). Fruit maturity at harvest influences firmness after storage, with lower initial firmness before storage dropping even further during storage relative to initially firmer fruit stored under the same conditions (Sass *et al.,* 1992). Børve *et al*. (2013) showed that fruit harvest time influenced fruit maturity with optimally harvested fruit having the best quality after storage. This implies that if variation in fruit maturity is present on a tree at a particular time, variation in fruit quality is to be expected.

6. Conclusion:

Consumers desire an apple that is visually appealing and provides a satisfactory eating experience. It is important to deliver a consistent supply of apples to consumers of acceptable quality. The prevalence of variation in quality parameters are negatively perceived and need to be prevented/mitigated, because an increased uniformity has shown an increase in perceived quality.

Duly it is essential for these external and internal characteristics to be as optimal and uniform as possible at harvest. Climatic and microclimatic conditions have a significant effect on fruit quality parameters. In South Africa, the climate is suboptimal in terms of winter chill and the tree architecture is complex, likely giving rise to a great degree of fruit variability at harvest. Much of the variation in size, colour, and shape are managed by sorting in pack houses, but the presence of inferior quality (sunburn, bruising and injuries etc.) fruit reduces the potential profit for growers as these are sent to juice processing. Although in development, sorting apples according to internal characteristics remains a challenge in the industry (Vanoli and Buccheri, 2012). Variation is present in the batches of fruit coming into the pack house and reduces the potential income for fruit growers in South Africa. Understanding the origin of variation in fruit quality and maturity could lead to optimised production practices. This is of particular importance for cultivars like ‘Granny Smith’ and ‘Golden Delicious’ which are strip picked and variation in storage maturity can lead to deterioration of the product and loss of income. This problem is also encountered with new full red strains of ‘Gala’ apples, which have typically been picked on ground colour, and visual determination of maturity has become a challenge for pickers.

Existing work has detailed sources of variation and with the rise of precision agriculture the scale of the variability has become more apparent. Future work should not neglect the influence of insufficient winter chill on fruit variability, because as the climate begins to warm, more regions will be confronted with these challenges.

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PAPER 1: Variation in fruit maturity and quality of ‘Golden Delicious’ apples from two climatically distinct regions.

Abstract

Fruit quality and maturity are paramount to the commercial viability of apple orchards. Fruit maturity has a direct effect on fruit quality and storage potential. Quality varies not only from season to season, but within a season many factors can contribute to the overall variation in an orchard. Understanding where this variation originates from, can help producers mitigate such issues more effectively. To determine variability in fruit quality and maturity, six full bearing ‘Golden Delicious’ trees were selected in orchards representing two climatically different apple production regions within the Western Cape, South Africa (11 orchards were in Elgin and 13 in the Koue Bokkeveld). All the fruit from two branches on each tree was harvested at commercial harvest and half of the fruit was stored in regular atmosphere for 12 weeks at -0.5 °C. Fruit appearance was measured using the variables: fruit aspect ratio, mass, and peel colour, and starch conversion percentage and flesh firmness were used as maturity indicators. The variance components investigated were region, orchard, tree, canopy position (top vs bottom fruit, sun exposed vs shaded fruit) and bearing wood (shoots vs spurs). Orchard variation was the greatest contributor to the total variability in both seasons, followed by tree-to-tree variation. The variation in starch conversion within an orchard was found to be considerable irrespective of region with most orchards having high numbers of fruit outside the optimum harvest standard. Canopy position (spatial positioning interacting with light exposure) influenced fruit firmness, with fruit from the upper canopy and sun exposed fruit being firmer. This effect was more pronounced after storage. Fruit colour (hue angle and lightness) correlated well before and after storage, but not with the other parameters such as starch conversion and/or flesh firmness. Results confirm that sorting fruit based on colour in a pack house is useful but addressing variation in maturity (starch conversion and fruit firmness), should be done at tree level and could differ across production regions. Horticultural practices that reduce canopy complexity are recommended to reduce fruit variability.

Keywords: colour, firmness, position, *Malus domestica,* starch

1. Introduction

Consumers desire fruit of high and consistent quality which has prompted producers to focus on the production of first-class apples (Sansavini *et al.,* 2004). External fruit traits such as shape, size, colour and peel blemishes, are contributing factors to perceived quality and drives the initial purchase of the fruit (Jaeger *et al.,* 1998; Kays, 1999). What brings the consumer back to the shelf and drives repeated purchases are the internal quality traits of the fruit, specifically texture, sweetness, acidity and the flavour profile (Daillant-Spinnler *et al.,* 1996; Jaeger *et al.,* 1998). Both the internal and external quality traits are affected by fruit maturity.

In the case of ‘Golden Delicious’, fruit yellow during maturation and continue do so after harvest in storage (Blažek *et al.,* 2003). Fruit flesh firmness is considered both a quality and maturity parameter and more mature fruit have lower firmness (Wills *et al.,* 1980). Fruit maturity at harvest also influences firmness after storage (Sass *et al.,* 1992). The direct influence of maturity on fruit quality suggests it can be viewed as a quality parameter. Børve *et al*. (2013) showed that fruit harvest time influenced fruit maturity with optimally harvested fruit having the best quality after storage. This implies that if variation in fruit maturity is present on a tree at a particular time, variation in fruit quality is to be expected.

Consumers presented with a batch of fruit are perceptive of, not only the quality, but also the uniformity of the fruit within the batch, and when consumers recognise a variation in size, colour, or other distinguishing characteristic, their perception of the overall fruit quality within the batch is negatively influenced (Richards and Patterson, 2000; Richards, 2000).

Variability in fruit quality can be introduced in several ways. When genetic factors are kept constant, the strongest effector of yield, quality, and variation in apple production is the environmental conditions, such as temperature and light (Palmer *et al*., 1989). Light availability and light quality, influences fruit size, colour and dry matter content (Corelli-Grappadelli and Lakso, 2004). As many biochemical and signalling pathways within a plant are temperature dependent, understanding how trees interact with temperature and temperature changes is imperative. Aggelopoulou *et al*. (2010) described significant spatial and temporal variation of fruit quality within an orchard, citing the effect of topography and angle of the slope (slope aspect), on microclimates within an orchard. Slope aspect and the direction of the slope, both influencing temperature and amount of light, had a marked impact on fruit quality leading to variation in fruit quality from tree to tree in the same orchard. Another possible source of variation arises from the type of bearing wood on which fruit is borne. Shoots and spurs do not necessarily have the same potential to produce high quality apples (Hirst and Ferree, 1995). The genetic makeup of the cultivar determines the bearing position that has the highest quality potential (Parisi *et al*., 2014).

Bepete and Lakso (1998) showed that insufficient light reduced fruit size. Lakso *et al*. (1999) looked at the carbon balance within the tree, assessing the relationship between light interception and shading in determining yield potential and productivity, linking this to the localized availability of photosynthates. Bastías and Corelli-Grappadelli (2012) showed that manipulation of the light conditions had a physiological impact on growth and fruiting habits in apples. Duly a complex canopy, with varying light conditions could produce erratic results in fruit quality within a single tree canopy. Even minor variations of light within a canopy have an influence on green colour development in 'Granny Smith', with innermost and outer canopy fruit being less green than partially shaded fruit (Fouché *et al.,* 2010). Conversely, light in excess causes photobleaching, and when coupled with elevated temperatures, sunburn can develop (Racsko and Schrader, 2012).

Temperature conditions also need to be contended with. Gouws and Steyn (2014) looked at red fruit colour development in two of the main apple production regions in South Africa, highlighting the temperature ranges required for optimal colour development. Additional challenges are faced in South African apple production, because of climate change. Predicted warming is believed to effect Mediterranean-like climates, and besides the challenges of increasing summer temperatures, a warmer/milder winter can also increase variation in fruit quality (Baldocchi and Wong, 2006; Midgley and Lötze, 2008). This is also true on a smaller, within-tree resolution; Aylor (1995) indicated that differences in air movement, temperature and humidity within the tree canopy influenced the spatial development of *Venturia inaequalis* (apple scab). In large tree canopies, variations in light distribution and temperature gradients have a physiological impact, influencing carbohydrate allocation to the different tree organs (Sperling *et al.,* 2017).

A myriad of factors influences fruit quality and knowing to what extent each factor contributes to the variation, will help direct producers in their efforts to produce more uniform fruit of good quality. Microclimate and bearing position can partially be manipulated by production practises such as choice of tree architecture, pruning, and tree spacing in orchards, all having an effect on light interception and distribution within the tree canopy (Dallabetta *et al.,* 2012). The aim of this study is to describe and quantify the variation in 'Golden Delicious' fruit maturity and quality parameters by considering pre-harvest variance components such as growing region, orchard, tree, canopy position (top vs bottom fruit, sun exposed vs shaded fruit) and bearing wood (shoot vs spurs).

2. Materials and methods

2.1. Plant material and study site

During the 2016/2017 season, 24 'Golden Delicious' orchards were selected from two different production regions in South Africa. In the Elgin (34.2°S, 19.0°E, 305 m.a.s.l.) region 11 orchards were used and 13 orchards in the Koue Bokkeveld (33.4° S, 19.5° E, 945 m.a.s.l.). These two regions are climatically different with Elgin receiving less winter chill (annual avg. ±700 Utah chill units (CU)) for dormancy alleviation compared to the Koue Bokkeveld (annual avg. of ±1400 Utah CU). Elgin received 795 Utah CU before the 2017 growing season and 965 before the 2018 season, while the Koue Bokkeveld had 1311 and 1199 Utah CU accumulated before the 2017 and 2018 seasons, respectively (Hortgro, 2017). 'Golden Delicious' typically requires 1400 Utah CU (Ogundeji and Jordaan, 2017). The following factors were standardised when selecting the orchards: rootstock

(M793), avg. annual yield (60-80 t/ha), solaxe-type central axis training system and row orientation (less than 10 degrees off north-south). Soil type, orchard size and tree height were not selected for. Standard commercial horticultural practises were followed in all the orchards and a rest breaking agent (0.26% cyanamide and 3% oil) was applied after bud swell. Six representative trees were selected in each orchard and two scaffold branches (perpendicular to row direction and on eastern side only) were identified (one branch in the top and one in the lower part of the tree). Branches with more than 30 fruit were selected and in cases where the branches carried fewer than 30 fruit, an additional adjacent branch was selected.

In the 2017/2018 season, the trial was repeated on 10 ‘Golden Delicious’ orchards selected from the same two production regions. Five orchards were selected in the Koue Bokkeveld and five orchards in Elgin with four trees randomly selected within each orchard. Orchard standardisation and branch selection practises were similar to the 2016/2017 season.

2.2. Fruit handling and harvest maturity

Fruit were harvested on, or within one day, of the commercial harvest date as determined by the producers. ‘Golden Delicious’ is commercially harvested at a minimum average starch conversion percentage (SC%) of 15% and a firmness minimum of 6.8 kg (Department of Agriculture, Forestry and Fisheries (DAFF), 2013). Although no maximum SC% is stipulated, commercial standard pack houses typical consider >50% starch conversion on ‘Golden Delicious’ as over mature. Duly orchards with an average starch conversion above 50% were excluded from the statistical analyses but are still presented when indicating the orchard variation. All the fruit from the top and bottom scaffold branches were harvested and sorted into the following eight categories (Shoots were defined as spurs when they were <5 cm long, and as long shoots if >5 cm.):

1. Top branch, sun exposed, on spurs.
2. Top branch, sun exposed, on long shoots.
3. Top branch, shaded, on spurs.
4. Top branch, shaded, on long shoots.
5. Bottom branch, sun exposed, on spurs.
6. Bottom branch, sun exposed, on long shoots.
7. Bottom branch, shaded, on spurs.
8. Bottom branch, shaded, on long shoots.

All fruit samples were stored in regular atmosphere at -0.5°C and processed within five days. The fruit from each orchard was split into two equal sample sets, with fruit from three trees in each set. Both sample sets underwent non-destructive analysis yielding the following parameters: fruit dimensions, fruit mass, fruit peel colour, and external defects. Fruit dimensions were measured as fruit length and diameter and expressed as a ratio (length:diameter) - this ratio is referred to as "fruit aspect" (Brown, 1960). Following the non-destructive analysis, the first sample set underwent destructive analysis to determine fruit firmness and percentage starch conversion. The second sample set was packed into standard export quality apple cartons, with the necessary packing material and stored at -0.5 °C in regular atmosphere storage (RA) for 12 weeks followed by a seven-day shelf-life period at room temperature. After the shelf-life period, the second sample set was submitted to the same non-destructive and destructive analysis as described before, excepting starch conversion tests. All measurements were completed at room temperature, within 24 hours after removal from cold storage. See Fig. 1 for a summary of the laboratory procedure to create the datasets.

Fruit length and diameter were measured in millimetre with an electronic calliper (Model EC799, Starret, USA). Fruit mass was recorded to the nearest gram by an electronic scale (Model C131AM, Hygeco, France). Fruit peel colour was determined using a chroma meter (Model CR-400, Konica Minolta, Japan) and expressed as XYZ colour space coordinates. The standard XYZ colour space coordinates measurements from the colourimeter were converted to the HCL colour space, of parameters: hue angle, chroma and lightness (Mahyar *et al.,* 2010). A single colour measurement was taken per fruit on the shaded cheek, and anywhere for fruit that were in the shade. Sunburn was scored using increasing severity of sunburn, with '1' representing blemish free and '6', representing necrotic stage sunburn (Schräder *et al*., 2003). Severely injured fruit, or fruit containing necrotic and diseased lesions, were discarded after the non-destructive analysis and not included in the destructive testing and storage. Fruit firmness was measured on two opposite peeled cheeks of the fruit using an 11 mm penetrometer (Model GS-20, GŰSS Manufacturing Ltd. South Africa), after which the fruit were cut horizontally in half. The fruit flesh of the lower half was painted with an iodine solution to visualise the starch content. The Unifruco Research Services pome fruit starch conversion chart was used to categorise the starch conversion percentage into the following categories: 5%, 10%, 20%, 25%, 30%, 40%, 60%, 70% and 80%. An additional 100% and 0% score was given to fruit on the upper and lower extremes to account for completely immature and overripe fruit.

2.3. Statistical analysis

The variance components were determined as a nested model with random effects using the Proc VarComp procedure in SAS (Version 5.1, SAS Institute Inc, USA) with individual fruit as the experimental unit. The contribution of canopy position, light exposure and bearing position were grouped together as "position" during the analysis and not analysed as individual entities. An ANOVA was performed on tree level, using the GLM procedure in SAS, followed by Fisher's Least Significant Difference test when the significance level was greater than p =0.05. For this analysis "position" was not grouped but analysed individually. All parameters were tested for possible correlations using the Pearson's correlation test in XLSTAT (Version 2015.04.36025, ADDINSOFT, France) at a significance level of $p = 0.05$. The Pearson's correlation coefficients were interpreted as follows: very weak (0.0-0.19), weak (0.20-0.39), moderate (0.40-0.59), strong (0.60-0.79), very strong (0.80-1.00) (Evans, 1996; Hinkle *et al*., 2003). Only strong and very strong correlations were commented on.

3. Results and discussion

To simplify and organise the plethora of statistical outcomes the individual parameters of the appearance and maturity indices are presented separately. For each of the parameters both the variance components and the ANOVA results are presented and discussed starting with the variance components, followed by only the highest order significant ANOVA results (main and/or interaction effects). Fig. 2 (2017 season) and Fig. 3 (2018 season) refer to the variance component analysis and Table 1a (2017 season) and 1b (2018 season) summarise the significant main effects and highest order interactions from the ANOVAs for the individual parameters. Results for the contribution of orchards and tree-to-tree variation for both years are presented in Table 1c. Orchard differences are discussed with the other variance components, but the contribution of tree-to-tree variance is discussed separately in section 3.3. Complete ANOVA tables, showing all the main and interaction results for each of the parameters, are available in Appendix A.

NOTE: *To maintain a logical separation between the ANOVA tables of the two seasons, Tables 2-12 pertain to the 2017 results followed by Table 13–21 for 2018. However, when the parameters are discussed, results from both years are considered and therefore the table numbers in the text are not always in order of appearance.*

3.1 Appearance parameters

3.1.1 Fruit aspect

Fruit aspect ratio, the relationship between length and diameter of a single fruit, has a strong genetic component but various environmental growing conditions can change the phenotypic expression (Brown, 1960). A larger fruit aspect ratio value is indicative of a fruit having a relatively smaller diameter for a given length. Fruit with a higher aspect ratio appear “taller” where fruit with a small aspect ratio appear “flatter” to the eye. Although Warrington *et al*. (1999) determined that temperatures during the growing season had little effect on fruit aspect within a given cultivar, Petri and Leite (2003) showed that the temperature conditions during winter dormancy can influence final fruit shape, with warmer temperatures resulting in smaller aspect ratios, producing ovoid shaped fruit.

In 2017 the variation in fruit aspect was primarily affected by region, orchard, and tree differences, contributing 33.3%, 9.1%, and 5.6%, respectively to total variation (Fig. 2). The unexplained variation was 51.6% and the remaining 0.4% of variation was collectively caused by differences in position, and the region x position interaction (Fig. 2). Region, orchard, and tree differences, were also the main contributors in 2018, contributing 20.7%, 10.1%, and 6.1%, respectively to total variation (Fig. 3). The unexplained variation was 62.7% and the remaining 0.4% of variation was collectively caused by position and the region x position interaction (Fig. 3).

The regional contribution to variation is illustrated using density curves in Fig. 4a and 4b, showing the distribution of the fruit aspect for Elgin and the Koue Bokkeveld in 2017 and 2018, respectively. Density is the relative frequency of an observation, divided by the frequency interval. The area under the curve is equal to 1. A higher density for a given aspect ratio value, means that relatively more observations were at that value. In 2017, fruit aspect in Eglin had a peak density of 0.25 (fruit aspect interval: 0.90-94) and in the Koue Bokkeveld it was 0.22 (fruit aspect interval: 0.97-1.01), thus showing that the Elgin fruit peaked at a lower fruit aspect value and a higher peak density than fruit from the Koue Bokkeveld. The 2018 season (Elgin: density of 0.18, fruit aspect interval 0.94-0.96; Koue Bokkeveld: density of 0.16, fruit aspect interval 0.97-1.01) showed a lower peak density for both regions compared to 2017, suggesting greater variation in the shape in the second season. The unexplained variation was just over 10% greater in 2018 than 2017, and because less of the variation was accounted for by region, a greater proportion of variation remained unexplained in the second season. Elgin had a higher mean aspect ratio in 2018 (0.948 ±0.036) than in 2017 (0.923 ±0.037) although the Koue Bokkeveld didn’t change much between 2017 (0.992 ±0.037) and 2018 (0.985 ±0.040). Elgin had more accumulated chilling for the 2018 season than that of 2017, likely explaining the difference in aspect ratio between the two years. The contribution of winter temperatures is quite apparent here, with milder winters producing fruit with a lower aspect and appearing “flatter” and this effect accentuating in even milder years. This concurs with what Petri and Leite (2003) found.

The orchard contribution (9.1% in 2017 and 10.1% in 2018) to the total variance in fruit aspect and the significant difference (p<0.0001 in both years, Table 1c) amongst the mean values of the orchards are best depicted with box plots for the individual orchards across regions (Fig. 5). In 2017, the fruit aspect across all orchards ranged from 0.76 to 1.17 and in 2018 from 0.79 to 1.16. Overall, this suggests a similar, high degree of variability for both years. When considering the regions, the interquartile range in Elgin fruit aspect seems marginally smaller than in the Koue Bokkeveld in 2017 but not in 2018. This points towards greater uniformity of fruit aspect in Elgin despite the high variability. This too concurs with the higher density peaks seen in Fig. 4. Some orchards from the Koue Bokkeveld in 2018 show a small degree of positive skewness, with the other box plots more normally distributed.

From the ANOVA results the highest order, significant interaction for 2017 was between region, tree position and light exposure (RxTPxLE) (p = 0.047, Table 1a and 2). When considering the mean fruit aspect ratio of this interaction it is clear that the biggest difference is between the two regions (Table 2). This is also confirmed by the large F-value for the region main effect (F=1567.09, Appendix A, Table 1). The overall mean fruit aspect ratio for the Koue Bokkeveld (0.992 ±0.037) was significantly larger compared to Elgin (0.923 ±0.037) with a *p*-value of <0.0001. The RxTPxLE interaction further illustrated differences within the regions in terms of tree position and light exposure, but although statistically significant, these differences are deemed too small to be of practical or commercial importance.

The interaction between region, tree position and bearing position (RxTPxBP) yielded statistically significant differences with a *p*-value of 0.029 (Table 1a and 3). Again, it is clear from the comparison of the means that the region effect is dominating with fruit from the Koue Bokkeveld again having a greater fruit aspect ratio compared to Elgin, thus taller fruit. Similarly, further differences within the interaction (in terms of tree and bearing positions) were statistically significant, but too small to consider them noteworthy.

Regardless of region, light exposure and bearing position (LExBP) yielded a significant interaction (*p* = 0.003; Table 1a and 4). Here it was evident that sun exposed fruit borne on shoots, had a larger mean aspect ratio than fruit borne on sun exposed spurs which were flatter. This was in accordance with the significant bearing position main effect (F = 20.02; *p*<0.0001; Appendix A; Table 1*)* but did not hold true for the shaded fruit, where the interaction with light exposure resulted in similar shoot and spur borne fruit mean aspect ratios.

Petri and Leite (2003) described apples from milder winter climates having a smaller fruit aspect. Our findings resonate with this as fruit from the Koue Bokkeveld region, which had a higher chill accumulation had a greater mean aspect ratio. Consumer preference on shape of apple fruit, was done by Hampson and Quamme (2000), showing no particular preference among Canadians. The judges responded with either neutral or a slight affinity to all shapes. Research done on fruit shape of litchi (Rahman *et al*., 2010) and watermelon (Sadrnia *et al*., 2007), suggests that uniform dimensions (shape) within a batch and a lack of visible deformities are more important and detectable by consumers. As this variation is easily recognisable, diligent sorting and packing after harvest should address this concern. From the results presented here there might be an increased risk of introducing variability when exporting containers with fruit from both regions simultaneously to the same buyer or supermarket.

It was also seen that depending on light exposure, shoots and spurs can bear fruit with different mean fruit aspect ratios. Literature comparing the effect of light and bearing wood on fruit aspect directly could not be found and considering the small degree to which these factors influenced fruit shape in only one of the two seasons, it does not grant further discussion.

3.1.2 Mass

Although fruit mass cannot be visually assessed by the consumer, it is indicative of fruit size. Fruit mass is important to consumers and is a regulated export criterion, with South African apples needing a minimum mass of 80 g before export is permitted (DAFF, 2013). Based on the correlation, producers are thus able to cull fruit based on a minimum diameter instead of weighing individual fruit. Literature shows that preference for size is not static. Canadians and many European countries prefer larger apples (Hampson and Quamme, 2000), while Norwegians prefer smaller apples

(Redalen, 1987). Having larger fruit does not necessarily indicate fruit of better quality, but to the producer larger apples for the same number harvested, means higher yields per unit area.

The variance component analyses illustrated in Fig. 2 and 3, also show the proportion that each factor contributes to the variation in fruit mass in 2017 and 2018, respectively. Region contributed strongly to variation in 2017 (23.4%) but not in 2018 (0.0%). Orchard-to-orchard (2017 12.4% and 2018 29.2%) and tree-to-tree (2017 13.1% and 2018 12.3%) variation in mass was present in both seasons. The contribution of orchard-to-orchard variation increased from the first season to the second, but tree-to-tree variation decreased. Position (combination of light exposure and bearing wood) and the region x position interactions contributed 1.2% in both seasons. The unexplained variation was smaller in 2018 (49.8%) than in 2019 (57.3%).

The regional effect is presented in Fig. 4c (2017) and 4d (2018) and shows the distribution of fruit mass for both regions using density curves. In 2017, Elgin peaked at interval 91.75-106.30 g (density: 0.19) and the Koue Bokkeveld at 120.85-135.4 g (density: 0.17). In the 2018 season, Elgin and the Koue Bokkeveld fruit mass both peaked at the 110.75-124.7 g interval, with Egin density 0.18 and that of the Koue Bokkeveld 0.15. There were 20 intervals of mass for the evaluation but because 2018 had a smaller range in a mass, the intervals were marginally smaller. Lower peak densities suggest greater variation. In both seasons Elgin appeared to have less variation in fruit mass than in the Koue Bokkeveld.

The box plots illustrate the variation amongst the orchards (Fig. 6a and b). There was a large range between minimum (24 g) and maximum (230 g) mass in 2017 across regions and in 2018 it ranged from 41 g to 235 g. Although the band that the interquartile ranges fell into was 60-160 g in 2017 and 78.75-172.25 g in 2018, the actual interquartile ranges appear to be greater in 2017 than in 2018. In terms of regions the interquartile ranges in Elgin were smaller than that of the Koue Bokkeveld. This, along with the density curves, suggest greater uniformity for fruit mass in Elgin than the Koue Bokkeveld and greater overall uniformity in the 2017 season. It is interesting, that the warmer Elgin winter (2017 season) resulted in the greater uniformity in fruit mass.

Considering the regional means, the average fruit mass in Elgin was 101.02 g (±21.82) in 2017 and 127.38 g (±22.84) in 2018 compared to the average mass of 125.37 g (±23.75) and 116.91 g (±29.30) in the Koue Bokkeveld in 2017 and 2018, respectively. The regional differences could not be looked at in isolation because of interactions with region and tree position (RxTP) in 2017 and a region and bearing position interaction in 2018 (RxBP). Tree position (TP) and bearing position (TPxBP) had a significant interaction in 2017 and the main effects of TP and light exposure (LE) were significant in 2018 (Table 1a and 1b).

In 2017, the interaction between region, tree position and light exposure (RxTPxLE) was significant with a *p*-value of 0.011 (Table 1a and 2). When comparing the means, the Koue Bokkeveld evidently had heavier fruit than Elgin. The interaction of region with tree position and light exposure within the Koue Bokkeveld, showed that shaded fruit on the lower branches had a significantly lower mass than the other positions in the tree. In Elgin, the interaction was not similar; fruit from the upper branches had a lower mass but were not significantly different from shaded fruit on lower branches. Sun exposed fruit on the bottom branches had the highest mass but was not significantly heavier than shaded fruit in the bottom of the tree. Overall, the largest differences are seen between the regions, which is confirmed by the large F-value of region (F=892.03, Appendix A, Table 2).

The interaction between tree position and bearing position (TPxBP) in 2017 was significant (*p*=0.050; Table 1a and 5). Regardless of position within the tree, shoots bore heavier fruit than spurs in the same position. The heaviest fruit were carried on shoots in the top part of the tree, significantly heavier than from bottom shoots. The mass of fruit borne on spurs did not differ significantly between the top and the bottom part of the tree.

In 2018, the combination of significant main effects and interactions did not overlap with what was seen in 2017. The region and bearing position (RxBP) interaction was significant (*p*=0.0004; Table 1b and 14) and in the Koue Bokkeveld fruit on spurs were heavier than fruit borne on shoots. The opposite was apparent in Elgin where shoots yielded heavier fruit than spurs. At a glance, it would appear that the Elgin fruit were heavier than fruit from the Koue Bokkeveld but in this case, the difference is confounded by the RxBP interaction.

Interestingly, in 2018, the main effect of tree position (TP) was statistically different for fruit from the top and bottom parts of the tree with a *p*-value of 0.003, but the LSD test did not confirm this. (Table 1b and 15). Even though the ANOVA indicates significant differences, the branch means are not significantly different according to other indicators. Typically, the leaves exporting carbohydrate to fruit in the top of the tree in a solaxed training system is exposed to light for a longer portion of the day and could therefore potentially fix more carbon increasing the fruit mass. Looking at the effect of light exposure (LE) in 2018, it had a significant effect (*p*= 0.046) on the mean mass of all the fruit (Table 1b and 16). The significance value is borderline and the indication of heavier sun exposed fruit should be interpreted in such a way.

In general, the 2017 season showed that production region played a large role in fruit mass, with the Koue Bokkeveld having heavier fruit. This was however not seen in the 2018 season. Reginato *et al*. (2019) found that winter climate has a direct influence on fruit mass. According to their study, fruit size was primarily dependent on winter climate (maximum average temperature during winter months) and to a lesser extent influenced by the effect of season length. They found that colder winters and longer growth seasons resulted in heavier fruit. Our results for 2017 agrees with their findings as the milder Elgin region resulted in lighter fruit compared to the Koue Bokkeveld. A possible explanation for the lack of effect in 2018, is that the difference in chill units between the regions was smaller in the winter preceding the 2018 harvest (234 units) than that of the 2017 harvest (516 units). It is likely that winter cold influences fruit mass, as shown by Reginato *et al*. (2019) for ‘Gala’ apples and could hold true for ‘Golden Delicious’, and the smaller difference in climate for the second season, suggests that the effect of other factors on fruit mass (such as historical crop load, fruit set distribution within tree and pollination efficacy) would be more pronounced (Hortgro, 2017).

Regarding bearing position, shoots bore heavier fruit compared to spurs except in the Koue Bokkeveld in 2018. The effect of light exposure on fruit mass was less evident in 2017, than in 2018 where the data showed sun exposed fruit was heavier than shaded fruit. This concurred with Wünsche and Lakso (2000) who found that increased light interception led to increased yields. Their study showed that individual parts of the tree that received more light had greater individual fruit mass. To address the effects of light distribution we refer to the review by Corelli-Grappadelli and Lakso (2007) that indicates that too much light overwhelms the photosystems and results in carbon assimilation inefficiencies and sunburn. They too comment on shaded portions of the tree that fix too little carbon for adequate fruit growth and promote research into optimising light distribution. In our study, the position of the fruit in the tree (top vs bottom branch) had a small influence on the individual fruit mass, but when considering the review by Corelli-Grappadelli and Lakso (2007), it is more likely that the light environment played a larger role in determining final fruit mass than tree position. In our study it can be qualitatively stated that the upper branches of the trees were exposed to more sunlight than the bottom branches (personal observation) but without the relevant measurements it cannot be quantified. Buler and Mika (2009) showed the light distribution in a solaxe training system, where mean illumination was over 3.7 times greater in the upper canopy compared to the lower.

3.1.3 Peel colour

Fruit peel colour, an external quality characteristic, is important across most fruit types. The pigments in the fruit that give rise to the perceived colour, are instrumental to the attractiveness of the fruit, as it is the first sensation apparent to consumers, and carries weight in governing their desire for the product (Leon *et al.,* 2006). Colour preference among consumers depends strongly on the cultivar in question. South African exporters are of the opinion that African consumers prefer a ‘Golden Delicious’ apple with a greener colour compared to Europeans who tolerate a yellower appearance (I. Adams 2021, personal communication, 22 November). It is generally accepted by South African producers that greener ‘Golden Delicious’ fruit fetch a higher market price in African countries compared to other markets that prefer yellower fruit.

Pathare *et al*. (2012) provides an excellent review on colour measurements in fresh and processed foods but only the following is pertinent to understanding the colour results presented in this paper:

- ‘Hue angle’ assigns a value (0-360) to the visual sensation of perceiving a colour, be it red, green, blue or a combination of them. All hue angle values were between 90 and 120, indicate that the dominant background colour is green (Little *et al.,* 1975). The lower the reported hue angle value the less green (yellower) fruit appears, and a higher hue angle value indicates a greener fruit. Thus, in the case of ‘Golden Delicious’ peel colour measurements a higher hue angle value denotes a greener (less yellow) colour.

- ‘Lightness’ measures how light or dark the colour is compared to a similarly illuminated white surface. - An object with a lightness value of zero would reflect no light and would appear be pure black, while an object with a lightness value of 100 (maximum) reflects all light and would be pure white. Hirst *et al*. (1990) said that increased lightness in fruit peel colour was perceived as yellower to the human eye. Duly, when interpreting ‘Golden Delicious’ peel colour, a lower lightness value will be considered as more preferred by the consumer. Hereafter, higher lightness is referred to as lighter green and lower lightness as darker green.
- ‘Chroma’, the quantitative attribute of colour, refers to the intensity or saturation of the hue angle. A higher chroma value denotes a brighter more vivid colour and lower chroma values are less vivid and almost faded to the eye. When it comes to describing ‘Golden Delicious’ peel colour, this parameter is not suitable to be used comparatively as it has no influence on the perception of yellow or green colour. Thus, only Hue angle and lightness will be discussed.

Fruit colour was measured and compared before and after harvest. It is generally expected that ‘Golden Delicious’ becomes yellower during storage (Blažek *et al*., 2003). Fruit at harvest that are yellower can be sorted or packed first to prevent further yellowing during storage (Cárdenas-Pérez *et al*., 2017). In most cases the results present below show statistically significant differences but may not be of practical (commercial) significance because the differences were very small and not likely to have an impact on the consumer. The difference between peel colour at harvest and after storage is commercially important because some pack houses “pre-sort fruit” based on colour using image and colour sensing technology (Zhang *et al*., 2014) and this sorting becomes futile if the colour change that occurs during storage is not related to colour at harvest.

Hue angle at harvest

When considering the variance component analysis, differences between orchards, trees, and position within the tree were the main sources of variation in 2017 contributing 12.9%, 9.9%, and 18.2%, respectively tot total variation. The unexplained hue angle variation was 57.5% and the remaining 1.5% was collectively caused by differences in region and the region x position interaction (Fig. 2). In 2018, the variation of hue angle at harvest was comparable to the previous season. The main causes of variation were again orchards, trees and position within the tree, contributing 19.7%, 9.5% and 13.7%, respectively. The unexplained variation was 57.1% and none of variation was caused by differences between region, trees nor the region x position interaction (Fig. 3).

Orchard-to-orchard variation was evident in the box plots of 2017 (Fig. 7a-b), yet in 2018, it would appear more homogenous barring a single orchard in Elgin that had much yellower fruit (Fig. 7c-d). It is likely that this lower performing orchard is responsible for the higher orchard-to-orchard variation reported in the 2018 season. In 2017, the interquartile ranges of the orchard fell between a hue angle of 110.21 and 114.53 and in 2018 it was between 110.50 and 114.54. The box plots were slightly negatively skewed in all the orchards. The first orchard in Elgin 2018 seems different from the other orchards from both regions. It displayed a wider range and its minimum and maximum are lower than the other orchards displaying more variability. Raese *et al*. (2007) showed that low nitrogen levels in 'Golden Delicious' can result in yellower fruit, and this could perhaps cause variability in fruit colour between orchards.

In 2017, region and tree position (RxTP) interacted significantly with a *p*-value of <0.0001 (Table 1a and 6). In both Elgin and the Koue Bokkeveld, the fruit from the bottom branch was greener than fruit from the top branch. For the respective tree positions, fruit from the Koue Bokkeveld were greener than those from Elgin. Looking at the F-values in Appendix A, Table 3, the TP main effect was the second largest contributor to changes in hue angle for both seasons tested. Region and bearing position (RxBP) interacted significantly (*p*=0.014; Table 1a and 7) in 2017. In the Koue Bokkeveld fruit borne on shoots were significantly greener than fruit borne on spurs, but in Elgin the hue angle was not affected by bearing position. Again, the regional effect was visible with fruit from the Koue Bokkeveld being greener than those from Elgin.

Light exposure (LE) is the largest contributor to changes in hue angle for both seasons. In Elgin the main effect of light exposure (LE) in 2017 was significant with a *p*-value of <0.0001 (Table 1a and 8). Shade fruit were greener than sun exposed fruit, which concurs with Ordóñez *et al*. (2016), who showed that 'Golden Delicious' fruit were greener under black shade netting.

In 2018, there were no interactions detected but the main effects of region (R) (*p*<0.0001; Table 1b and 13) tree position (TP) (p<0.0001; Table 1b and 15) and light exposure (LE) (p<0.0001; Table 1b and 16) were significant. Fruit from the Koue Bokkeveld were greener than fruit from Elgin. Fruit from the bottom branch were greener than fruit from the top branch and shaded fruit were greener than sun exposed fruit. The regional difference is of little commercial significance, but the differences that arise from tree position and light exposure may have implications. The difference is mainly due to light exposure and reducing the amount of light may result in greener fruit, as in the study on 'Granny Smith' by Fouché *et al*. (2010).

In summary, the hue angle after harvest in both seasons showed that the Koue Bokkeveld produced greener fruit compared to Elgin. Across regions, position within the tree and light exposure had a consistent effect. The lower branch had greener fruit than those from the upper branch, and shaded fruit were greener than sun exposed fruit. Fouché *et al*. (2010) showed that 'Granny Smith' fruit from the outer canopy were less green than the intermediate canopy, but that fruit from very shaded areas were also less green; although plant spacing is not mentioned in their study, it is apparent that they used trees on seedling rootstocks that typically grow vigorously and produce a larger canopy volume than the orchards in our study. This ties in well with these results where sun exposed fruit were yellower than shaded fruit as outer canopy fruit are sun exposed and inner canopy more shaded. These two effects (position within the tree and light exposure) are likely linked because light exposure on a bottom branch is typically less than the top branch. The correlation between these two factors is seen throughout this paper as fruit in the top part of the tree are more sun exposed and fruit in the bottom of the tree are more often shaded.

Hue angle after storage

In 2017, the variation of hue angle values after storage was primarily caused by orchard, tree, and fruit position within the tree, each contributing 27.8%, 11.7%, and 11.3%, respectively (Fig. 2). The unexplained variation was 46.5% and the remaining 2.8% of variation was caused by the region x position interaction. Region did not contribute to variation in hue angle after storage (Fig. 2). The main sources of variation the following season were also orchard, tree, and position differences, with contributions similar to 2017; 28.2%, 13.8%, and 11.2%, respectively. The unexplained variation was 46.8% but in 2018 position in the tree and the region x position interaction did not contribute to any variation in the hue angle (Fig. 3).

These results are similar to hue angle at harvest, but the orchard component had an increased contribution after storage. Since orchard differences increased during storage, it would be beneficial for orchards to be grouped at harvest so that yellower groups could be marketed earlier. The box plots (Fig. 8) show that the interquartile range of hue angle in 2017 ranged from 102.44 to 111.57 and in 2018 it ranged from 99.73 to 108.97. The first orchard in Elgin 2018 again, similar to the hue at harvest results, showed a wide box plot which also had the lowest hue angle.

In 2017, light exposure and bearing position (LExBP) interacted significantly (p = 0.0142; Table 1a and 4). For sun exposed fruit, fruit borne on shoots were greener than fruit borne on spurs. For shaded fruit, bearing position did not affect hue angle. Shaded fruit were greener than sun exposed fruit. Appendix A, Table 4 showed LE having the largest F-value, indicating that of the main effects it contributed most strongly to hue angle after storage in both seasons.

Fruit position in the tree and bearing position (TP x BP) interacted significantly in 2017 with a p-value of 0.0074 (Table 1a and 5). On the bottom branch, fruit borne on shoots were greener than fruit born on spurs, but on the top branch hue angle did not differ between bearing positions. Fruit from the bottom branch were greener than fruit on the top branch.

In 2017, region and tree position (RxTP) interacted significantly with a p-value of <0.0001 (Table 1a and 6). In both Elgin and the Koue Bokkeveld, fruit from the bottom branch were greener and the difference in hue angle between the fruit from different tree positions was greater in the Koue Bokkeveld than in Elgin.

In 2018 with a p-value of <0.0001, tree position (TP) had a significant effect (Table 1b and 15). Fruit from the bottom branch were greener than fruit from the top branch.

The effect of light exposure (LE) was significant with a p-value of <0.0001 (Table 16). Shade fruit were greener than sun exposed fruit. This was in line with the 2017 season where shaded fruit were greener too. Likewise, the effect of tree position on fruit colour was consistent between seasons where lower branches had greener fruit.

The results were consistent when comparing the hue angle before and after harvest. After storage the fruit were notably yellower than before. What was evident was the difference in greenness between fruit from the top and bottom branches, where fruit from the Koue Bokkeveld showed a larger difference between tree positions than fruit from Elgin. The canopy and microclimatic effect on peel colour is described by Fouché *et al*. (2010) on 'Granny Smith' peel colour, where less light and lower temperature in the canopy produced greener fruit and those directly exposed to the sun could be bleached but does nothing to explain the difference between the regions. It is important to note that Fouché found that paler apples were produced from the most shaded portions of the tree.

Lightness at harvest

Orchard and tree differences were the greatest contributors to the lightness variation at harvest in 2017, contributing 28.2% and 15.5%, respectively. The unexplained variation was 45.8% and the remaining 6.1% of variation was collectively caused by differences in position and the region x position interaction. Region did not contribute to variation in lightness at harvest (Fig. 2).

The contribution of the variance components changed in 2018. The primary sources of variation now also included the position in the tree compared to 2017. The contributing values were 20.3%, 12.6% and 9.6%, respectively for orchard, tree, and position. The unexplained variation was higher in 2018 (57.1%) with a negligible combined contribution (0.4 %) from growing region and the region x position interaction (Fig. 3).

Box plots were used again to illustrate the large contribution of orchard differences (Fig. 9.). The interquartile range of lightness in 2017 was from 68.11 to 76.11 and in 2018 it ranged from 69.58 to 76.96. It appears that there were greater differences among orchards in 2017 than in 2018. The exception here again is the single orchard in Elgin that had markedly higher lightness.

In 2017, the interaction between region and tree position (RxTP) was significant (*p* <0.0001; Table1a and 6). Fruit from the top branch in the Koue Bokkeveld were lighter than fruit from the lower branch, this pattern was not present in in the Elgin region.

In the first season region and bearing position (RxBP) interacted significantly with a *p*-value of 0.006 (Table 1a and 7). Across regions fruit borne on spurs were lighter than fruit borne on shoots. Regional differences were only seen where fruit were borne on shoots in Elgin which were lighter than fruit borne on shoots in the Koue Bokkeveld, but at a commercially insignificant margin.

In 2017, the interaction between region and light exposure (RxLE) was significant with a *p*-value of 0.014 (Table 1a and 11). In both regions sun exposed fruit were lighter in colour than shaded fruit. Sun exposed fruit from Elgin were lighter than fruit from the Koue Bokkeveld.

The second season saw main effects region, tree position, light exposure and bearing position significant with *p*-values of <0.0001 but none of the interactions were significant. (Table 1b). Fruit from Elgin were lighter in colour than in the Koue Bokkeveld (Table 13). Tree position showed the upper branches having lighter fruit (Table 16). Sun exposed fruit were lighter than the shaded fruit (Table 16). It is probable that fruit from the upper canopy are more likely to be sun exposed, partially explaining why tree position had an effect on lightness. Bearing position showed fruit borne on spurs being lighter than those borne on shoots (Table 18).

Regionally the first season showed some aspects where Elgin fruit were lighter than fruit from the Koue Bokkeveld, and in 2018 Elgin showed overall to have lighter fruit than the Koue Bokkeveld. Across both seasons and regions, fruit from the upper branches were lighter than fruit from the lower branches. It is also seen, across seasons, that sun exposed fruit were lighter than shaded fruit. Fouché *et al*. (2010) showed outer canopy 'Granny Smith' fruit being lighter than the inner canopy. Inner and outer canopy differences were accounted for by light conditions. Here the difference between the upper and lower branch as well as the light exposure of fruit reacted similarly. Interestingly, bearing position had a consistent effect on lightness that was not present in the other colour parameters. Fruit borne on spurs were lighter than fruit born on shoots. Literature on this effect could not be found, but it is known that higher nitrogen levels produce greener fruit (Hansen, 1980) and perhaps it can be explained with nutrient supply to the fruit through the different shoot types.

Lightness after storage.

Variation of the lightness values after storage in 2017 was primarily affected by orchard and tree differences, contributing 28.2% and 15.5%, respectively. The unexplained variation was 45.8% and the remaining 10.5% of variation was collectively caused by differences in position and the region x position interaction. Again, region did not contribute to the variation in lightness after storage (Fig. 2).

The largest contributors did not change in the second season and the variation of lightness after storage was again predominantly affected by orchard and tree differences, contributing 24.1% and 11.9%, respectively. The unexplained variation was 60.1% and the remaining 4.0% of variation was caused by differences in position. Region and the region x position interaction did not contribute to variation in lightness at harvest (Fig. 3).

The box plots of lightness values after storage indicate variability between the orchards (Fig. 10). The interquartile range of lightness in 2017 was from 69.65 to 78.83 and in 2018 it was higher at 71.67 to 80.93 (Fig. 10). The ranges are wider than what was seen for the fruit at harvest (Fig. 9). It appeared that overall variability of lightness within orchards increased after storage. The ratio of chlorophyll to carotenoids in 'Golden Delicious' peel determines the peel colour and as the fruit ripens, chlorophyll levels drop while carotenoids remain the same, resulting in the change in appearance from green to yellow (Workman, 1963). Knee (1972) observed a similar reduction in chlorophyll in 'Cox orange Pippin' as fruit matured but here carotenoids also initially began to decline until the climacteric where after it increased. Kuckenberg (2008) showed that there was a lower level of chlorophyll in the sunlit cheek of a 'Golden Delicious' apple than that of the shaded cheek. The increase in variability in colour after storage could be due to the lower level of chlorophyll on the sun exposed apples only being apparent once it has begun to break down.

In 2017 region and tree position (RxTP) interacted significantly with a *p*-value of <0.0001 (Table 1a and 6). In Elgin, fruit from the bottom branch were lighter in colour than those from the top branch. In the Koue Bokkeveld fruit from the top branch were lighter in colour than those from the bottom branch. The difference in lightness between the tree positions was greater in the Koue Bokkeveld than in Elgin. In 2017, region and bearing position (RxBP) interacted significantly (*p*=0.012; Table 1a and 7). Across regions, fruit borne on spurs were lighter in colour than fruit borne on shoots. Regional differences were only seen where fruit were borne on spurs in Elgin that were lighter in colour than fruit borne on spurs in the Koue Bokkeveld. As was the case before storage, the main effect of light exposure (LE) had a significant effect with sun exposed fruit being lighter in colour than shaded fruit after storage (*p*-value <0.0001; Table 1a and 8).

In 2018, the main effects of tree position (TP) and light exposure (LE) were significant (Table 1b). Fruit from the top branch were lighter in colour than fruit from the bottom branch (*p*= 0.0113; Table 1b and 15) and sun exposed fruit were lighter than shaded fruit (*p*= 0.002; Table 1b and 16).

The results for lightness after storage were comparable to lightness at harvest regarding the effect of tree position and light exposure. In both seasons light exposure showed sun exposed fruit being lighter in colour than shaded fruit. It is also seen that, similar to at harvest, the fruit borne on shoots were lighter in colour than fruit borne on spurs. This was an interesting find but literature relating to this this could not be found. A difference between at harvest and after storage was noted regarding tree position. It appeared that there was greater variation in lightness after storage than at harvest. After storage the Koue Bokkeveld fruit from the top branch were lighter than fruit from the lower branch which is consistent with the results at harvest. An inconsistency arose with fruit from the bottom branch in Elgin which were lighter than fruit from the top branch, whereas the results at harvest showed no difference between the fruit from the top and bottom branches in Elgin. Due to milder winter conditions in Elgin, trees are more basal dominant than in the Koue Bokkeveld (Cook and Jacobs, 1999) and it is likely that with a more conical shaped tree, instead of columnar, more light would be present at the bottom of the canopy in Elgin than in the Koue Bokkeveld, but this would not explain why this difference disappeared after storage. Stage of fruit maturation does effect fruit colour due to chlorophyll degradation in the fruit peel as maturity advances (Knee, 1972), and this evidence of higher lightness of fruit peel colour in the lower branches in Elgin initially appeared in part to be a matter of maturity. The idea is later invalidated where it was seen that the fruit from lower branches in the Koue Bokkeveld were the most advanced in maturity but the effect was not present there. This lends more credence to canopy structure and light exposure as chief driving force of peel lightness.

3.1.4 Sunburn

Sunburn is well described in literature and its origin is predominantly metrological, and the exposure of susceptible fruit peel to inducing conditions namely high temperatures and irradiance (Piskolczi *et al*., 2004; Racsko and Schrader, 2012). Sunburn is a well understood phenomenon and common in 'Golden Delicious' produced under South African summer conditions. Duly a "sun exposed" category was included in this trial, where it was expected to give high sunburn results compared to the other categories. This can also serve as a quality check for our methods and statistical analysis. It was expected that only sun exposed fruit had sunburn, and that the upper branch in the tree (being more exposed to the sun) also showing more sunburn. Differences between the regions could be expected if summer temperatures were markedly different during the susceptible times.

Our 2017 sunburn results show that variation was primarily affected by fruit position and accounted for 40.1% of the variation. The unexplained variation was 50.1% and the remaining 9.8% was collectively caused by differences among orchards, trees, and the region x position interaction. Regional differences did not contribute to variation in sunburn (Fig. 2).

A similar trend is seen in 2018, with fruit position being the largest contributor (37.0%) The unexplained variation was 52.0% and the remaining 11.0% of variation was collectively caused by differences across regions, orchards, trees, and the region x position interaction (Fig. 3).

In 2017, the interaction between region, tree position and light exposure (RxTPxLE) was significant (*p*=0.0033; Table 1a and 2). Sun exposed fruit within both tree positions and regions had more sunburn than shaded fruit. As expected, the top branches showed more sunburn than the bottom branches in both regions. Sun exposed fruit from the bottom branch in Elgin, had more sunburn than sun exposed fruit from the bottom branch in the Koue Bokkeveld. Again, the basal dominance in Elgin referred to earlier, was likely a factor here, with more light in the lower canopy that in the Koue Bokkeveld (Cook and Jacobs, 1999). Bearing position had a significant effect on sunburn with a *p*-value of 0.049 (Table 1a and 10), but this is only just within the 5% significance level. Fruit borne on shoots showed more sunburn than fruit borne on spurs.

In 2018 we found a significant four-way interaction: region, tree position, light exposure, and bearing position (RxTPxLExBP) (*p*= 0.034; Table 1b and 20). Sunburn, regarding bearing position within light exposure, tree position and region did not differ, except for fruit on shoots from the Koue Bokkeveld, bottom sun exposed fruit had more sunburn than fruit on spurs. As expected, sunburn due to light exposure consistently showed more sunburn on sun exposed fruit, except in the Koue Bokkeveld where fruit from sun exposed spurs on the bottom branch did not differ from shaded spurs on the bottom branch. Regarding tree position, the greatest degree of sunburn in Elgin was on the top branch and the greatest degree of sunburn in the Koue Bokkeveld was on the bottom branch.

As anticipated, position (TP LE and BP) accounted for the largest amount of variation in both seasons but of these, light exposure contributed most when looking at the high F-values in both seasons (1595.28 in 2017 and 386.24 in 2018; Appendix A Table7). Fruit position in the tree also played a role because of its interaction with light exposure where the upper branch is exposed to sunlight for a longer period during the day. Of interest is the effect of bearing position where shoots showed more sunburn than spurs. This is likely due to the weight of the apple moving the shoot into the sun as its mass increased during the growing season. Due to this, fruit may also shift from a shaded position earlier in the growing season to a sun exposed position when it is heavier later in the season. Fruit that is not acclimated to sunlight is more susceptible to sun damage (Chen *et al*., 2008).

3.2 Maturity parameters

3.2.1 Starch conversion percentage

As apple fruit ripen and start to mature, the accumulated starch in the flesh is hydrolysed to sugar (Krotkov and Helson, 1946). This conversion is used as an indication of fruit maturity with low starch/high sugar deemed more mature.

Differences across orchards and trees were the largest contributors to overall variation in starch conversion (SC%) in 2017, contributing 21.3% and 28.5%, respectively. The unexplained variation was 43.0% and the remaining 7.2% of variation was collectively caused by differences in position and the region x position interaction. Region did not contribute to overall variation in SC% (Fig. 2).

Less of the variation was accounted for in the second season. The three main contributors of variation in 2018 were orchards, trees and the region x position interaction, contributing 10.6%, 8.8%, and 9.1%, respectively. The unexplained variation was higher at 68.9%, and the remaining 2.6% of variation was caused by positional differences. Region, again, did not contribute to overall variation in SC% (Fig. 3).

In both seasons the differences among the orchards were stark (Fig. 11a and b). Not only was there high variation among the orchards but within orchard a large proportion of fruit ranged from below to very high above SC% specification of 15% at harvest. The orchards with a mean SC% above 50% were included in the box plots to illustrate the variation in harvest maturity but were excluded from further statistical analysis to avoid the use of over “mature” orchards. The 2018 season showed more condensed ranges for the box plots (Fig. 11b) compared to the 2017 results. These orchards were harvested closer to optimum than in the previous season, reducing to some extent the variation due to orchard differences (Fig. 1; Fig. 2). Growers use SC% to aid in harvesting at the correct maturity, but the inherent variability in SC% makes it challenging to do so (Smith *et al*., 1979).

In 2017, the interaction between region, tree position and light exposure (RxTPxLE) was significant (*p*=0.040; Table 1a and 2). In the Koue Bokkeveld, fruit from the top branches had a lower SC% percentage than the bottom branches. In Elgin the role of tree position was less clear; shaded fruit followed the same pattern with the top-shaded fruit having a lower SC% than the bottom-shaded fruit, but sun exposed fruit did not differ significantly between the tree positions (*p*=0.040; Table 2).

In Elgin, sun exposed fruit had a lower mean SC% than shaded fruit and this is consistent regardless of tree position (*p*=0.040; Table 2). In the Koue Bokkeveld however, tree position interacted differently with light exposure than in Elgin. Sun exposed fruit from the top branches in the Koue Bokkeveld had a lower starch conversion percentage than shaded fruit, and on the bottom branches in the Koue Bokkeveld sun exposed fruit had a higher SC% than shaded fruit (*p*=0.040; Table 2).

The effect of bearing position, in 2017, was statistically significant (*p*=0.001) but the SC% across shoots and spurs do not differ significantly (Table 1a and 10). In 2018 the interaction between region and tree position (RxTP) was significant (*p*<0.0001; Table 1b and 17). Both regions showed a lower mean SC% in the top branches than the bottom branches, but the difference between the top and the bottom branches was significantly larger (*p*<0.0001; Table 17) in the Koue Bokkeveld than in Elgin. Region also interacted with light exposure (RxLE) with sun exposed fruit from the Koue Bokkeveld showing a lower starch conversion percentage than shaded fruit, but in Elgin sun exposed and shaded fruit did not differ (*p*<0.0001, Table 1b and 19).

Less of the overall variation was explained in the second season and some effects appeared to be more consistent across seasons than others. The differences between tree positions were relatively consistent, with fruit from the upper branches showing lower starch conversion than in the lower branches. In the 2017 season the multilevel interaction (RxTPxLE) confounded the tree position effect in Elgin. However, the effect of tree position in the 2018 was clearer where it showed a significant difference between the top and bottom branches in the Koue Bokkeveld than in Elgin. Light exposure was less determinate, but fruit generally showed lower SC% in sun exposed fruit than shaded fruit in contrast to Ju *et al*. (1999) who found that 'Fuji' apples did not differ in maturity due to light environment. This aspect may be cultivar specific, as postulated by Drogoudi and Pantelidid (2011), and 'Golden Delicious' maturation may be more affected by light exposure. The extent of the sensitivity to light exposure is however questionable as exceptions were present in this study; the maturity of the fruit on the lower branches in the Koue Bokkeveld in 2017 and Elgin in 2018 seemed unaffected by the light exposure. As the lower branches of the tree received less light compared to the upper branches, it is possible that the difference in light received between sun exposed and shaded fruit from the lower branches was smaller than that of the upper branches. It is reasonable to assume that where the difference between sun exposed and shaded fruit is smaller, as in the lower branches, the effects observed would be weaker.

Noting the difference in SC% between canopy positions, it would be practically feasible to do separate starch tests for the top and bottom of the tree and pick accordingly. This would allow for greater uniformity of stored fruit. Already, some growers pick the bottom fruit from two successive orchards without ladders and workers return for the fruit in the top at a later stage (usually within 7 days). The differences seen between shaded and sun exposed fruit could be addressed by planting higher density orchards with a more uniform light environment. Higher density planting is not the only solution, and as Robinson *et al*. (1991) explained, tree canopy structure in low density plantings, can be manipulated to allow for adequate light distribution. He too noted that with higher density plantings less time and expertise is required to achieve the same light conditions.

Blanpied and Silsby (1992) highlighted the drawbacks of the use of a starch index as an indicator of maturity stating that apples from lightly cropped trees may falsely test as being immature while on a heavily cropped tree the starch iodine test will incorrectly indicate an inflated maturity. The test can also be inaccurate because starch in the fruit core area is consistent with an immature fruit but the absence thereof is not necessarily an indication of maturity (Krotkov and Helson, 1946) and thus staining patterns can be misinterpreted if fully immature fruit are present in a sample. Despite the inherent flaws in using SC% as an indicator of picking maturity, the ease of field application and low cost of the test drives its current usage. Further work to use hyper-spectral imaging analysis aims to remove the subjectivity of human evaluation and refine the measurability of this parameter (Menesatti *et al*., 2009).

3.2.2 Firmness

Fruit flesh firmness is used together with SC% as an indicator of fruit maturity across a wide range of fruit crops, but especially in pome and stone fruits (Sams, 1999). South African apple producers use firmness (in conjunction with starch conversion percentage) to determine the commercial harvest date of an orchard. It also correlates well with eating satisfaction and is thus a useful measure of both maturity and quality (Wills *et al.,* 1980). Fruit firmness has an impact on future eating quality and storage ability of apples making uniformity in this parameter at harvest and after storage a crucial aspect of apple production (Sass *et al*., 1992).

Firmness at harvest

In 2017, the variation in firmness at harvest was mainly caused by orchard and tree differences, 5.8% and 10.8%, respectively. The unexplained variation was 80.9% and the remaining 2.5% of variation was collectively caused by differences in region, positions, and the region x position interaction (Fig. 2). The second season had less unexplained variation (61.2% in 2018). Orchards, and trees contributed 25.9% and 9.9%, respectively. The remaining 2% of variation was caused by differences across fruit positions. Region and the region x position interaction did not contribute to variation in firmness at harvest (Fig. 3). A large amount of the explained variation originates from orchards and tree differences and little is explained by the other factors making it important to quantify and describe these effects. Fruit firmness is not a direct physiological trait and depends on cellular properties such as cell size, and the bonding between cells and cell walls (Harker *et al*., 1997). The complexity behind fruit firmness is likely why the high unexplained variation is present in both seasons.

The box plots in Fig. 12 illustrate the variation of orchards. Both seasons saw a similar interquartile range: 6.98 to 8.90kg in 2017 and 6.66 to 8.93kg on 2018. In 2017 the orchard with the highest mean firmness in Elgin was 8.40 kg and the lowest 7.44 kg (0.94 kg difference), where in the Koue Bokkeveld the highest was 8.03 kg and the lowest 7.51 kg (0.52 kg difference; Fig.12). The same is seen in 2018, where the differences between the highest and lowest firmness in Elgin is 1.42 kg and in the Koue Bokkeveld 0.66 kg (Fig.12) Orchards appeared to differ more from one another in 2018 than in 2017 (Fig. 12).

In 2017, the interaction between region and tree position (RxTP) was significant with a *p*-value of 0.003 (Table 1a and 6). In the Koue Bokkeveld, tree position did not affect fruit firmness, but in Elgin fruit from the top branches showed a greater firmness than fruit from the bottom branches. The effect of TP on firmness, present in Elgin but not the Koue Bokkeveld, is perhaps because firmness is not purely related to maturity. Fruit size has an influence; Blanpied *et al*. (1978) showed a strong negative correlation between fruit mass and fruit firmness. This explains the result here when noting that the fruit from the upper branches in Elgin had smaller fruit than fruit from the bottom branches. The main effect of bearing position was significant and showed that fruit borne on shoots were firmer than fruit borne on spurs ($p<0.0001$; Table 1a and 10).

In 2018, the interaction between region and bearing position (RxBP) was significant ($p= 0.030$; Table 1b and 14). In Elgin there were no differences between fruit borne on shoots and fruit borne on spurs and the results also did not differ from spur borne fruit in the Koue Bokkeveld. In the Koue Bokkeveld, fruit borne on shoots were significantly firmer than those on spurs (Table 14). BP was the driver in this interaction with an F-value of 11.45 compared to the F-value of region, 0.51 (Appendix A, Table 9). Tree position (TP) effected fruit firmness significantly with fruit from the top branches were firmer than fruit from the lower branches ($p= 0.004$; Table 1b and 15). This indicates more mature fruit from the lower branches as seen with SC% results, adding evidence to ripening differences between the top and bottom branch. The difference is commercially significant, because if the fruit firmness for ‘Golden Delicious” drops below 5.4 kg, it is rejected for export (DAFF, 2013). Even a marginal increase in fruit firmness can have substantial commercial consequence.

Sams (1999) discussed numerous factors that influence fruit firmness, ranging from genetic, environmental, cultural and physiological. Shifting focus to environmental causes, Blanpied *et al*. (1978) stated that sun exposed fruit had a greater firmness than shaded fruit, likewise Klein *et al*. (2001) showed that firmness was higher in sun exposed fruit.

Firmness after storage.

Besides the unexplained variation of 64.9% in 2017, the variation in firmness after storage was affected by orchards and trees, contributing 12.1% and 15.7%, respectively. The remaining 7.3% of variation was caused by differences across positions. Region and the region x position interaction did not contribute to the total variation in firmness after storage (Fig. 2). As in the previous season, the variation in firmness after storage in 2018 was mainly affected by orchards and trees, contributing 38.5% and 7.5%, respectively. The unexplained variation was 46.4% and the remaining 7.6% of variation was collectively caused by differences across positions and the region x position interaction. Region did not contribute to variation in firmness at harvest (Fig. 3).

When considering box plots to investigate the variation brought about by orchards (Fig.13), the interquartile range was 4.02 to 6.13 kg in 2017 and 4.01 to 6.51 kg in 2018. A clearer difference was seen again when looking at the range from the highest mean firmness in a region to the lowest mean firmness. In 2017, the means in Elgin differed by 1.20 kg, where in the Koue Bokkeveld it was less with 1.00 kg (Fig. 13). In 2018, the differences were still larger in Elgin (1.42 kg) compared to the Koue Bokkeveld (1.17 kg) and larger than the previous season. These differences between highest and lowest firmness increased from harvest to after storage, suggesting that variation was higher after storage than at harvest (Fig. 12 and 13).

In 2017, tree position and light exposure (TPxLE) interacted significantly with a *p*-value of 0.039 (Table 1a and 9). In the top branches, sun exposed fruit were firmer than shaded fruit and the bottom branches showed a similar result with sun exposed fruit again being firmer than shaded fruit. LE, with an F-value of 100.95, had a larger effect on firmness than TP with an F-value of 59.94 (Appendix A, Table 10). Bearing position and region were present as main effects, with a *p*-values of <0.0001 and 0.048 respectively (Table 1a). Fruit borne on shoots were firmer than fruit borne on spurs (Table 10). After storage fruit were firmer in Koue Bokkeveld than in Elgin (Table 12).

In 2018, region and tree position (RxTP) interacted significantly with a *p*-value of 0.008 (Table 1b and 17). In the Koue Bokkeveld, fruit from the top branches were firmer than fruit from the bottom branches but in Elgin the tree position did not affect fruit firmness. The difference seen in the Koue Bokkeveld (0.49 kg) is a fair margin, and is larger than the difference in firmness at harvest over both regions (0.23 kg; Table 15). It is possible that the difference is magnified after storage, suggesting that even if a grower detects a small difference in fruit firmness between the top and bottom of the tree, it may be beneficial to segregate the fruit from the two positions during harvest. The tree position and light exposure (TPxLE) interaction showed that fruit from both tree positions were firmer when sun exposed than shaded (*p*=0.022; Table 21). The shaded fruit from the bottom branch was significantly less firm than shaded fruit from the top branch (*p*=0.022; Table 1b and 21). Bearing position had a significant effect and fruit borne on shoots were firmer than fruit borne on spurs (*p*=0.0003; Table 1b and 18).

The same factors (region, tree position, and bearing position) as seen in firmness at harvest, also effected the fruit firmness after storage. Tree position also showed firmer fruit in the upper branch but it was less clear than at harvest. This is likely explained by the effect of light exposure on fruit firmness which showed that sun exposed fruit were firmer than shaded fruit. Klein *et al*. (2001) showed a difference between sun exposed and shaded fruit firmness, but the observed effects in their study disappeared after 10 days of shelf life. The shelf life in our study was shorter (7 days), and is perhaps why we could still observe the difference in firmness after storage. This effect of LE seems only to have manifested after storage but the effect of TP was present at harvest, where one expects more sunlight in the upper canopy, may have hidden the effect of light exposure prior to storage. Furthermore, it would appear that the effect of light exposure modifies the storage potential of the fruit since its effect is stronger after storage than before. Kuckenberg *et al*. (2008) showed that for a sun exposed fruit, the firmness did not differ between the shaded and sunlit cheek indicating a more uniform firmness across the fruit. It can be deduced from the results that sunlit fruit, may possibly have better storage potential than shaded fruit, especially because the difference only manifested after storage.

3.4 Correlations between parameters

Many of the appearance and maturity parameters correlated with one another, with significant correlations shown in bold in Table 22 and 23. Although many correlations were significant only stronger and commercially relevant correlations were discussed. A strong negative correlation (r=-0.62) was found in 2018 between fruit mass and fruit firmness, both before and after harvest (Table 23). In 2017 the correlation between mass and firmness at harvest was moderately negative (r=-0.47), and the correlation between mass after storage firmness was weakly negative (r=-0.34; Table 22). De Salvador *et al*. (2006) found weak correlations between mass and other quality parameters, except for size dimensions arguing that larger fruit have more mass. Blanpied (1978) noted that a negative correlation existed between fruit size and fruit firmness but did not provide any possible reasons for this. Johnston *et al*. (2002) suggested that smaller fruit possibly ripened slower than larger fruit. Saei *et al*. (2011) noted that the correlation between size and firmness was only present when fruit were harvested at advanced maturity but they showed that dry matter concentration was a better indication of firmness regardless of maturity.

Strong correlations were present among the colour parameters both at harvest and after storage (Table 22 and 23). Comparing colour from at harvest to after storage, hue angle at harvest had a strong positive correlation with hue angle after storage in both seasons (r=0.71 in 2017; r=0.77 in 2018). Lightness too showed strong positive correlations from harvest to after storage for both years (r=0.84 in 2017; r=0.74 in 2018). Li *et al*. (2009) described a method to combine colour measurements computationally, using only hue angle and lightness and terms it “value”, for the purpose of sorting fruit. Our results also confirmed that sorting fruit appearance based on colour in a pack house is useful, but addressing variation, in starch conversion and fruit firmness, should be done at tree level and could differ across production regions.

Moderate to weak correlations were present between maturity and colour parameters (Table 22 and 23), and although it is already a standard practise for apple pack houses to use image and colour sensing technology to automate packing in most fruit and vegetables (Zhang at al., 2014), this would only be sorting for appearance and not necessarily internal/eating quality. The effect of light exposure appears to be a stronger driver in fruit colour than that of maturity. It may be that if the light environment fruit were exposed to was more uniform, the effect of light on colour would be removed and a stronger correlation between colour and maturity would be present. Future research on this could prove useful. Although not ubiquitous in application, the use of hyperspectral imaging can detect bruises, and determine firmness and total soluble solids (TSS) in apple fruit (Wang *et al*., 2015). A method described by Mendoza *et al*. (2011) provides an acceptable level of accuracy for sorting 'Golden Delicious' among others, but it cannot yet be implemented in real time.

Sunburn and hue angle at harvest had a strong negative correlation in 2017 (r =-.067) and a moderately negative correlation in 2018 (r=-0.49). Shade netting can be used to reduce the amount of sunlight reaching the fruit, and Ordóñez *et al*. (2016) detailed the effect of black coloured net in producing greener fruit. They suggested that not only was chlorophyll degradation slowed, maturation was also delayed by the net further deferring yellow colour development. In a low light environment carotenoid production, a product of photo-protective processes in the skin, will be reduced and duly yellow colour development will be retarded (Demmig-Adams, 1990).

The correlation between starch conversion and firmness at harvest (r=-0.29) in 2017 was weakly negative (Table 22). In 2018 there was a stronger correlation between starch conversion and firmness before (moderate negative: r=-0.48) and after (moderate negative: r= -0.42) storage (Table 23). This serves to show little agreement between starch conversion and fruit firmness as maturity parameters. Previous research done by Knee and Smith (1989) showed a correlation between apple firmness and maturity, with firmer fruit being less mature, but the inconsistencies of using SC% as a maturity index limits its utility.

The correlation between firmness at harvest and firmness after storage was moderate and positive in both seasons (r=0.40 in 2017; r=0.59 in 2018), but relatively stronger in the second season (Table 22; Table 23). Firmness of fruit at harvest translates well through storage and it is current practise to store batches of fruit with the highest firmness for the latest packing date. The challenge that remains is that when fruit firmness is inconsistent within a batch and it is stored according to the mean fruit firmness, high variability will be retained throughout storage making mean quality parameters less reliable.

3.3 Tree-to-tree effect

Tree-to-tree variation accounted for a notable portion of variation for most variables measured in both seasons and was highly significant for all variables except for sunburn in 2018 (Table 1c). The only parameter measured where this was not significant was sunburn. This was unexpected as precision horticultural studies done by Aggelopoulou *at al.* (2010) mapped the yield and quality of fruit in an orchard, and while a great deal of variation in yield from tree-to-tree was recorded, they found little variation in fruit quality.

Even though rootstocks could be standardised for this paper, it was not possible to standardise soil conditions. It is generally accepted that soil in the Western Cape is highly variable across a small area, making soil heterogeneity, even within a relatively small orchard, a reality. Umali *et al*. (2012) focused on mitigating the effect of soil variation in apple orchards, emphasising intensive soil sampling to find "pockets or zones" within an orchard that require additional management or amelioration. If the nutritional or mineral relationship with a parameter of interest, such as firmness, is not known, it might be possible to identify the problem areas in an orchard, but the remedial action remains elusive.

Some simple orchard establishment errors can also contribute to variability, such as incorrect planting depth. Care should be taken during planting to ensure that trees are not buried above the rootstock-scion graft union as the variability caused by scion rooting is underestimated. This negates the effect of the rootstocks and can introduce large variation that could be easily avoided. Our study recommends that the surprisingly high tree-to-tree variation in all the tested quality parameters should be addressed through dedicated research using a precision agriculture approach and modern technology.

The high variation in maturity complicates the sampling of fruit in determining harvest date. Typically, growers only sample ten fruit from an orchard and often it is not a representative sample. Increasing the sample size and deliberately sampling from different positions (top, bottom, sun and shade), would provide a more accurate indication of orchard maturity and if a clear pattern emerges, the orchard could be harvested accordingly. This sampling should be extended beyond ‘Golden Delicious’ to full red and green cultivars where picking on ground colour is not possible.

4. Conclusion

Light exposure is likely to be the major external factor that drives variability within the canopy. It often interacted with the other factors, especially bearing position and position within a tree (top vs. bottom). Fruit firmness after storage was higher for sun exposed fruit, confirming that adequate light is important for the development of high-quality fruit. Two-dimensional canopy structures or slender spindle could create a more favourable light environment. A flatter canopy or “fruiting wall” would better light distribution and reduce the variability caused by the fruit’s position within the tree. Modern planting systems present much simpler canopies that have additional benefits, such as easier mechanisation. There exist rootstock evaluation sites from studies in the previous decades that could be used to determine their influence on uniformity in quality and rate of maturation.

Variation of starch conversion within an orchard was excessive irrespective of region, with most orchards having relatively high portions of fruit outside of the optimum harvest standard. This warrants an investigation into the sampling of different zones of a tree to determine if a more selective picking strategy, appose to the “single-strip” harvest approach currently in practice, could have advantages when harvesting ‘Golden Delicious’. Where large differences in maturity exist between the top and bottom canopy, an initial pick can be performed without ladders, thus in the bottom of the tree first. Once the maturity in the upper canopy has advanced, a second pass through the orchard can be made with ladders/platforms. Another way to reduce variation would be to use 1-methyl-cyclopropene (1-MCP) before harvest. Varanasi *et al*. (2013) showed that critically timed (applied at 30% starch conversion) were the most effective. Greene (2005) evaluated the timing of aminoethoxyvinylglycine (AVG) treatment to elicit the best response in terms of delaying maturation.

Evidence exists for both compounds to retard ripening but effect different pathways. Perhaps if both were used, albeit at high cost, it would result in a far more uniform crop.

Because of the contrasting winter conditions, it was expected that growing region would have a notable influence on the quality parameters, but surprisingly, only appears to have influenced fruit aspect and mass. In fact, orchard variation, followed by tree-to-tree variation, was the greatest contributor to the total variability in both seasons. The position of fruit inside a tree had a marked influenced on colour (hue angle and lightness) but also had some influence on the starch conversion percentage and fruit firmness. Perhaps difference in flowering time between top and bottom branches is one of the causal elements in the difference observed. It is known for South African apple production sites for the top of the tree to bloom a few days after the bottom. This can be addressed by modifying rest breaking treatments to target the top and bottom at different time interval to increase the dosage to the upper portion of the tree.

To address the variation in practice, the unfavourable macroclimatic effects cannot be mitigated directly, barring the use of low chill cultivars. Bearing position is related to genetics but understanding a cultivar’s natural bearing habit and architecture can assist in custom pruning practices to exploit/mitigate certain traits. For instance, ‘Golden Delicious’ is a tip bearer and pruning strategies aim to maximise the number of fruit borne on short shoots. Light exposure is likely easier to confront. A more uniform light environment would remove some of the variation seen from differences in shaded and sun exposed fruit. This can potentially be done by planting higher density orchards or by more aggressive light management in existing lower density orchards. The most effective way would be to address it when planning future orchards than to ameliorate existing trees. Currently there is a drive to replant a greater proportion, 8-10% than the standard of 3-5% per annum.

The minor regional differences observed suggest that although a difference in chill accumulation was present between the two regions, it may be that it did not result in a more condensed bloom in the Koue Bokkeveld. It would be beneficial to further investigate the effects of the elements that were standardised in this paper, i.e., row orientation, planting distance, rootstock, and cultivar. Additionally, information on the effect of nets on variation in fruit quality should also be considered.

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Tables

Table 1a: Statistically significant ANOVA results (main effects and/or highest order interactions) for the fruit maturity and quality variables for the **2017** season with reference to the individual ANOVA tables (*p*=0.05). Region (**R**), Tree Position (**TP**), Bearing Position (**BP**), Light Exposure (**LE**).

Variable	Source of variability	F value	*p*-value	ANOVA Tables
Fruit aspect	R x TP x LE	3.98	0.047	Table 2
	R x TP x BP	4.78	0.029	Table 3
	LE x BP	8.83	0.003	Table 4
Mass	R x TP x LE	6.54	0.011	Table 2
	TP x BP	3.87	0.050	Table 5
Hue-angle at harvest	R x TP	17.99	<0.0001	Table 6
	R x BP	6.06	0.014	Table 7
	LE	521.91	<0.0001	Table 8
Hue-angle after storage	LE x BP	6.09	0.014	Table 4
	TP x BP	7.28	0.007	Table 5
	R x TP	37.38	<0.0001	Table 6
Lightness at harvest	R x TP	79.60	<0.0001	Table 6
	R x BP	7.55	0.006	Table 7
	R x LE	6.06	0.014	Table 11
Lightness after storage	R x TP	46.90	<0.0001	Table 6
	R x BP	6.37	0.012	Table 7
	LE	21.33	<0.0001	Table 8
Sunburn	R x TP x LE	8.68	0.003	Table 2
	BP	3.90	0.049	Table 10
Starch conversion	R x TP x LE	4.27	0.040	Table 2
	BP	10.37	0.001	Table 10
Firmness at harvest	R x TP	9.36	0.002	Table 6
	BP	19.48	<0.0001	Table 10
Firmness after storage	TP x LE	4.31	0.039	Table 9
	BP	16.20	<0.0001	Table 10
	R	3.96	0.048	Table 12

Table 1b: Statistically significant ANOVA results (main effects and/or highest order interactions) for the fruit maturity and quality variables for the **2018** season with reference to the individual ANOVA tables (*p*=0.05). Region (**R**), Tree Position (**TP**), Bearing Position (**BP**), Light Exposure (**LE**).

Variable	Source of variability	F value	*p*-value	Table reference
Fruit aspect	R	166.05	<0.0001	Table 13
Mass	R x BP	13.09	0.0004	Table 14
	TP	9.30	0.003	Table 15
	LE	4.04	0.046	Table 16
Hue-angle at harvest	R	6.59	0.011	Table 13
	TP	96.70	<0.0001	Table 15
	LE	64.07	<0.0001	Table 16
Hue-angle after storage	TP	58.80	<0.0001	Table 15
	LE	35.64	<0.0001	Table 16
Lightness at harvest	R	48.31	<0.0001	Table 13
	TP	55.72	<0.0001	Table 15
	LE	56.30	<0.0001	Table 16
	BP	4.60	0.033	Table 18
Lightness after storage	TP	6.92	0.011	Table 15
	LE	10.24	0.002	Table 16
Sunburn	R x TP x LE x BP	4.58	0.034	Table 20
Starch conversion	R x TP	20.09	<0.0001	Table 17
	R x LE	19.36	0.0001	Table 19
Firmness at harvest	R x BP	4.99	0.030	Table 14
	TP	9.09	0.004	Table 15
Firmness after storage	R x TP	7.58	0.008	Table 17
	BP	15.15	0.0003	Table 18
	TP x LE	5.59	0.022	Table 21

Table 1c: ANOVA summary of **fruit aspect, mass, hue angle, lightness, sunburn, starch conversion percentage** and **firmness** variation across orchards and among trees as a source of variation for the 2017 and 2018 seasons.

Variable	Source	2017		2018	
		F-value	*p*-value	F-value	*p*-value
Fruit aspect	Orchard	38.20	**<0.0001**	15.83	**<0.0001**
	Tree	4.11	**<0.0001**	3.19	**<0.0001**
Mass	Orchard	49.16	**<0.0001**	50.72	**<0.0001**
	Tree	8.36	**<0.0001**	5.57	**<0.0001**
Hue angle at harvest	Orchard	38.25	**<0.0001**	25.65	**<0.0001**
	Tree	5.41	**<0.0001**	3.47	**<0.0001**
Hue angle after storage	Orchard	42.36	**<0.0001**	32.18	**<0.0001**
	Tree	5.87	**<0.0001**	6.11	**<0.0001**
Lightness at harvest	Orchard	100.61	**<0.0001**	30.80	**<0.0001**
	Tree	8.92	**<0.0001**	5.01	**<0.0001**
Lightness after storage	Orchard	72.53	**<0.0001**	19.23	**<0.0001**
	Tree	8.39	**<0.0001**	4.39	**0.001**
Sunburn	Orchard	7.40	**<0.0001**	5.32	**<0.0001**
	Tree	1.50	**0.001**	1.04	0.426
Starch conversion (%)	Orchard	66.24	**<0.0001**	21.71	**<0.0001**
	Tree	17.22	**<0.0001**	6.75	**<0.0001**
Firmness at harvest	Orchard	9.55	**<0.0001**	24.25	**<0.0001**
	Tree	4.17	**<0.0001**	4.38	**<0.0001**
Firmness after storage	Orchard	23.66	**<0.0001**	47.73	**<0.0001**
	Tree	7.33	**<0.0001**	3.33	**0.002**

Table 2: Mean values from **2017** for light exposure, starch conversion percentage, **fruit aspect**, **mass**, and **sunburn**, and the respective statistical relationships thereof, for the interaction between region, tree position and light exposure (**RxTPxLE**). Means with different letters are significantly different (p<0.05).

Region	Tree position	Light exposure	Starch conversion (%)	Fruit aspect	Mass (g)	Sunburn (score)
Koue Bokkeveld	Top	Sun	29.6 f	0.981 b	128.49 a	1.52 a
		Shade	33.5 e	0.990 a	128.18 a	0.25 d
	Bottom	Sun	54.5 a	0.983 b	128.23 a	0.84 c
		Shade	49.7 b	0.986 ab	117.34 b	0.08 e
Elgin	Top	Sun	35.0 de	0.927 c	99.29 d	1.45 a
		Shade	39.6 c	0.919 d	99.58 d	0.29 d
	Bottom	Sun	37.6 cd	0.923 cd	104.06 c	1.11 b
		Shade	46.2 b	0.921 d	101.42 cd	0.15 e
$LSD_{0.05\%}$			3.61	0.006	3.211	0.102
p-value			0.040	0.047	0.011	0.003

Table 3: Mean values in **2017** for fruit aspect and statistical relationships thereof, for the interaction between region, **tree position** and **bearing position** (RxTPxBP). Means with different letters are significantly different (p<0.05).

Region	Tree position	Bearing position	Fruit aspect
Koue Bokkeveld	Top	Shoot	0.991 a
		Spur	0.980 b
	Bottom	Shoot	0.986 ab
		Spur	0.984 b
Elgin	Top	Shoot	0.926 c
		Spur	0.921 c
	Bottom	Shoot	0.927 c
		Spur	0.917 d
$LSD_{0.05\%}$			0.006
p-value			0.029

Table 4: Mean values in **2017** for **fruit aspect** and **hue angle after storage**, and the respective statistical relationships thereof, for the interaction between light exposure and bearing position (**LExBP**). Means with different letters are significantly different (p<0.05).

Light exposure	Bearing position	Fruit aspect	Hue angle after storage (degrees)
Sun	Shoot	0.960 a	107.35 b
	Spur	0.950 b	106.82 c
Shade	Shoot	0.960 a	108.21 a
	Spur	0.956 a	108.17 a
$LSD_{0.05\%}$		0.004	0.251
p-value		0.003	0.014

Table 5: Mean values in **2017** for **mass** and **hue angle after storage** and the statistical relationships thereof, for the interaction between tree position and bearing position (**TPxBP**). Means with different letters are significantly different (p<0.05).

Tree position	Bearing position	Mass (g)	Hue angle after storage (degrees)
Top	Shoot	118.09 a	107.04 c
	Spur	114.24 bc	107.01 c
Bottom	Shoot	114.24 b	108.64 a
	Spur	111.69 c	108.09 b
$LSD_{0.05\%}$		2.259	0.251
p-value		0.050	0.007

Table 6: Mean values in **2017** for **hue angle at harvest, hue angle after storage, lightness at harvest, lightness after storage**, and **firmness at harvest** and the respective statistical relationships thereof, for the interaction between region and tree position (**RxTP**). Means with different letters are significantly different (p<0.05).

Region	Tree position	Hue angle at harvest (degrees)	Hue angle after storage (degrees)	Lightness at harvest (value)	Lightness after storage (value)	Firmness at harvest (kg)
Koue Bokkeveld	Top	112.35 c	106.70 d	72.20 a	74.46 a	7.80 b
	Bottom	113.33 a	108.63 a	71.05 c	73.42 d	7.77 b
Elgin	Top	112.16 d	107.41 c	71.79 b	73.75 c	7.99 a
	Bottom	112.73 b	108.06 b	71.71 b	74.06 b	7.76 b
$LSD_{0.05\%}$		0.105	0.252	0.143	0.237	0.089
p-value		<0.0001	<0.0001	<0.0001	<0.0001	0.002

Table 7: Mean values in **2017** for **hue angle at harvest, lightness at harvest** and **lightness after storage**, and the respective statistical relationships thereof, for the interaction between region and bearing position (**RxBP**). Means with different letters are significantly different (p<0.05).

Region	Bearing position	Hue angle at harvest (degrees)	Lightness at harvest (value)	Lightness at storage (value)
Koue Bokkeveld	Shoot	112.87 a	71.40 c	73.61 c
	Spur	112.76 b	71.90 a	74.32 a
Elgin	Shoot	112.38 c	71.65 b	73.74 c
	Spur	112.49 c	71.85 a	74.05 b
$LSD_{0.05\%}$		0.105	0.143	0.237
p-value		0.014	0.006	0.012

Table 8: Mean values in **2017** for **hue angle** and **lightness after harvest** and the respective statistical relationships thereof regarding light exposure (**LE**). Means with different letters are significantly different (p<0.05).

Light exposure	Hue angle at harvest (degrees)	Lightness after storage (value)
Sun	112.16 b	74.16 a
Shade	113.08 a	73.74 b
$LSD_{0.05\%}$	0.074	0.167
p-value	<0.0001	<0.0001

Table 9: Mean values in **2017** for **firmness after storage**, and the respective statistical relationships thereof, for the interaction between tree position and light exposure (**TPxLE**). Means with different letters are significantly different (p<0.05).

Tree position	Light exposure	Firmness after storage (kg)
Top	Sun	5.16 a
	Shade	4.83 c
Bottom	Sun	4.91 b
	Shade	4.69 d
$LSD_{0.05\%}$		0.074
p-value		0.039

Table 10: Mean values in **2017** for **starch conversion percentage, firmness at harvest, firmness after harvest,** and **sunburn**, and the respective statistical relationships thereof regarding bearing position (**BP**). Means with different letters are significantly different (p<0.05).

Bearing position	Sunburn (score)	Starch conversion (%)	Firmness at harvest (kg)	Firmness after storage (kg)
Shoot	0.74 a	39.57 a	7.89 a	4.95 a
Spur	0.67 b	41.25 a	7.77 b	4.85 b
$LSD_{0.05\%}$	0.051	1.795	0.062	0.053
p-value	0.049	0.001	<0.0001	<0.0001

Table 11: Mean values in **2017** for **lightness after storage** and the respective statistical relationship thereof, for the interaction between region and light exposure (**RxLE**). Means with different letters are significantly different (p<0.05).

Region	Light exposure	Lightness at harvest (value)
Koue Bokkeveld	Sun	71.98 b
	Shade	71.37 c
Elgin	Sun	72.19 a
	Shade	71.32 c
$LSD_{0.05\%}$		0.143
p-value		0.014

Table 12: Mean values in **2017** for **firmness after storage**, and the respective statistical relationship thereof regarding regions (**R**). Means with different letters are significantly different (p<0.05).

Region	Firmness after storage (kg)
Koue Bokkeveld	4.92 a
Elgin	4.87 b
$LSD_{0.05\%}$	0.053
p-value	0.048

Table 13: Mean values in **2018** for **fruit aspect, hue angle-angle at harvest,** and **lightness at harvest,** and the respective statistical relationships thereof regarding regions (**R**). Means with different letters are significantly different (p<0.05).

Region	Fruit aspect (ratio)	Hue-angle at harvest (degrees)	Lightness at harvest (value)
Koue Bokkeveld	0.992 a	113.32 a	71.62 b
Elgin	0.948 b	113.09 b	72.62 a
$LSD_{0.05\%}$	0.007	0.179	0.283
p-value	<0.0001	0.011	<0.0001

Table 14: Mean values in **2018** for **mass** and **firmness at harvest**, and the respective statistical relationships thereof, for the interaction between region and bearing position (**RxBP**). Means with different letters are significantly different (p<0.05).

Region	Bearing position	Mass (g)	Firmness at harvest (kg)
Koue Bokkeveld	Shoot	112.41 c	7.84 a
	Spur	120.77 b	7.45 b
Elgin	Shoot	130.14 a	7.59 b
	Spur	124.76 b	7.57 b
$LSD_{0.05\%}$		4.858	0.175
p-value		0.0004	0.030

Table 15: Mean values in **2018** for **mass, hue angle-angle at harvest, hue angle-angle after storage, lightness at harvest, lightness after storage,** and **firmness at harvest**, and the respective statistical relationships thereof regarding light exposure (**TP**). Means with different letters are significantly different (p<0.05).

Tree position	Mass (g)	Hue angle at harvest (degrees)	Hue angle after storage (degrees)	Lightness at harvest (value)	Lightness after storage (value)	Firmness at harvest (kg)
Top	124.01 a	112.83 b	105.42 b	72.57 a	75.77 a	7.70 a
Bottom	121.57 a	113.63 a	106.84 a	71.73 b	75.22 b	7.47 b
$LSD_{0.05\%}$	3.412	0.178	0.403	0.283	0.518	0.121
p-value	0.003	<0.0001	<0.0001	<0.0001	0.011	0.004

Table 16: Mean values in **2018** for **mass, hue angle-angle at harvest, hue angle-angle after storage, lightness at harvest** and **lightness after storage**, and the respective statistical relationships thereof regarding light exposure (**LE**). Means with different letters are significantly different (p<0.05).

Light exposure	Mass (g)	Hue angle at harvest (degrees)	Hue angle after storage (degrees)	Lightness at harvest (value)	Lightness after storage (value)
Sun	126.43 a	112.61 a	105.11 b	73.08 a	76.22 a
Shade	120.34 b	113.61 b	106.77 a	71.54 b	74.00 b
$LSD_{0.05\%}$	3.429	0.179	0.406	0.285	0.521
p-value	0.046	<0.0001	<0.0001	<0.0001	0.002

Table 17: Mean values in **2018** for **starch conversion** and **firmness after storage**, and the respective statistical relationships thereof, for the interaction between region and tree position (**RxTP**). Means with different letters are significantly different (p<0.05).

Region	Tree position	Starch conversion (%)	Firmness after storage (kg)
Koue Bokkeveld	Top	21.43 c	5.52 a
	Bottom	39.47 a	5.03 c
Elgin	Top	20.59 c	5.21 b
	Bottom	26.39 b	5.15 bc
$LSD_{0.05\%}$		3.682	0.157
p-value		<0.0001	0.008

Table 18: Mean values in **2018** for **lightness at harvest** and **firmness after storage**, and the respective statistical relationships thereof regarding bearing position (**BP**). Means with different letters are significantly different (p<0.05).

Bearing position	Lightness at harvest (degrees)	Firmness after storage (kg)
Shoot	71.98 b	5.34 a
Spur	72.40 a	5.16 b
$LSD_{0.05\%}$	0.282	0.110
p-value	0.033	0.0003

Table 19: Mean values in **2018** for **starch conversion** and the respective statistical relationship thereof, for the interaction between region and light exposure (**RxLE**). Means with different letters are significantly different (p<0.05).

Region	Light exposure	Starch conversion (%)
Koue Bokkeveld	Sun	18.12 c
	Shade	33.46 a
Elgin	Sun	25.10 b
	Shade	21.87 b
$LSD_{0.05\%}$		3.708
p-value		<0.0001

Table 20: Mean values in **2018** for **sunburn** and the respective statistical relationships thereof, for the interaction between region, tree position, light exposure and bearing position (**RxTPxLE**). Means with different letters are significantly different (p<0.05).

Region	Tree position	Light exposure	Bearing position	Sunburn (score)
Koue Bokkeveld	Top	Sun	Shoot	1.12 bc
			Spur	0.97 c
		Shade	Shoot	0.06 ef
			Spur	0.06 ef
	Bottom	Sun	Shoot	1.67 a
			Spur	0.38 de
		Shade	Shoot	0.00 f
			Spur	0.06 ef
Elgin	Top	Sun	Shoot	1.76 a
			Spur	1.43 ab
		Shade	Shoot	0.49 d
			Spur	0.15 def
	Bottom	Sun	Shoot	1.14 bc
			Spur	1.26 bc
		Shade	Shoot	0.05 ef
			Spur	0.06 ef
$LSD_{0.05\%}$				0.371
p-value				0.034

Table 21: Mean values in **2018** for **firmness after storage** and the respective statistical relationship of the interaction between tree position and light exposure (**TPxLE**). Means with different letters are significantly different (p<0.05).

Tree position	Light exposure	Firmness after storage (kg)
Top	Sun	5.48 a
	Shade	5.24 b
Bottom	Sun	5.38 ab
	Shade	4.97 c
$LSD_{0.05\%}$		0.162
p-value		0.022

Table 22: Pearson correlation matrix for all variables in 2017. Values in bold are different from 0 with a significance level of p =0.05

Variable	Fruit aspect	Mass harvest	Hue-angle harvest	Hue-angle storage	Lightness harvest	Lightness storage	Sunburn	Starch conversion	Firmness harvest	Firmness storage
Fruit aspect	1.00	**0.14**	**0.13**	0.03	**-0.09**	-0.03	**-0.09**	**-0.08**	0.03	**-0.05**
Mass	**0.14**	1.00	**-0.18**	-0.06	0.04	-0.04	**0.17**	-0.02	**-0.47**	**-0.34**
Hue angle harvest	**0.13**	**-0.18**	1.00	**0.71**	**-0.71**	**-0.49**	**-0.67**	**0.37**	-0.12	-0.13
Hue angle storage	0.03	**-0.06**	**0.71**	1.00	**-0.81**	**-0.84**	**-0.39**	**0.41**	**-0.21**	**-0.23**
Lightness harvest	**-0.09**	0.04	**-0.71**	**-0.81**	1.00	**0.84**	**0.32**	**-0.40**	**0.24**	**0.22**
Lightness storage	-0.03	-0.04	**-0.49**	**-0.84**	**0.84**	1.00	0.10	**-0.37**	**0.20**	0.06
Sunburn	**-0.09**	**0.17**	**-0.67**	**-0.39**	**0.32**	0.10	1.00	**-0.19**	0.04	**0.28**
Starch conversion	**-0.08**	-0.02	**0.37**	**0.41**	**-0.40**	**-0.37**	**-0.19**	**1.00**	**-0.29**	-0.06
Firmness harvest	0.03	**-0.47**	-0.12	**-0.21**	**0.24**	**0.20**	0.04	**-0.29**	1.00	**0.40**
Firmness storage	**-0.05**	**-0.34**	-0.13	**-0.23**	**0.22**	0.06	**0.28**	-0.06	**0.40**	1.00

Table 23: Pearson correlation matrix for all variables in 2018. Values in bold are different from 0 with a significance level of p =0.05

Variable	Fruit aspect	Mass harvest	Hue-angle harvest	Hue-angle storage	Lightness harvest	Lightness storage	Sunburn	Starch conversion	Firmness harvest	Firmness storage
Fruit aspect	1.00	**0.33**	**0.26**	0.07	**-0.37**	-0.12	**-0.22**	**0.21**	**-0.29**	**-0.37**
Mass	**0.33**	1.00	-0.04	-0.09	-0.03	-0.16	0.07	**0.19**	**-0.62**	**-0.62**
Hue angle harvest	**0.26**	-0.04	1.00	**0.77**	**-0.77**	**-0.58**	**-0.49**	**0.33**	**-0.26**	**-0.35**
Hue angle storage	0.07	-0.09	**0.77**	1.00	**-0.74**	**-0.85**	**-0.22**	**0.30**	**-0.26**	**-0.25**
Lightness harvest	**-0.37**	-0.03	**-0.77**	**-0.74**	1.00	**0.74**	**0.36**	**-0.45**	**0.36**	**0.46**
Lightness storage	-0.12	-0.16	**-0.58**	**-0.85**	**0.74**	1.00	0.12	**-0.32**	**0.36**	**0.36**
Sunburn	**-0.22**	0.07	**-0.49**	**-0.22**	**0.36**	0.12	1.00	-0.10	0.09	0.15
Starch conversion	**0.21**	**0.19**	**0.33**	**0.30**	**-0.45**	**-0.32**	-0.10	1.00	**-0.48**	**-0.42**
Firmness harvest	**-0.29**	**-0.62**	**-0.26**	**-0.26**	**0.36**	**0.36**	0.09	**-0.48**	1.00	**0.59**
Firmness storage	**-0.37**	**-0.62**	**-0.35**	**-0.25**	**0.46**	**0.36**	0.15	**-0.42**	**0.59**	1.00

Figures

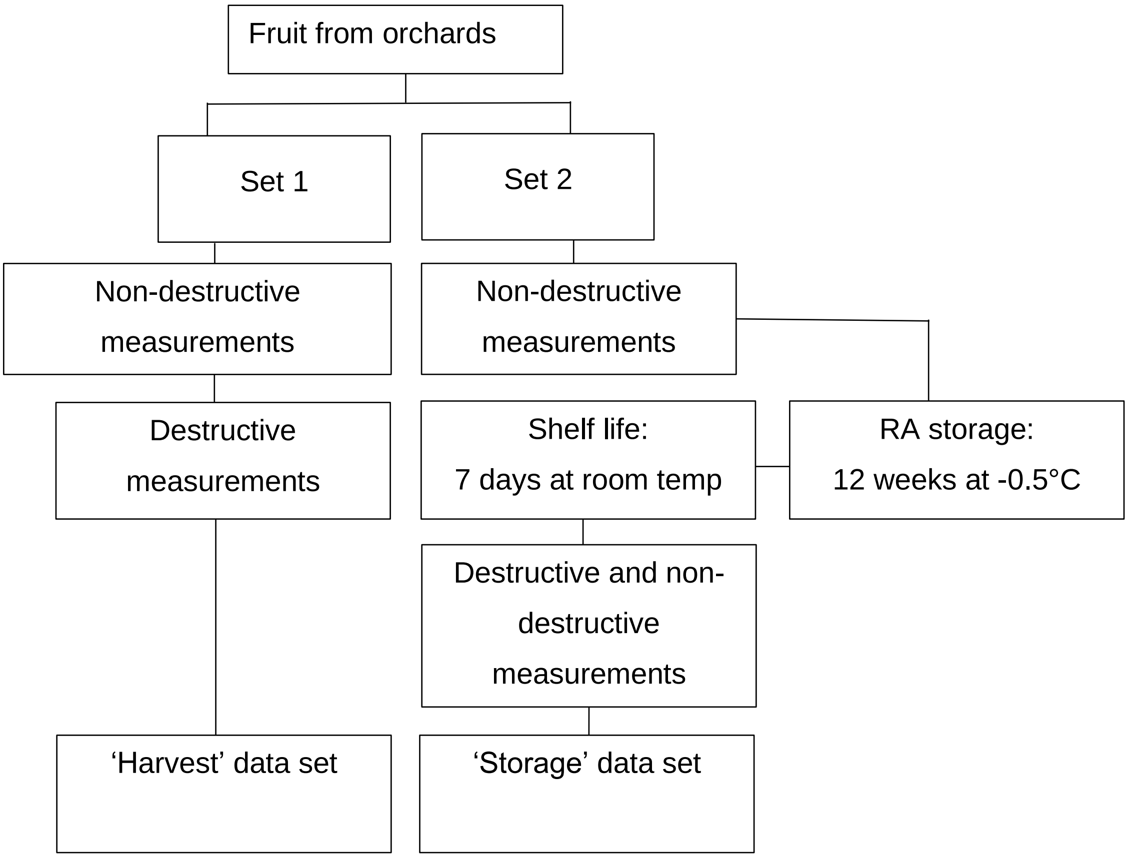

Figure 1: Flow diagram to illustrate the sequence of events to create a "Harvest" and "Storage" dataset.

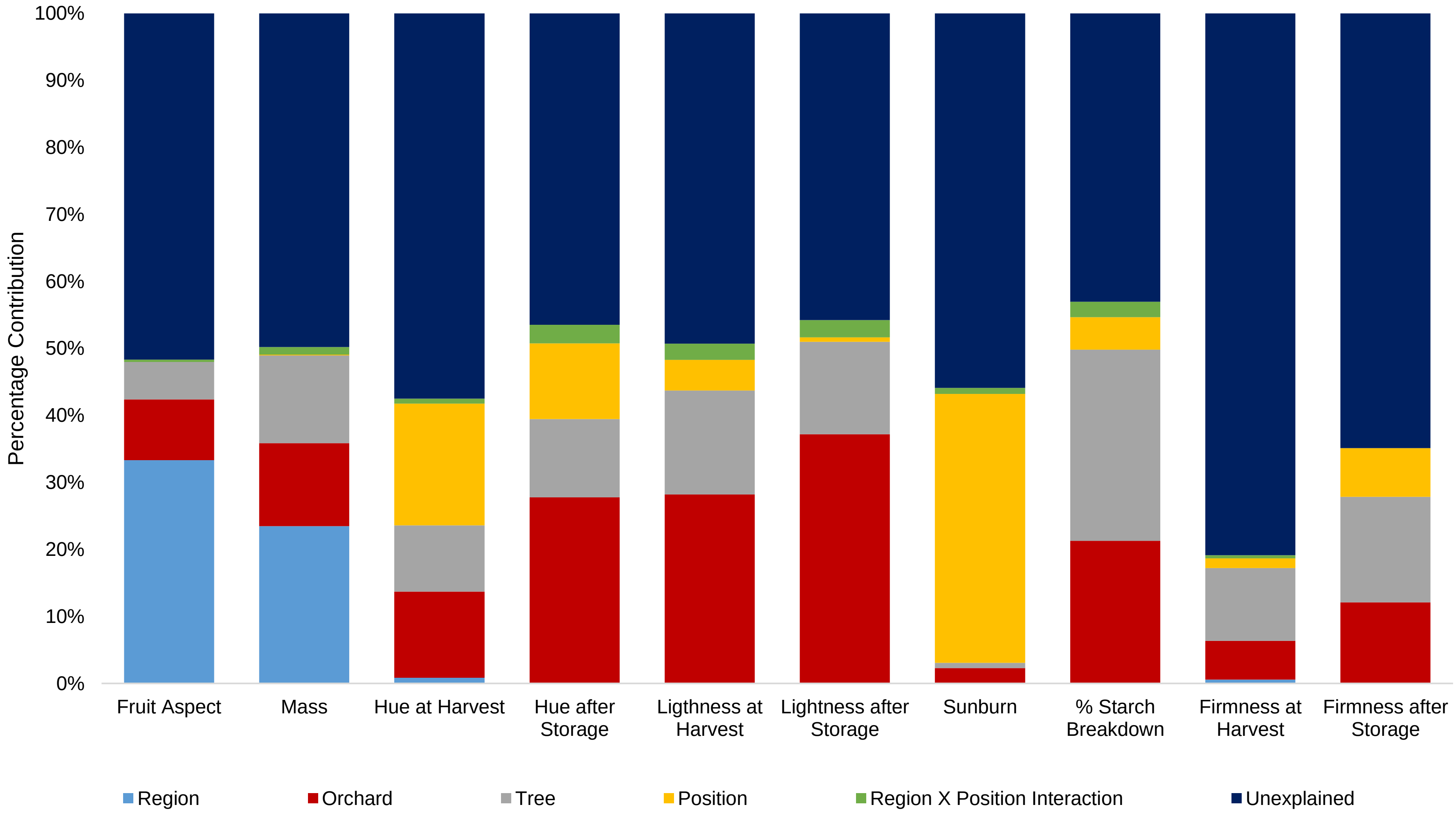

Figure 2: Variance component analysis for **2017** indicating the percentage contribution of each component towards the measured variable.

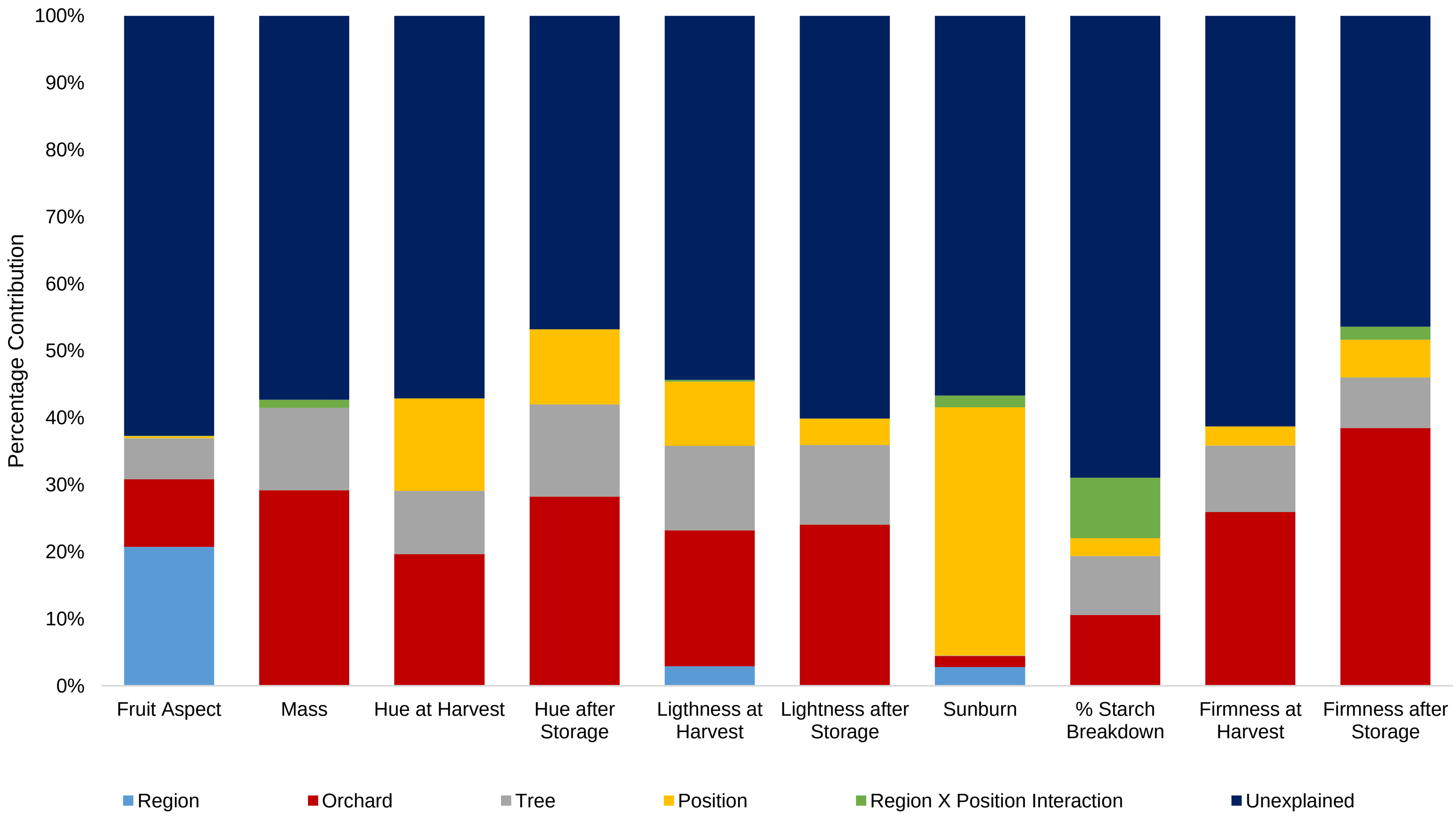

Figure 3: Variance component analysis for **2018** indicating the percentage contribution of each component towards the measured variable.

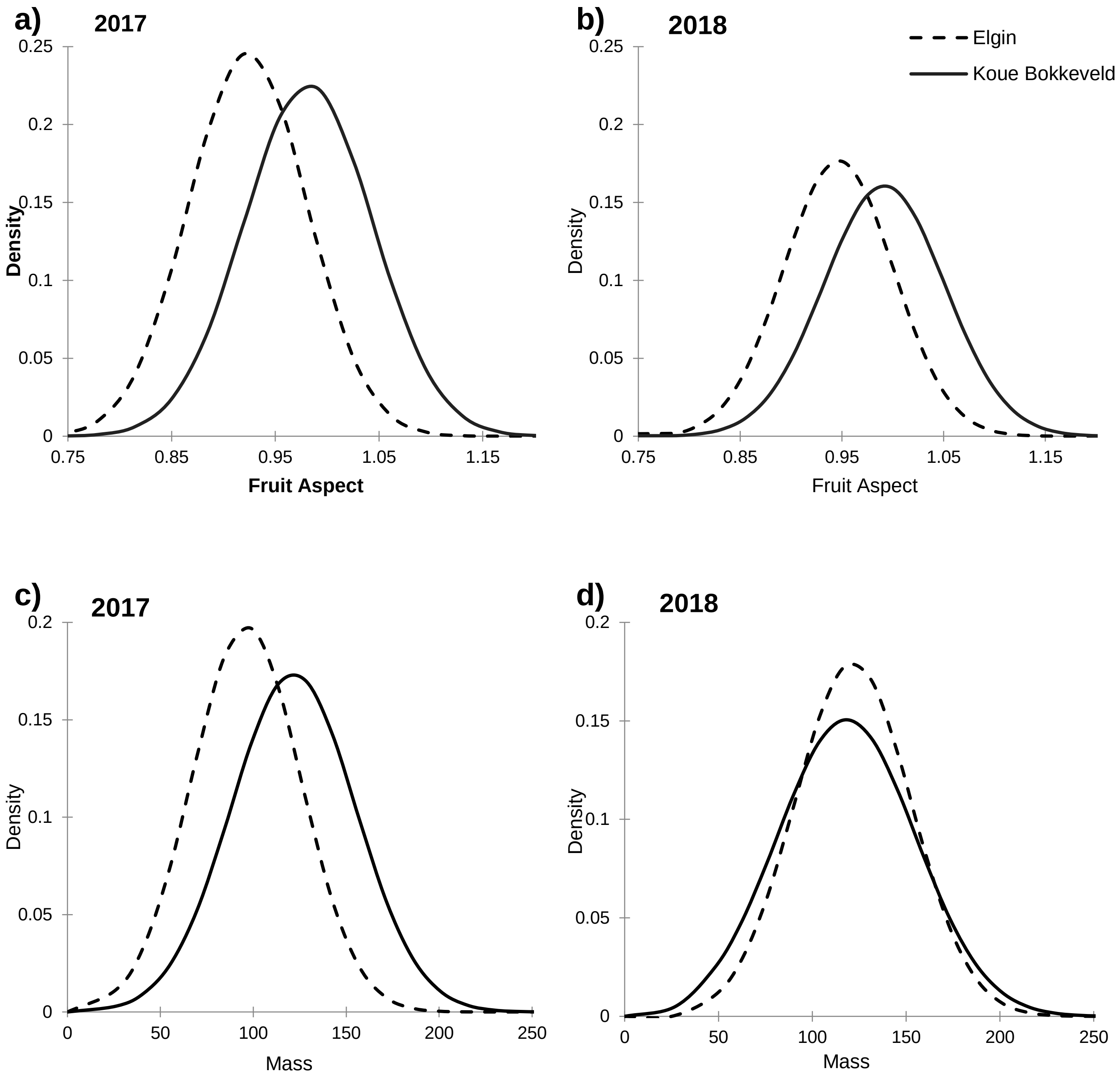

Figure 4: Density curves showing the distribution of fruit aspect in Elgin and the Koue Bokkeveld for 2017 (a) and 2018 (b). Density curves showing the distribution of fruit mass in Elgin and the Koue Bokkeveld for 2017 (c) and 2018 (d).

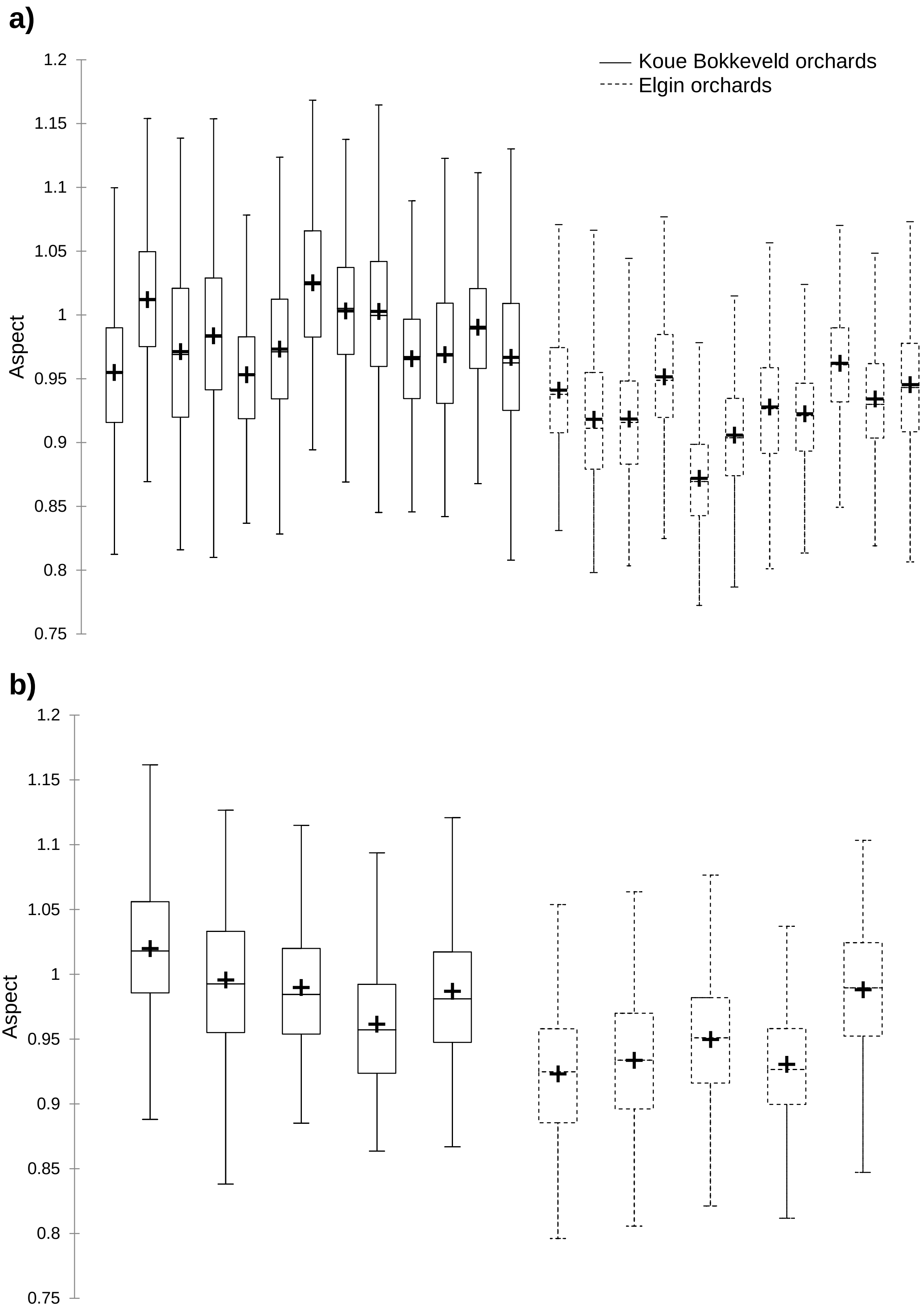

Figure 5: Box plots of fruit aspect per orchard for the a) 2017 and b) 2018 seasons. The box plot splits the data in four quartiles with an equal number of data points in each. The centre horizontal line in the median and the **+** is the mean.

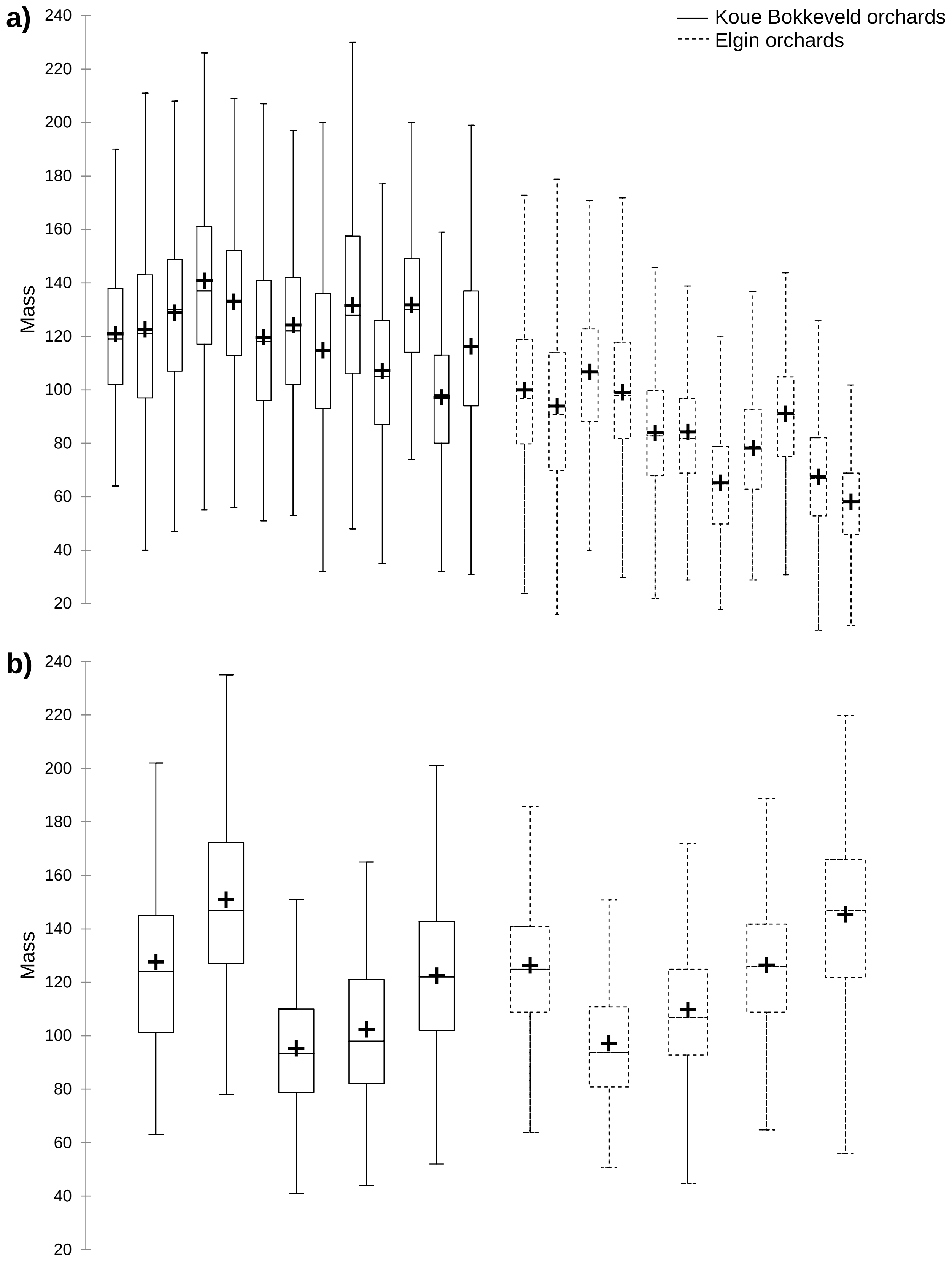

Figure 6: Box plots of fruit mass per orchard for the a) 2017 and b) 2018 seasons. The box plot splits the data in four quartiles with an equal number of data points in each. The centre horizontal line in the median and the **+** is the mean.

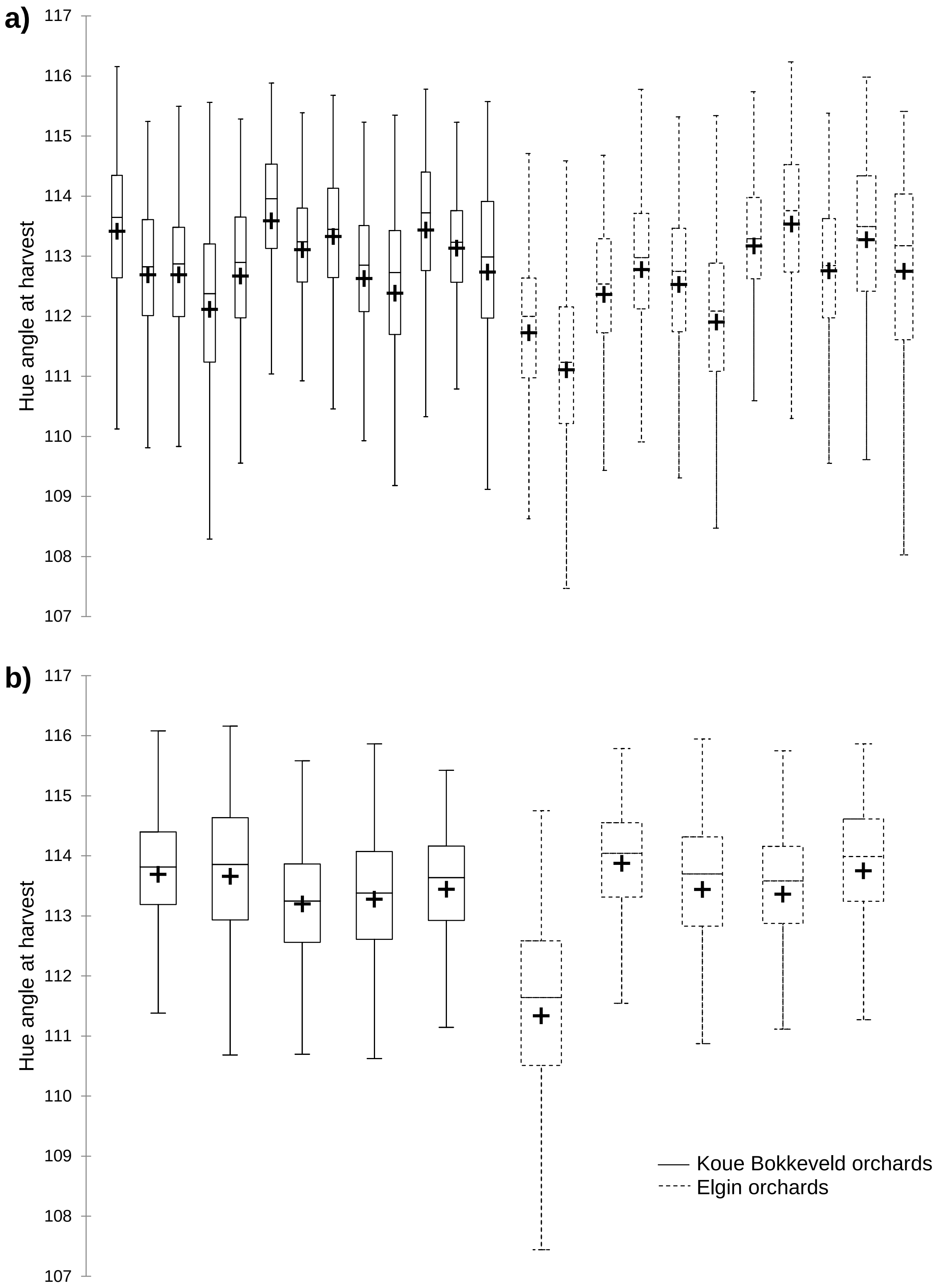

Figure 7: Box plots of hue angle at harvest per orchard for the a) 2017 and b) 2018 seasons. The box plot splits the data in four quartiles with an equal number of data points in each. The centre horizontal line in the median and the **+** is the mean.

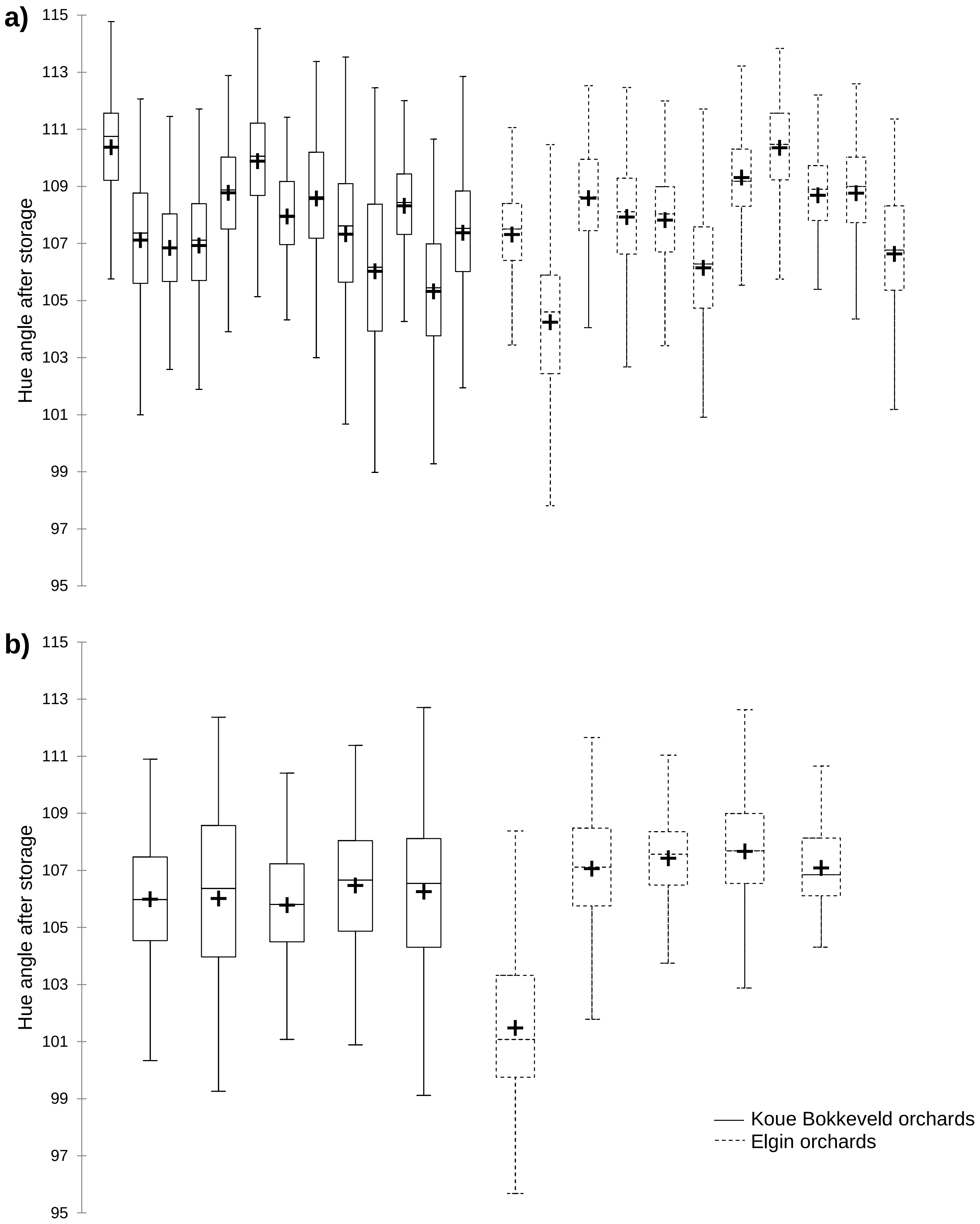

Figure 8: Box plots of hue angle after storage per orchard for the a) 2017 and b) 2018 seasons. The box plot splits the data in four quartiles with an equal number of data points in each. The centre horizontal line in the median and the **+** is the mean.

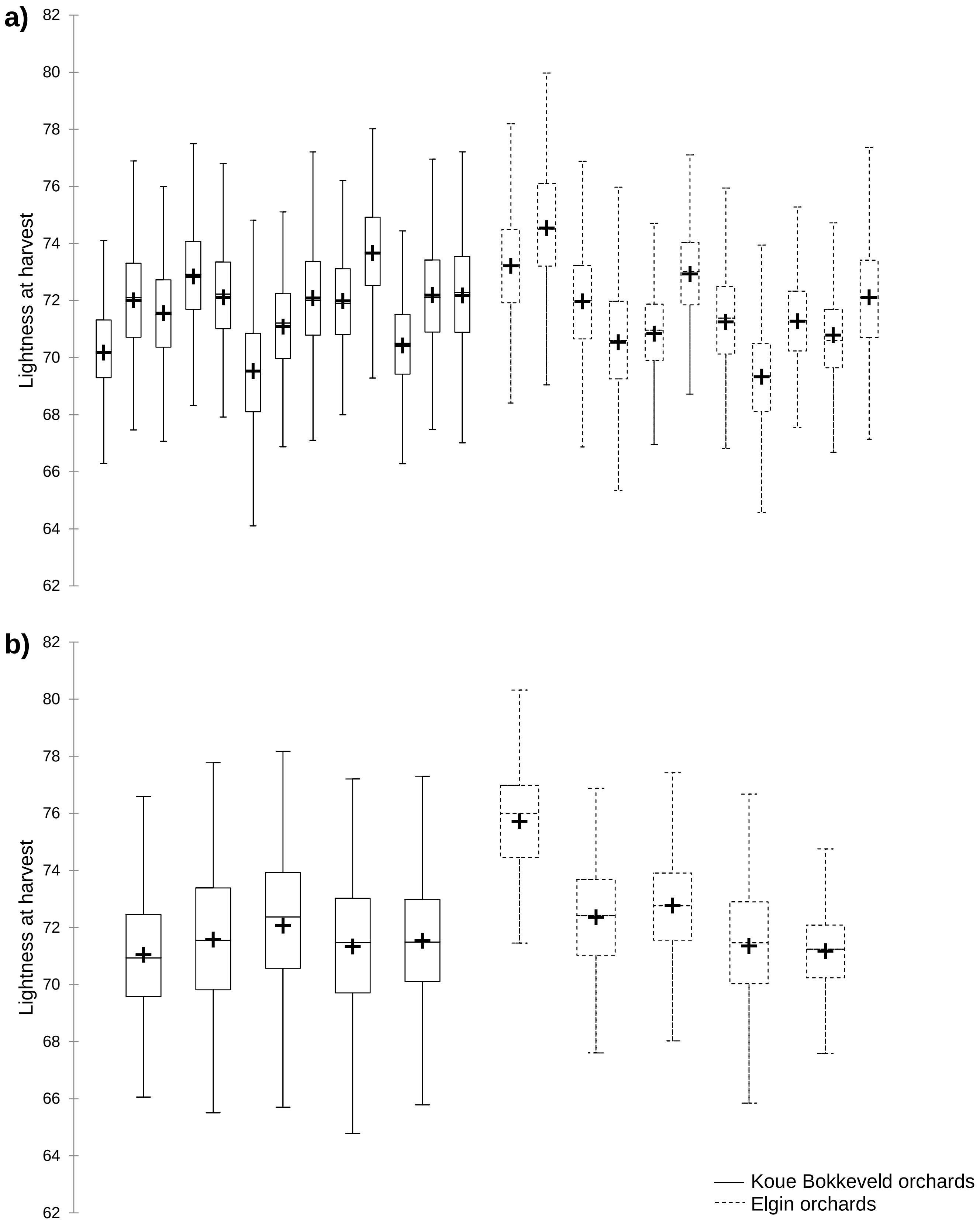

Figure 9: Box plots of lightness at harvest per orchard for the a) 2017 and b) 2018 seasons. The box plot splits the data in four quartiles with an equal number of data points in each. The centre horizontal line in the median and the **+** is the mean.

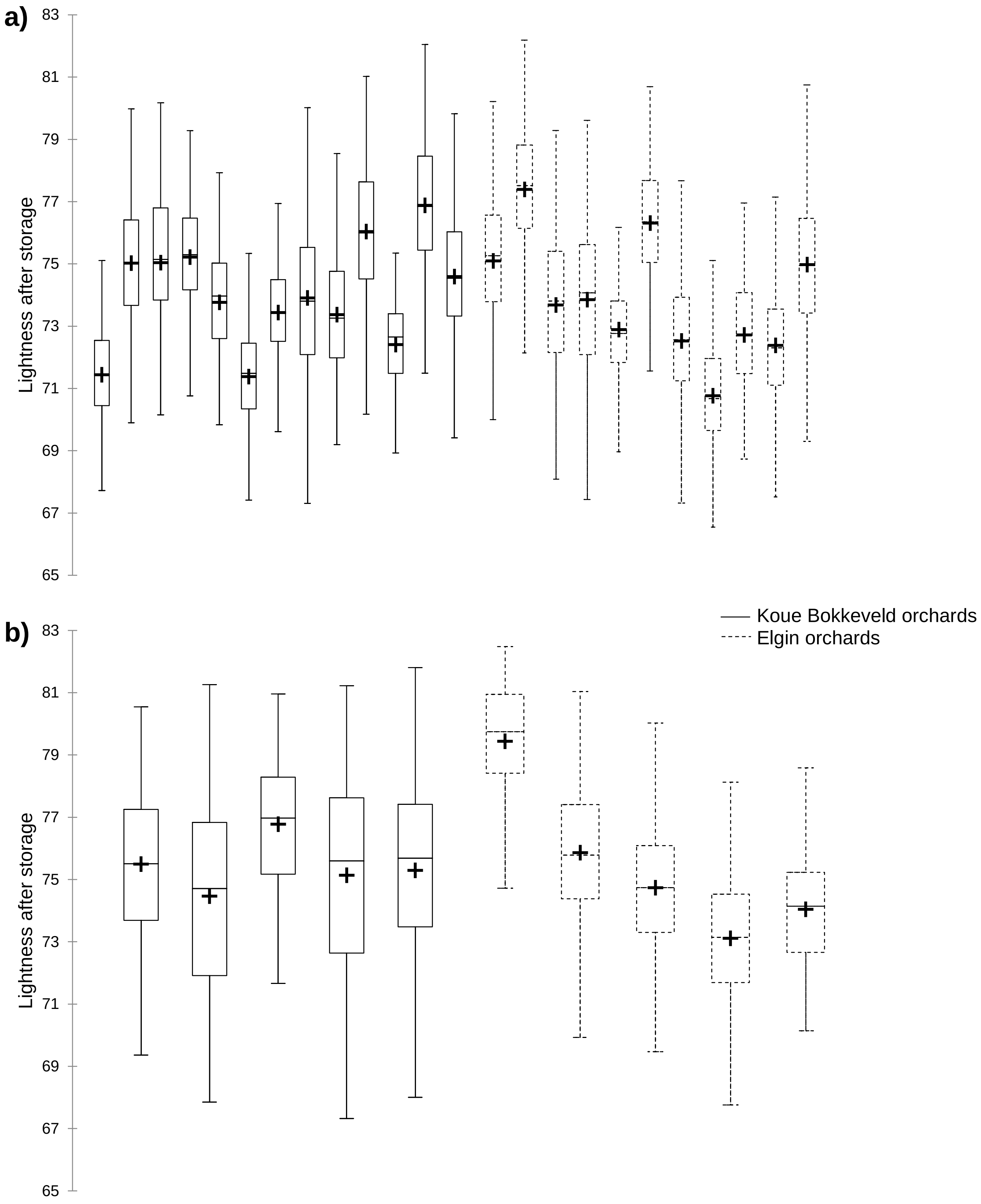

Figure 10: Box plots of lightness after storage per orchard for the a) 2017 and b) 2018 seasons. The box plot splits the data in four quartiles with an equal number of data points in each. The centre horizontal line in the median and the **+** is the mean.

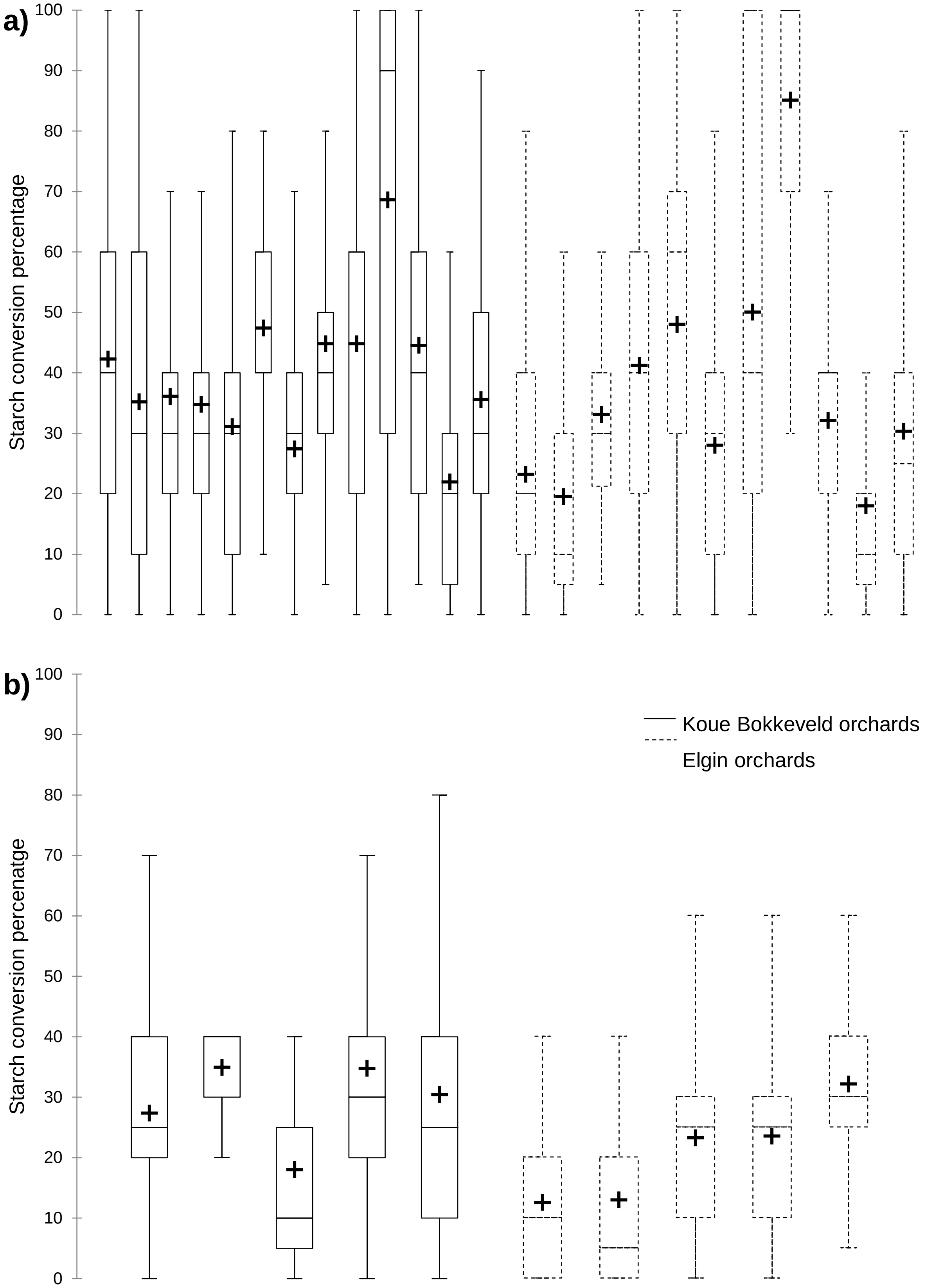

Figure 11: Box plots of starch conversion percentage per orchard for the a) 2017 and b) 2018 seasons. The box plot splits the data in four quartiles with an equal number of data points in each. The centre horizontal line in the median and the **+** is the mean.

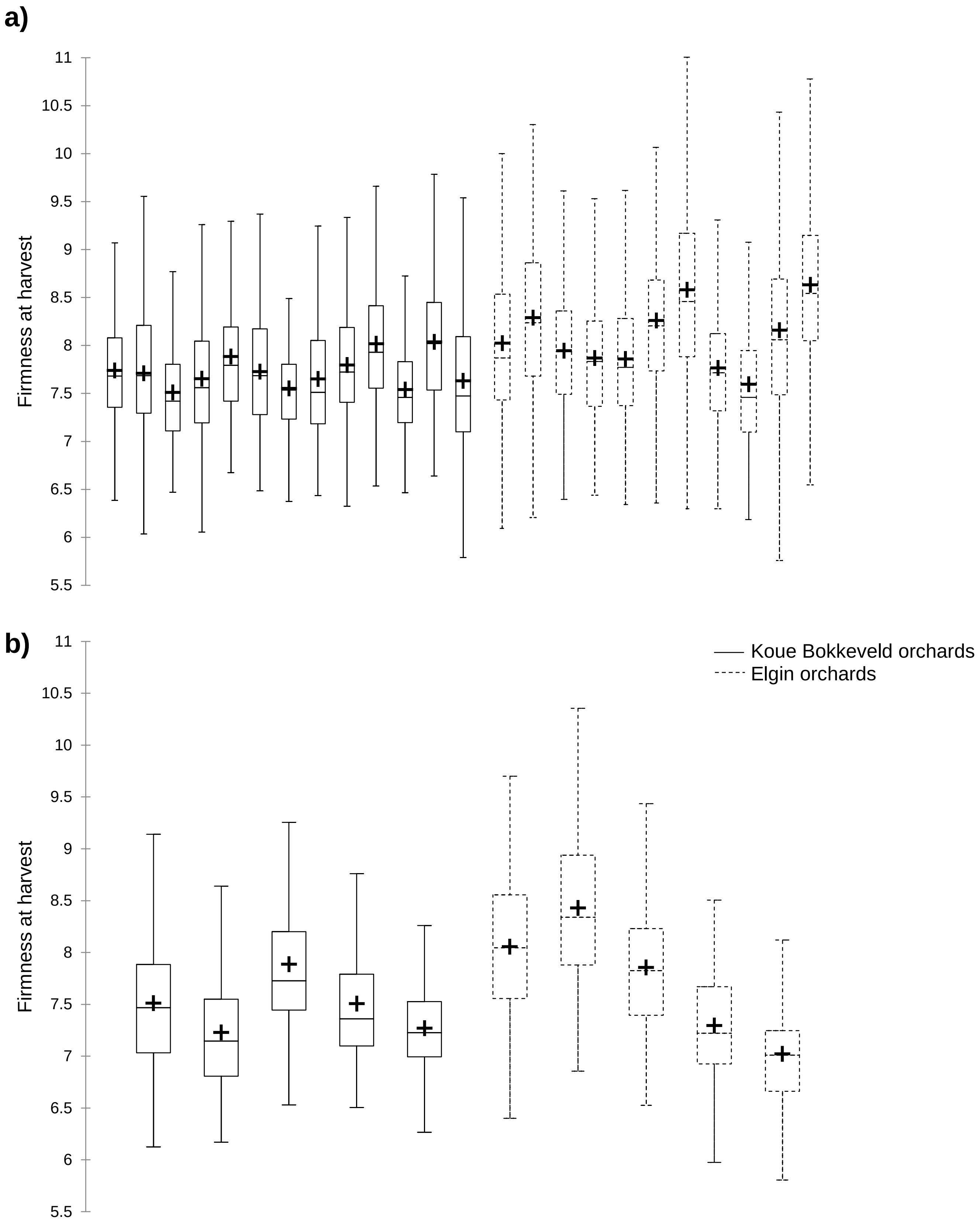

Figure 12: Box plots of firmness at harvest per orchard for the a) 2017 and b) 2018 seasons. The box plot splits the data in four quartiles with an equal number of data points in each. The centre horizontal line in the median and the **+** is the mean.

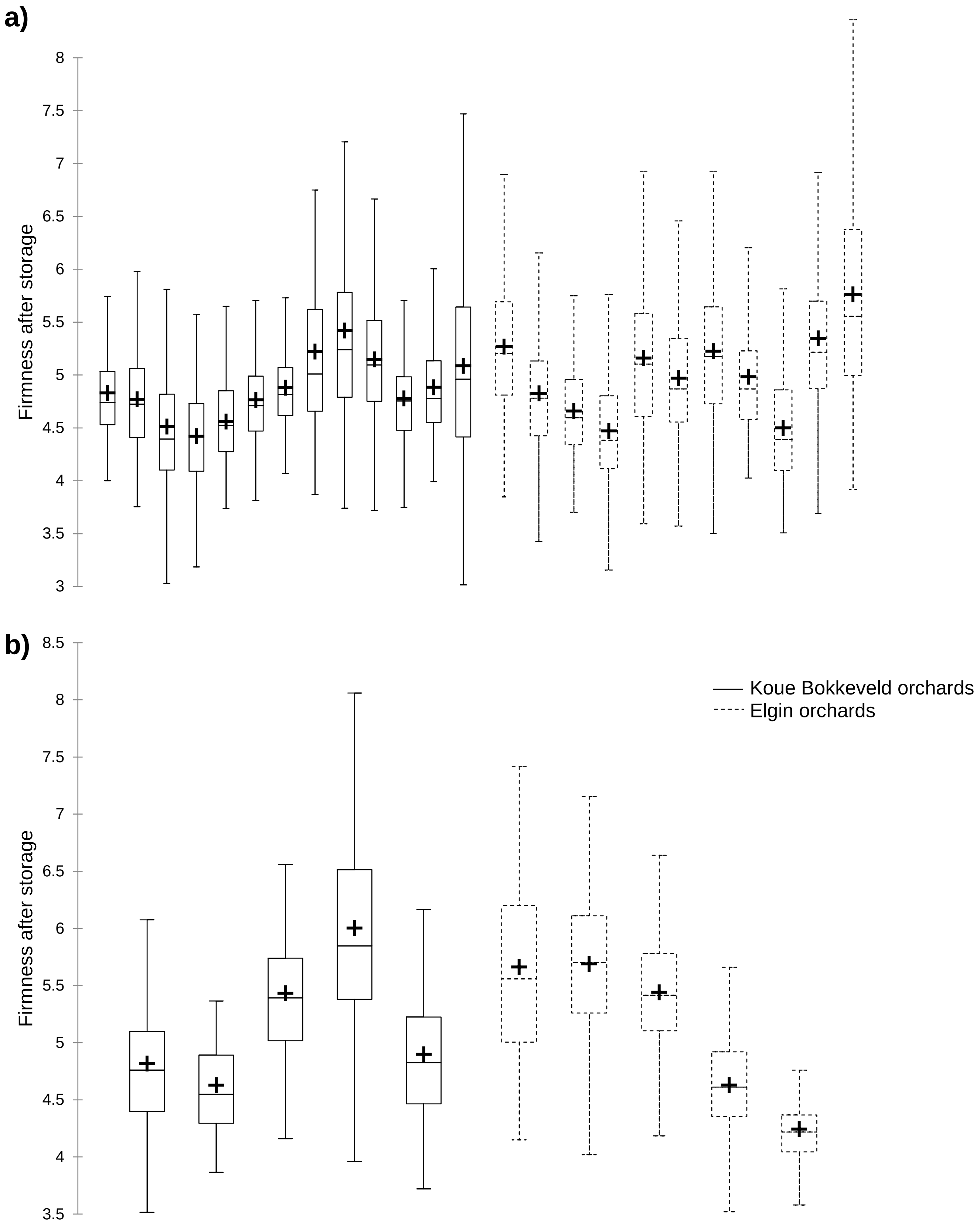

Figure 13: Box plots of firmness after storage per orchard for the a) 2017 and b) 2018 seasons. The box plot splits the data in four quartiles with an equal number of data points in each. The centre horizontal line in the median and the **+** is the mean.

PAPER 2: The influence of relative bloom date on 'Golden Delicious' fruit maturity, quality and variability.

Abstract

Insufficient winter chill results in protracted bloom. In deciduous fruit trees this can lead to management issues during the season and results in mixed maturities in fruit at harvest. Climate change and the resultant warming may increase the difficulties already experienced. The extent to which flowering time or relative bloom date (RBD: classed as early, full bloom, and late) influenced maturity and quality of 'Golden Delicious' fruit was quantified. Flower clusters were tagged with dates during bloom in two climatically distinct production regions of South Africa. Fruit aspect (height:diameter), weight, colour, flesh firmness, and starch conversion percentage (SC%) were measured at harvest with a portion of the fruit being measured after 12 weeks of regular atmosphere storage at -0.5°C. Protracted bloom of 15 to 16 days was observed in both production regions. The parameters measured showed large variation, but much of the variance remained unexplained. RBD explained less of the variation compared to variation between orchards and trees. RBD had a notable effect on SC% and fruit mass. Fruit from the warmer winter region showed the early clusters producing the fruit with higher SC% than that of late clusters but in the cooler winter climate there was no difference. This work serves to describe and quantify the influence of protracted bloom on fruit production regions that lack sufficient winter cold.

Keywords: blossom, variation, position, region, apple

1. Introduction

Climate change is putting pressure on agriculture. Annual crops are relatively easy to move to more suitable regions. Perennial crop production, like that of apples, is particularly vulnerable to climate change because it cannot be relocated and requires years of production to generate a suitable return on investment (Glen *et al*., 2013). Apple, in particular, is adversely affected by warmer temperature. Warming temperatures were seen to advance the bloom dates of apple trees throughout continental Europe, reducing the variability between regions (Legave *et al*., 2013). Although this paper suggested that warmer springs lead to earlier bloom in European apple crops, it also highlighted that bloom date was less advanced in Mediterranean and coastal regions of Europe due to progressively milder winters resulting in insufficient chill accumulation.

Grab and Craparo (2011) showed the same trend in South Africa, with ‘Golden Delicious’ particularly susceptible to the warming winter temperatures. Predicted warming is believed to effect Mediterranean-type climates to a greater extent, and in the case of temperate fruit crops a warmer/milder winter increases variation in fruit quality and other growth-related aspects (Baldocchi and Wong, 2006; Midgley and Lötze, 2008). Most of the apple growing regions in the Western and Eastern Cape provinces of South Africa accumulate insufficient winter chill leading to a protracted endodormancy induction phase, a low maximum bud dormancy level followed by an extended-release period (Cook *et al*., 2017). This results in various adverse symptoms in spring ranging from delayed, prolonged bud break, formation of “bare necks” (unbranched shoots), decrease in vigour and extended flowering periods (Cook and Jacobs, 2000). The extended bloom arising from warmer winters, is expected to have effects such as delayed maturation and mixed maturities at harvest (Cook and Jacobs, 2000). Mixed maturity in a tree is difficult to manage during the growing season and complicates harvesting, especially for a cultivar such as ‘Golden Delicious’ that is traditionally harvested as a single strip pick. Not all fruit will be harvested at optimal maturity and fruit are often harvested too early to avoid overripe fruit within a batch. Chemical rest breaking agents can be applied to partially alleviate the symptoms of extended bloom and is part of the standard apple production regime in mild winter areas (Petri, 1985). The success of chemical rest breaking is often variable and unreliable as it is influenced by factors such as temperatures after application and water status of the soil (Erez, 1994). Protracted bloom continues to be a challenge in mild winter production regions and with the prediction of increasing winter temperatures in South Africa, marginal regions are also increasingly at risk (Midgley and Lötze, 2008).

After a great deal of unexplained variation was reported in Paper 1 in all parameters measured, date of blossom was considered as a possible additional source of variation. To date protracted bloom has been studied well (Cook *et al*., 2017; Erez 2000; Funes *et al*., 2016; Gadoury, 2015; Hauagge and Cummins, 1991), but besides the resultant mixed maturity, it has not been linked directly to final fruit quality. This paper aims to determine if blossom date has an influence on fruit maturity and quality parameters at harvest and after cold storage and to quantify the effect of blossom date on the variance in said parameters.

2. Materials and methods

2.1. Plant material and study site

The trial was performed during the 2017/2018 season using 11 ‘Golden Delicious’ orchards in the climatically contrasting regions of the Koue Bokkeveld (annual avg. of ±1400 Utah chill units) and Elgin (annual avg. ±700 Utah chill units), similar to the sites described in Paper 1 of this thesis. Five orchards were selected in the Koue Bokkeveld and six orchards in the Elgin region with the orchard selection protocol and horticultural practises identical to that mentioned in Paper 1. The bloom dates of the two regions can differ up to fourteen days in some years, with the colder Koue Bokkeveld region, typically being earlier.

2.2. Flower monitoring and pre-harvest measurements

Prior to bud break, four trees were randomly selected in each orchard and two scaffold branches (perpendicular to row direction and on eastern side only) were identified of which one was in the upper and one in the lower part of the tree. The orchards in both regions received a spray application of mineral oil and hydrogen cyanamide (tank mix of 3% mineral oil with 0.26% cyanamide) as chemical rest breaking agent during bud swell. To record the chronology of bud break, the selected branches were monitored, and flower clusters were tagged upon opening. At the first sign of flowering, the orchards in the Koue Bokkeveld were visited Monday, Wednesday and Friday, while Elgin sites were visited Tuesdays, Thursdays and Saturdays. Flower clusters were tagged as they opened. A cluster was considered “open” once the third flower within the cluster had opened. For clusters that had opened on the day between visits, during the next visit these clusters were tagged as having opened the previous day. For the additional day gap over the weekend, clusters that were about to open, but did not yet qualify as open, were tagged as opening the following day (e.g., a site visit in Elgin on a Saturday, if a cluster only had the king flower open it would be tagged as having opened on the Sunday). All flowers that opened later than the nineth day after full bloom were identified by not having a tag at harvest and these fruits were grouped as bloom on the last day of tagging. Following fruit set, the clusters were thinned according to commercial practice, starting with chemical thinning and followed up with hand thinning to a maximum of three fruitlets per cluster. All the tags remained on the trees until harvest.

2.3. Post-harvest measurements

All the fruit from the selected branches were harvested on the commercial harvest date in similar categories as described in Paper 1. The post-harvest handling of the fruit and the fruit quality measurements were also similar to that described in Paper 1, with the following measurements taken: fruit size (diameter and height), weight, peel colour, flesh firmness, and starch conversion percentage. Similar to Paper 1, the standard XYZ colour space coordinates measurements from the colourimeter were converted to the HCL colour space, of parameters: hue, chroma and lightness. Only the lightness and hue angle metrics were used. Further detail on ‘Golden Delicious’ fruit colour is given in Paper 1. Two trees were used for each of the data sets (”Harvest” and “Storage”). The data collection was structured in such a way that each parameter was known for each individual fruit.

2.4. Calculations and Statistical analysis

The overall variability within each of the quality parameters was determined using a box and whiskers plot to depict the mean, median and distribution or the parameter (Fig. 1). Fruit shape was expressed as the fruit aspect ratio which is fruit height divided by diameter.

The fruit were traced back to their flowering time and depicted relative to the full bloom date of the orchard. The full bloom date of all the orchards were synchronised and the relative bloom date (RBD) for each fruit was determined relative to the full bloom date, which was given as a “day zero” value. Flowers that bloomed before the full bloom date were given a negative number according to the number of days before full bloom and those that flowered after were assigned a positive number of days relative to the full bloom date. See Fig. 2 for detail. All fruit that resulted from flowers that opened before the full bloom date, were classed as “Early”, those at full bloom as “Full Bloom” and fruit from flowers that opened after the full bloom date as “Late” (See Fig. 3).

To determine the contribution of each variance component (orchard, tree, position within the tree) on RBD, a variance component analysis was performed separately for each of the regions. The RBD data were treated as a nested model with random effects using the Proc VarComp procedure in SAS (Version 5.1, SAS Institute Inc, USA) with individual fruit as the experimental unit. This was followed by a variance component analysis on all fruit parameters to determine the contribution of each variance component, this time including RBD as a variance component. For the variables where the RBD contributed >1.5% of the total variability, the means were compared using the GLM procedure in SAS, followed by Fisher’s least significant difference test when the significance level was <0.05. Using the same “>1.5%” threshold, density curves were drawn for each bloom class using XLSTAT (version 2015.04.36025) to illustrate the distribution across each class. Correlations between the variables was done by means of the Pearson’s correlation test in XLSTAT (version 2015.04.36025) at a significance level of $p = 0.05$. The correlation coefficients were interpreted as very weak (0.0-0.19), weak (0.20-0.39), moderate (0.40-0.59), strong (0.60-0.79), very strong (0.80-1.00) according to Evans (1996) and Hinkle *et al*. (2003).

3. Results

Only the effect of RBD is pertinent to describing the variance in this paper, where other components of variation were described in Paper 1 (e.g. region, tree, position etc.).

3.1 Overall variability

Fig. 1 shows the variability of the various parameters as box and whisker diagrams of each parameter per region.

Fruit aspect ranged from 0.80 to 1.09 in Elgin and from 0.84 to 1.14 in the Koue Bokkeveld. The interquartile range in the Koue Bokkeveld (0.08) was larger than in Elgin (0.07; Fig. 1a). Considering the results for fruit mass, the box plot was positively skewed in the Koue Bokkeveld but in Elgin it was normally distributed. The range (167 g) and interquartile range (46 g) in the Koue Bokkeveld was larger than in Elgin (160 g and 43 g; Fig. 1b).

Regarding colour, the peel lightness values at harvest were normally distributed in both regions (Fig. 1c) but after storage lightness value was negatively skewed in the Koue Bokkeveld and positively skewed in Elgin (Fig. 1 d). At harvest, the values ranged from 65.3 to 77.8 in the Koue Bokkeveld and 66.7 to 78.1 in Elgin (Fig. 1c). The range in values were larger after storage with values from 67.9 to 81.8 in the Koue Bokkeveld and 68.7 to 82.1 in Elgin (Fig. 1d). The interquartile range increased from harvest to after storage from 3.29 to 4.03 in the Koue Bokkeveld and 2.88 to 3.61 in Elgin (Fig. 1c and 1d).

Peel hue angles in the Koue Bokkeveld ranged from 110.8° to 116.3° at harvest and 99.4° to 112.9° after storage (Fig. 1e and 1f). In Elgin the hue angles at harvest ranged from 110.2° to 115.9° and from 101.8° to 111.6° after storage (Fig. 1e and 1f). The hue angles, both at harvest and after storage, were normally distributed in the Koue Bokkeveld but both were skewed negatively in Elgin before harvest and after storage (Fig. 1e and 1f). The interquartile range in hue angle increased between harvest and after storage from 1.39° to 3.39° in the Koue Bokkeveld and 1.02° to 1.68° in Elgin (Fig. 1e and 1f).

Fruit firmness ranged from 5.95 to 8.98 kg at harvest and 3.52 to 7.03 kg after storage in the Koue Bokkeveld (Fig.1g and 1h). In Elgin the range in fruit firmness was from 5.72 to 9.43 kg at harvest and 3.52 to 7.18 kg after storage. Data was skewed to higher firmness at harvest for both regions (Fig. 1g), and after storage firmness data was skewed to higher firmness in the Koue Bokkeveld but normally distributed in Elgin (Fig. 1h). Firmness at harvest had an interquartile range of 0.79 kg in the Koue Bokkeveld and 0.97 kg in Elgin (Fig. 1g), and after storage the ranges were 1.00 kg and 1.06 kg, respectively (Fig. 1h).

The data for starch conversion percentage (SC%) ranged from 0% to 80% in the Koue Bokkeveld and from 0% to 60% in Elgin (Fig. 1i). The interquartile range in the Koue Bokkeveld was from 10 to 40% and from 5% to 30% in Elgin. The data was skewed to higher SC% in both regions (Fig. 1i).

Considering the variance in relative blossom day, the range in the Koue Bokkeveld was 16 days and in Elgin it was 14 days (Fig 1j). The data was skewed to later flowering and had an interquartile range of 2 days in both regions (Fig. 1j).

3.2 Relative bloom day (RBD) as a variable

Across the two regions the flowering time lasted for five weeks with the first flower clusters tagged on 2 October and the last on 3 November. A greater proportion of fruit in Elgin (45.2%) came from flowers at full bloom compared to that of the Koue Bokkeveld (33.0%) (Fig. 2). When the bloom dates were classed into "Early" "Full Bloom" and "Late", the number of fruit from early flowers was similar for the two regions (Elgin 29.3%; Koue Bokkeveld 30.4%; Fig. 3). The Koue Bokkeveld saw a greater proportion of its fruit being from late flowers than fruit from Elgin, with 36.6% and 25.5%, respectively (Fig. 3).

Using the categories as in Paper 1, tree position (top and bottom branch), light exposure (sun and shade) and bearing wood (shoot and spur), fruit were further split into bloom classes (Table 1). The distribution of the data does not allow for sensible analysis of variance due to low representation in some categories. In Elgin some categories (early, shaded fruit and late shaded spurs) had less than 1% of the data and top branch spurs with 9.3% and 12.3% of the fruit (Table 1). The Koue Bokkeveld had empty categories for early shaded fruit on the bottom branch, and shaded full bloom fruit on the bottom branch (Table 1). The majority of the fruit was on sun exposed spurs in both regions.

A variance component analysis was performed on RBD as a variable, with orchard, tree and position being the components. Fig. 4 shows the relative contribution of orchard, tree, position and the orchard X position interaction, on variance in RBD. It showed that more of the variation was unexplained in the Koue Bokkeveld (76%) than in Elgin (67%) (Fig. 4). For RBD in Elgin, orchard to orchard variation gave rise to 10% of the variation, tree to tree 18% and the orchard x position interaction 5%. Position did not contribute to variation in RBD in Elgin (Fig. 4). In the Koue Bokkeveld, tree to tree, position, and the orchard x position interaction contributed 12%, 6% and 6% to variation in RBA, respectively, but orchards did not contribute to variation in RBD (Fig. 4).

3.3 RBD as a variance component

Similar to paper 1, variance component analyses were performed to provide insight into the sources behind the variability described above. Again, the differences between orchards (Orchard), tree-to-tree differences (Tree) and position within the tree (Position) were considered. More importantly, RBD (Relative Blossom Day) was now also considered a source of variability for each parameter and included in the analysis. The effect of orchard, tree and position was discussed in Paper 1 and will not form part of this paper as emphasis is placed on RBD. The Position x RBD interaction was included in the analysis but explained less than 1% of the overall variance in both regions and was therefore not included in further discussions (Fig. 5 and 6). Any variability not explained by these components were seen as "Not Explained" by the model. The regions were analysed separately, and results are presented in Fig. 5 for Elgin and Fig. 6 for the Koue Bokkeveld.

In Elgin RBD gave rise to 1.6% of the variation in fruit aspect and 3.5% in mass, (Fig. 5). For lightness value, RBD contributed 0.12% of the variation in lightness at harvest and 0.00% after storage. Similarly, variance in hue angle at harvest and after storage was also poorly explained by RBD, 0.48% and 0.00% respectively (Fig. 5). RBD did influence the maturity parameters though. For SC% it contributed 7.4% to variation and in firmness at harvest and after storage it contributed 8.2% and 12.2% respectively (Fig. 5).

In the Koue Bokkeveld RBD appeared to contribute less to variation than in Elgin. For fruit aspect, 2.6% of the variation was due to RBD, and for mass it was 7.0% (Fig. 6). As in Elgin, RBD had a negligible impact on fruit colour, lightness value and hue angle, both at harvest and after storage in the Koue Bokkeveld. For maturity parameters, RBD had no effect on variation in SC% but did for firmness at harvest (8.3%) and after storage (0.65%; Fig. 6).

To depict the variability of the bloom classes, density curves are presented for each variable from Fig. 7 to Fig 11. Density curves were only drawn for parameters where RBD contributed more than 1.5% of the variation. This same threshold is later used to determine which variables to perform ANOVA's on.

Fruit aspect in Elgin showed a higher peak density for full bloom fruit than those from early and late flowers indicating greater uniformity in full bloom fruit that those from early and late flowers (Fig. 7a). In the Koue Bokkeveld, all bloom classes had a similar peak density indicating similar variation within each bloom class for fruit aspect (Fig. 7b).

Considering fruit mass, the density curves for Elgin (Fig. 8a) showed fruit borne from late flowers having a lower peak, an indication of greater variation in mass than early or full bloom fruit. In the Koue Bokkeveld early fruit peaked at a higher mass (Fig. 8b) suggesting a higher mean mass than other bloom classes but the ANOVAs showed differences between the means (Table 2). In the Koue Bokkeveld fruit from full bloom varied more than those from early or late flowers.

The SC% distribution curves in Elgin showed a low peak density for late fruit compared to early and full bloom (Fig. 9a). This suggests greater variability in SC% for fruit from late flowers in Elgin. In the Koue Bokkeveld fruit from both early and late flowers had a lower peak density than that of full bloom flowers (Fig. 9b). Fruit from full bloom flowers had the greatest uniformity in SC%.

The density curves for firmness at harvest in Elgin showed progressively declining peak density from early, to full bloom and late having the lowest peak density (Fig. 10a). The peaks occurred at different firmness values but the ANOVA later will address the differences in means. What is seen here is that fruit from early flowers had the least variation in firmness followed by full bloom fruit being more variable and late fruit having the most variation. The same progression is seen in the Koue Bokkeveld, with each successive bloom class having more variation in flesh firmness, but the difference between early and full bloom peaks are smaller than in Elgin (Fig. 10b).

After storage, the distribution curve in Elgin once again showed a progressive decline in peak density from early to late (Fig. 11a). The curves had a wider spread suggesting that firmness was more variable after storage for all bloom classes than at harvest. The Koue Bokkeveld showed lowering of peak densities, with early fruit dropping the most relative to at harvest (Fig. 11b). Full bloom fruit had the highest peak density and duly had the most uniform firmness after storage in the Koue Bokkeveld than the other bloom classes.

To compare the means, ANOVA results are presented in Table 2 for the variables where RBD contributed more than 1.5% to the variation. Fruit aspect in Elgin showed that fruit borne from full bloom flowers had the greatest aspect ratio (taller) (*p*=0.016; Table 2). In the Koue Bokkeveld early and full bloom flowers yielded fruit with the same aspect ratio, and fruit from late flowers had a significantly smaller aspect ratio (flatter) (*p*<0.0001; Table 2). In both regions full bloom flowers had the tallest fruit.

In Elgin, the fruit mass between the bloom classes differed significantly (*p*<0.0001; Table 2). Fruit borne on early flowers had a greater mass than fruit from full bloom and late flowers (Table 2). Full bloom and late flowers were similar (Table 2). With a p-value of <0.0001, bloom classes showed significant differences in the Koue Bokkeveld (Table 2). Early and full bloom flowers produced the heaviest fruit with late flowers yielding fruit of significantly lower mass.

Regarding the maturity parameter, SC%, the ANOVA results showed significant differences for Elgin (*p*<0.0001) but not the Koue Bokkeveld (*p*=0.101; Table 2). Starch conversion in Elgin showed a decline from early, mid, to late flowers (Table 2). In Elgin, fruit firmness had significant differences between bloom classes both before and after storage with a p<0.0001 (Table 2). At harvest the early fruit had the lowest firmness followed by firmer full bloom fruit; fruit from late flowers had the highest firmness. The same pattern is presented after storage with early fruit having the lowest firmness, but full bloom and late fruit did not differ significantly from one another. This points to a greater variation in fruit firmness after storage compared to at harvest. In the Koue Bokkeveld fruit firmness both at harvest (p=0.0027) and after storage (p<0.0001) differed amongst the bloom classes (Table 2). At harvest the early and full bloom fruit did not differ significantly from one another but the late fruit were significantly firmer than early fruit. The same pattern was seen after storage where early and full bloom did not differ in firmness but late fruit were significantly firmer than both early and late fruit in the Koue Bokkeveld. As in Elgin, the differences in firmness for the bloom classes in the Koue Bokkeveld appeared larger after storage than at harvest.

The correlations between the parameters for each bloom class are presented in Appendix B and only the relationships that are of significant commercial value are discussed. SC% and flesh firmness at harvest showed significant correlations (Fig. 12). At harvest, all bloom classes in Elgin showed a significant negative correlation between SC% and firmness (early: r=-0.59, *p*=0.001; full bloom: r=-0.69, *p*<0.0001; late: r=-0.82, *p*<0.0001; Fig. 12a). The strength of the correlation was moderate for early fruit, strong for full bloom fruit and very strong for late fruit. In the Koue Bokkeveld, only full bloom fruit showed a significant negative correlation of moderate strength (full bloom: r=-0.51, *p*=0.02; Fig. 12). After storage the correlation between firmness and SC% at harvest was significant but weaker in Elgin for full bloom (moderate) and late fruit (strong) and the correlation for early fruit was not significant (full bloom r=-0.51, *p*=0.011; Late r=-0.61; *p*<0.0001) (Fig. 13a). There were no significant correlations for SC% at harvest and firmness after harvest in the Koue Bokkeveld (Fig. 13b).

Correlations between fruit firmness at harvest and firmness after storage are shown in Fig. 14. In Elgin (Fig. 14a), firmness of early fruit at harvest did not correlate with firmness after storage but showed a strong positive correlation for full bloom fruit (r=0.76; *p*<0.0001*)* and a moderately positive correlation for late fruit (r=0.57; *p*=0.001). In the Koue Bokkeveld, only full bloom fruit showed a significant strong correlation (r=0.61; *p*=0.026 (Fig. 14b).

In both regions, mass and firmness at harvest showed significant negative correlations for each bloom class. Early fruit in Elgin had a weak correlation (r=-0.37; *p*=0.03), but for full bloom (r=-0.73; *p<0.0001*) and late fruit (r=-0.72; *p<0.0001*) the correlations were strong (Fig. 15a). In the Koue Bokkeveld the correlation was very strong for early fruit (r=-0.82; *p<0.0001*), strong for full bloom fruit (r=-0.72; *p*=0.001) and moderate for late fruit (r=-0.61; *p*=0.002; Fig.15b). Mass and firmness after storage also had significant correlations across bloom classes bar early fruit (*p*=0.21) from the Koue Bokkeveld (Fig. 16). Early (r=-0.54; *p=0.01*), full bloom (r=-0.81; *p<0.0001*) and late (r=-0.70; *p<0.0001*) fruit from Elgin had moderate, very strong, and strong correlations respectively (Fig. 16a). Full bloom fruit (r=-0.79; *p<0.0001)* from the Koue Bokkeveld had a strong correlation between mass and firmness after storage with late fruit (r=-0.70; *p=0.02)* having a moderate correlation (Fig. 16b).

4. Discussion

4.1 Overall variability

The parameters measured showed large variation, with the variance component analysis showing high unexplained variance. RBD explained less of the variation compared to orchards and trees. It would be prudent to highlight how the methodology could possibly affected the variability. Initial fruit set, as well as fruit thinning could have reduced the variation in mass, as the instruction given to labourers is to remove the smallest fruit from each cluster. Changes in light exposure during the season could also have influenced the quality of the sampled fruit. In this study fruit were categorised according to the light exposure at harvest and this could not account for the seasonal change in exposure as shoots grew, and fruit that were sun-exposed early in the season would later be classified as “shaded” fruit. This change in exposure through the season can influence the colour development as shown for ‘Granny Smith’ apples in a study by Fouché *et al*, (2010), where fruit that were exposed early in the season and then later shaded were greener than fruit that were shaded for the whole season. In our study, both the fully shaded and more recently shaded fruit would be classified as “shaded” fruit thereby increasing variation in green colour, amongst other things, within the “shaded” category.

Consumers are sensitive to the appearance of fruit when making purchasing decisions, but their perception of the quality is negatively influence by variations in quality within a bag or carton of fruit (Richards and Patterson, 2000; Richards 2000). The variation in appearance parameters (fruit aspect, mass and colour) can easily be sorted after harvest in the pack house with existing computer vision systems and weight sensors (Zhang at al., 2014). The challenge arises with maturity parameters, SC% and firmness, which are typically measured destructively preventing them from being used as a sorting metric. It is possible to detect some of these parameters using hyperspectral imaging as detailed in the review by Wang *et al*. (2015). The use of these techniques is not yet implemented in packing systems shifting attention to the causal factors in variation to identify and address them.

Some measures exist to address the variation before harvest by using either aminoethoxyvinylglycine (AVG; Venburg *et al*., 2008) or 1-methylcyclopropene (1-MCP; Varanasi *et al*., 2013) to delay and thereby homogenise fruit maturation. AVG delays maturation by temporarily inhibiting ethylene biosynthesis and needs to be applied 3-4 weeks before anticipated harvest for optimal effectivity in ‘Golden Delicious’ whereas 1-MCP is an ethylene competitive inhibitor with a recommended application date of 1 week before anticipated harvest. Both chemicals can operate independently but Yuan and Carbaugh (2007) showed that a combination treatment of each product had the best results in slowing the ripening process to delay maturation date. Wang and Dilley (2001) showed that a combination of AVG and ethephon (and ethylene-releasing molecule that stimulates ripening) resulted in greater uniformity in terms of flesh firmness in ‘Gala’ apples.

Since fruit are harvested on the same day, but not all flowers on the tree bloomed simultaneously, attention must be given to blossom date as a possible source of variation. Grab and Craparo (2011) looked at the effect of warming climate from 1973 to 2009 in South Africa on full bloom dates and showed that full bloom dates advanced on average by 1.6 days per decade for the period measured. Studies on quantifying the duration of bloom were not found. Elgin received 965 Utah chill units (CU) before the 2018 season, and the Koue Bokkeveld 1199 Utah CU accumulated before the 2018 season (Hortgro, 2017). It is typical of apples produced in mild winter regions to have delayed and erratic bud break and protracted bloom (Cook, 2007). Duly it could be assumed that the Koue Bokkeveld would have a more condensed bud break than Elgin but this was not the case with a greater proportion of fruit from full bloom flowers in Elgin than in the Koue Bokkeveld. The explained variation in RBD was greater in Elgin than in the Koue Bokkeveld. It may be that execution of thinning strategies differed between the relevant growers, since in Elgin where producers believe, they are predisposed to having smaller fruit take a harder line when thinning off the smallest fruitlets. Temperature after rest breaking application is important determining the efficacy of the treatment (Erez *et al.,* 1971). Temperatures were not logged in the orchards but some of the differences observed may be from the influence of the early spring temperature.

4.2 Effect of RBD on fruit appearance

Fruit aspect ratio is the relationship between length and diameter of a fruit. A larger fruit aspect ratio means fruit will have a relatively smaller diameter for a given length and appear “taller” whereas fruit with a small aspect ratio appear “flatter” to the eye. The expression of fruit shape is predominantly genetic and will vary from cultivar to cultivar, but environmental conditions can influence the phenotype (Brown, 1960). Apple fruit shape is typically independent of growing season temperatures (Warrington *et al*., 1999) but Petri and Leite (2003) showed that the temperature conditions during winter dormancy can influence final fruit shape, with warmer temperatures resulting in smaller aspect ratios, producing ovoid shaped fruit. In our study, the RBD accounted for some of the variation in fruit aspect; “full bloom” fruit from both regions and “early” fruit from the Koue Bokkeveld had the taller “more typical” shape of a ‘Golden Delicious’. When considering the findings of Paper 1, most of the variation was caused by regional differences (fruit aspect in the Koue Bokkeveld was greater than in Elgin) with orchards and trees contributing to a lesser extent. This difference could be attributed to the winter conditions being milder in Elgin (965 Utah CU) than the Koue Bokkeveld (1199 Utah CU) concurring with finding of Petri and Leite (2003). These differences suggested that fruit from full bloom flowers in the milder winter area was of preferred quality in terms of fruit aspect. These fruit were similar in shape to fruit from the early flowers in the Koue Bokkeveld production region. Full bloom and early fruit from the colder production region were tallest, and there was less variability in the fruit aspect ratio in the cooler winter climate. This is unlikely to affect consumer choices though as Hampson and Quamme (2000) showed in their consumer study in Canada, that apple fruit shape had little influence on preference.

The ultimate fruit size at harvest is an important quality parameter that is influenced by temperature in the dormant and growing season. Fruit size is visually assessed by the consumer, and fruit mass is used in practise to indicate size in a pack house. Typically (but not always), larger fruit are more desirable to the consumer but there is no evidence that larger fruit are of superior quality as quality is a combination of many parameters. Preference for specific sizes varies between population with Canadians and most central Europeans preferring larger apples (Hampson and Quamme, 2000) and Scandinavians opting for smaller apples (Redalen, 1987). For the farming unit, larger fruit, if from the same number of fruit, results in more kilograms produced per unit area and hence larger fruit is desirable. With larger fruit a cost reduction due to efficiency gains is seen at harvest, storage and packing. In a kiwi fruit study, Richardson *et al*. (2019) reported that kiwi fruit from early opening flowers had a greater mass and matured sooner than those that opened in the middle and late end of the bloom window. Similarly, our study showed RBD influenced final fruit mass in both regions but to a lesser extent than in the kiwi fruit study. Our differences between bloom classes were small but did show a similar pattern of late flowers resulting in smaller fruit than early flowers. Reginato *et al*. (2019) showed that the maximum average winter temperature is the most important in determining fruit size with a secondary effect of increased season length with a lower average daily maximum temperature producing larger fruit. In our study the early flowers produced relatively heavier fruit than other bloom classes, pointing to season length being causal. To detect mechanisms at play other than season length, comparing the flower quality at blossom could prove useful.

4.3 Effect of RBD on fruit maturity:

Apple fruit maturity is an important parameter when determining the harvest date of an orchard. The biological composition of the fruit at harvest determines many of the post-harvest qualities and storability of the fruit. Two important metrics for determining apple fruit maturity are SC% and fruit firmness.

As apple fruit ripen and enter maturation, the accumulated starch is hydrolysed to sugar (Krotkov and Helson, 1946), which is responsible for the sweet taste that makes apples desirable. A simple starch test by a horizontally slicing an apple and covering it with an iodine solution will reveal the sugar content as a white surface against the black starch background can be scored as a percentage starch breakdown. Fruit flesh firmness is used across a wide range of fruit crops, but especially in pome and stone fruits (Sams, 1999) as a maturity indicator as the cell walls separating during ripening coincides with a decrease in fruit firmness. In apples, it correlates well with eating satisfaction and is a useful measure of maturity, quality and storability (Wills *et al.,* 1980). For apple producers in South Africa, the export policy for 'Golden Delicious' states a minimum average SC% of 15% and a firmness minimum of 6.8 kg (Department of Agriculture, Forestry and Fisheries (DAFF), 2013). Even though a maximum threshold is not set for SC%, packhouses impose guidelines to receive more uniform batches of fruit. These guidelines are typically more stringent than those set out by DAFF as overripe fruit are more likely to develop storage disorders. To further increase uniformity, the fruit harvested form orchards of advanced maturity are often handled separately from fruit harvested from orchards at optimal maturity. Even with the use of pre-sorting equipment, this practise remains relevant.

When "strip-picking" fruit from early flowers would typically have a longer growing season (more days of growth and development) and would be expected to be the first to mature. Since firmness and SC% are related to maturity, it is expected that relative bloom date should influence this. RBD had the greatest contributions to variation in firmness and SC% compared to the other parameters measured. SC% in both regions was most uniform in full bloom flowers but for firmness at harvest an increase in variation was apparent from early, to full bloom to late fruit. As anticipated, bloom classes influenced SC% in Elgin, but in the Koue Bokkeveld SC% was consistent across all the classes. This was surprising as the Koue Bokkeveld had a more protracted bloom period with less of its fruit in the full bloom class. A similar pattern was seen for firmness in both regions with a progressive decrease in firmness in fruit from early to late flowers. The trend was less evident in in the Koue Bokkeveld. The time of on-tree development had a greater impact on fruit in Elgin than the Koue Bokkeveld. Perhaps heat units during the growing season had a stronger effect on development than the winter cold, but the growing season temperatures were not recorded for this study.

Fruit firmness at harvest and SC% correlated significantly in Elgin with R-values becoming progressively larger from early to late fruit with a similar pattern for firmness after storage. Late fruit had the strongest correlations between SC% and firmness in Elgin, but in the Koue Bokkeveld this correlation was only present for firmness at harvest of full bloom fruit. This suggests that RBD had little effect on maturity in the Koue Bokkeveld but a notable effect in Elgin. It is similar for the correlations between firmness at harvest and firmness after storage, where again correlations were stronger in Elgin than in the Koue Bokkeveld adding more evidence that RBD was a stronger determining factor in maturity in Elgin than the Koue Bokkeveld.

Literature notes a negative correlation between apple fruit size and fruit firmness (Blanpied *et al.,* 1978). This correlation was also present in our results, and all fruit, regardless of flower class, showed a negative correlation between fruit mass and firmness before and after harvest. Apple fruit that develop over a shorter period of time are expected to be smaller compared to fruit with a longer growing season. Tomala *et al*. (1992) focused their study primarily on bitter pit but also showed bloom date relative to full bloom effecting fruit size; where fruit from later flowers were notably smaller. Reginato *et al*. (2019) also showed that season length affected fruit mass with longer seasons producing larger fruit. Thus, with strong correlations between the firmness and maturity and less mature fruit being firmer, it appeared that the later flowers produced smaller fruit that were less mature (Knee and Smith., 1989). Protracted bloom can exacerbate this phenomenon as the later fruit have size development curtailed. Maggs (1975) showed that a fruit’s potential weight decreases by 2 grams for every day’s delay after full bloom. Since date of blossom influences both mass and physiological maturity it is reasonable to assume that the number of days the fruit spent on the tree (from blossom to harvest) had a strong bearing on these qualities.

5. Conclusion

RBD (as a variable) showed large variation within and across production regions, and when determining the sources of variation, much of variability remained unexplained for both regions. Variability of ‘Golden Delicious’ fruit in terms of quality and maturity was high and compared to orchard and tree differences, RBD explained a relatively small portion of the overall variability in ‘Golden Delicious’ fruit. This is in itself rather interesting since we have always assumed that protracted bloom under low chill conditions in South Africa would contribute significantly to variation in fruit maturity.

Fruit aspect was affected by the RBD, indicating that it has an effect on the appearance properties of the fruit. Correlations of maturity parameters (SC% and Firmness) were strongest for fruit from full bloom flowers and early flowers resulted in earlier maturation.

The effect of RBD on final fruit mass is commercially consequential. Early fruit in Elgin constituted a third of the volume harvested and were 7.5% larger in size than full bloom and late fruit. In the Koue Bokkeveld, late fruit were 14% lighter than those of early and full bloom flowers. Thus, shifting bloom of late flowers earlier in a well yielding orchard (90 t/ha), this could result in an additional 4.8 t/ha in Elgin and 4.5 t/ha in the Koue Bokkeveld due to the increased mass alone. This is assuming that the cause in increased yield was due to having a longer development time and not accounting for possible differences in flower quality among the early, full bloom and late flowers. Paper 3 provides an investigation into the flower quality of early and late flowers.

Typically, orchards in South Africa are thinned chemically from petal drop targeting mostly the late flowers. This is clearly not as effective as desired, as up to one third of the fruit on the tree at harvest came from late flowers. Perhaps adapting thinning techniques or further developing mechanical thinning to a point of utility, fewer of the fruit from late flowers will be left on the trees and a more uniform crop will be present. Spur thinning in spring, or artificial spur extinction (ASE), is effective in higher density orchards (>2500 trees per hectare) and results in predictable fruit set early in the season (Breen *et al*., 2012). Breen *et al*. (2012) showed that after ASE was applied chemical thinning was no longer necessary and the resultant hand thinning was less complex. ASE allows for the removal of the weakest positions and should therefor increase the uniformity in fruit quality.

This study established that relative blossom day has an effect on fruit quality parameters, but further work on flower quality at different bloom times from different positions is needed to uncover the mechanisms at play. The length of the growing season (time of on tree development) plays a role in determining final fruit quality. Additionally, investigating flower quality among the different positions, could yield valuable information.

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Tables

Table 1: The percentage of fruit from each bloom class in Elgin and the Koue Bokkeveld (KB) split into tree position (top and bottom), light exposure (sun and shade), and bearing wood (shoot and spur).

Tree position	Light Exposure	Bearing wood	Early		Full Bloom		Late	
			Elgin	KB	Elgin	KB	Elgin	KB
Bottom	Sun	Shoot	1.87	1.38	3.4	2.02	3.91	3.72
		Spur	5.87	9.45	7.23	10.08	3.83	7.54
	Shade	Shoot	0.68	0	1.79	0	1.62	0.42
		Spur	0.77	0.11	1.45	0	0.94	0.42
Top	Sun	Shoot	1.45	0.53	3.06	1.59	1.45	2.34
		Spur	8.17	11.57	9.28	11.15	3.57	10.62
	Shade	Shoot	2.47	2.12	6.64	2.97	4.94	3.82
		Spur	8	5.2	12.34	5.2	5.28	7.75

Table 2: Summarised ANOVA results of Elgin and Koue Bokkeveld, showing the LSD, F and *p*-values of all the variables with flower class as a factor.

Parameter	Aspect Ratio		Mass (g)		Starch conversion (%)		Firmness at Harvest (kg)		Firmness after Storage (kg)	
Region	Elgin	KB	Elgin	KB	Elgin	KB	Elgin	KB	Elgin	KB
Early flowers	0.944 b	0.995 a	129.00 a	122.43 a	27.72 a	25.79 a	7.30 c	7.39 b	4.96 b	5.28 b
Full Bloom	0.952 a	0.990 a	122.41 b	120.54 a	22.03 b	30.49 a	7.50 b	7.51 ab	5.34 a	5.07 b
Late flowers	0.942 b	0.973 b	119.25 b	103.18 b	17.84 c	29.71 a	7.78 a	7.64 a	5.43 a	5.52 a
LSD	0.008	0.009	4.35	5.56	3.56	4.88	0.16	0.14	0.16	0.22
F-value	4.17	13.38	9.07	29.38	12.99	2.3	14.65	5.98	17.42	10.48
p-value	0.016	<0.0001	<0.0001	<0.0001	<0.0001	0.101	<0.0001	0.003	<0.0001	<0.0001

Figures

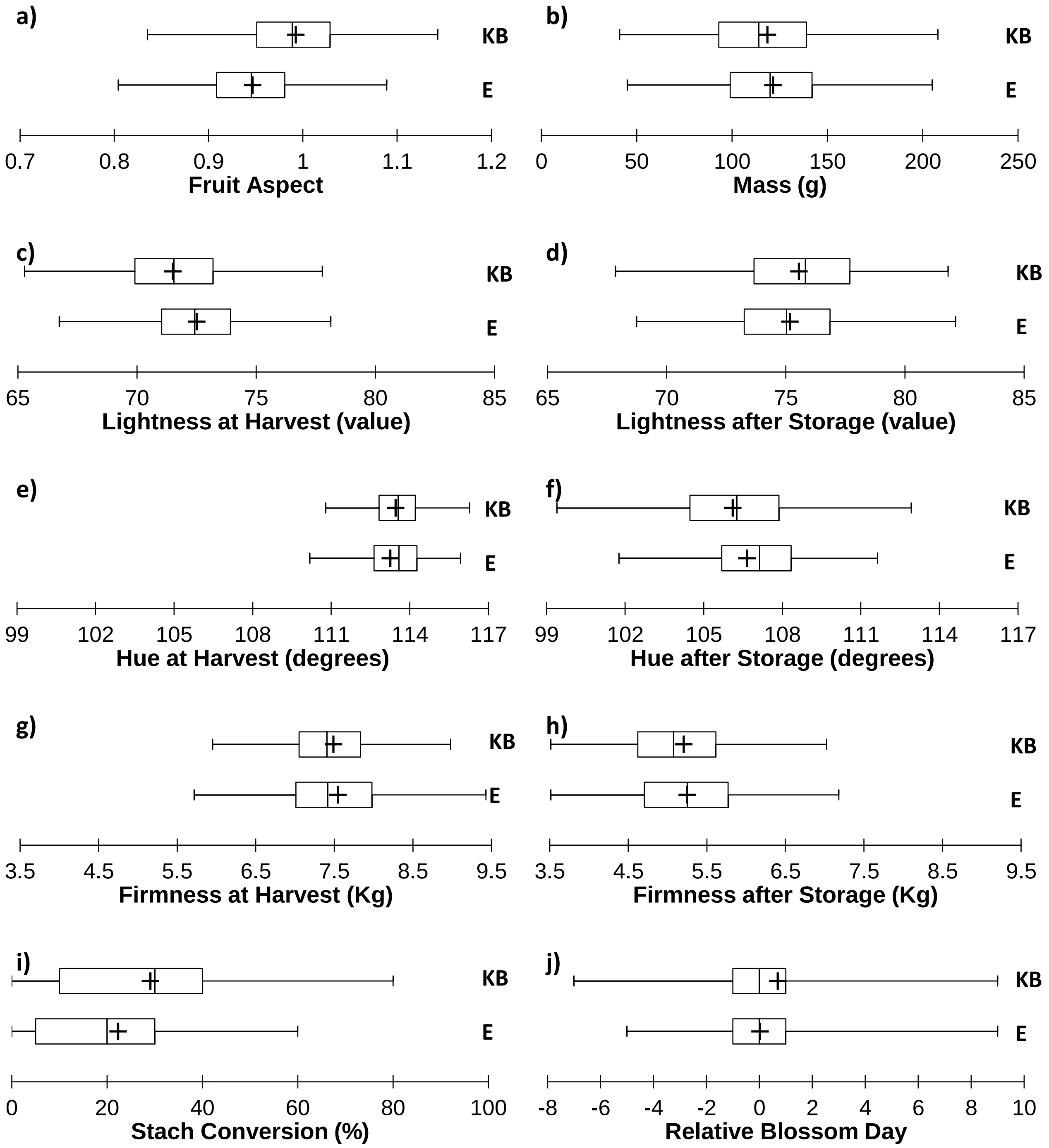

Figure 1: Box and whisker plot of fruit aspect (a), mass (b), lightness at harvest (c), lightness after storage (d), hue at harvest (e), hue after storage (f), firmness at harvest (g), firmness after storage (h), starch conversion (i), and blossom day (j) for the Koue Bokkeveld (KB) and Elgin (E). The box plot splits the data in four quartiles with an equal number of data points in each. The vertical line in the box is the median and the + indicates the mean.

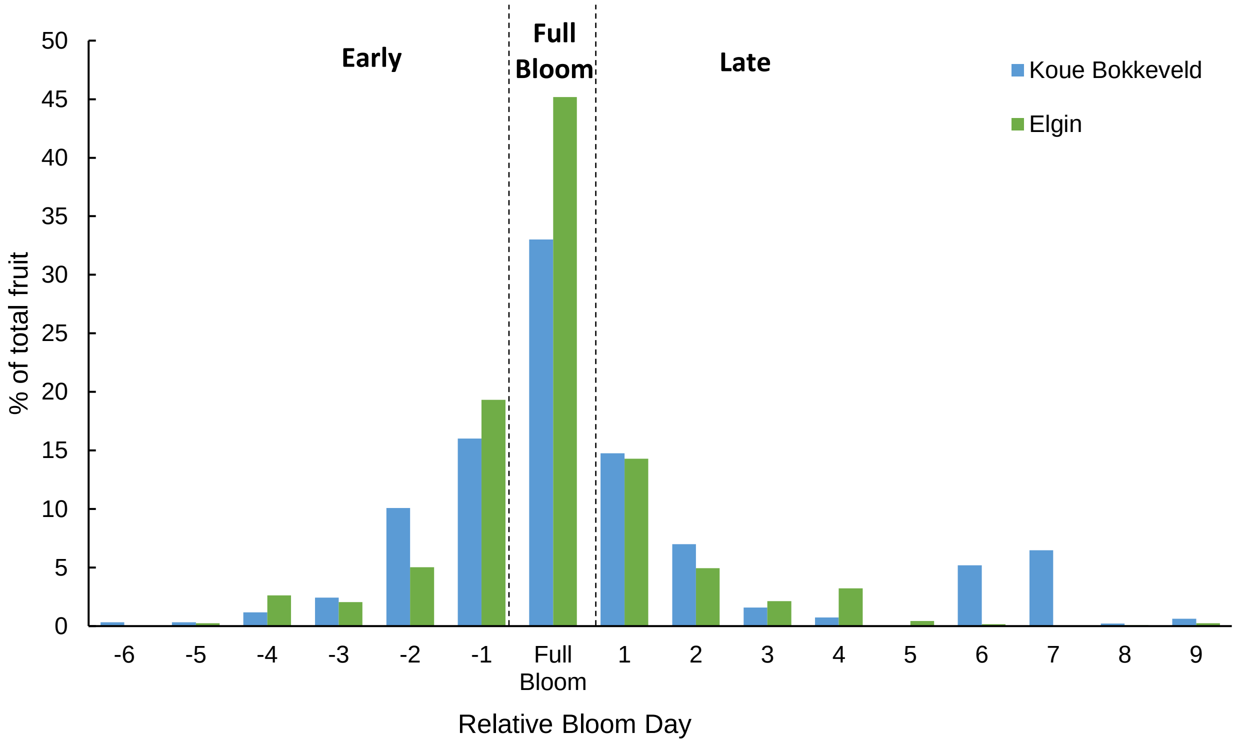

Figure 2: The percentage of fruit harvested from for each relative blossom day in the Koue Bokkeveld and Elgin. Full bloom dates were aligned to day zero and fruit from flowers that opened earlier are marked with a negative number of days and fruit that were from later flowers indicated with a positive number. The dotted line separates the bloom classes.

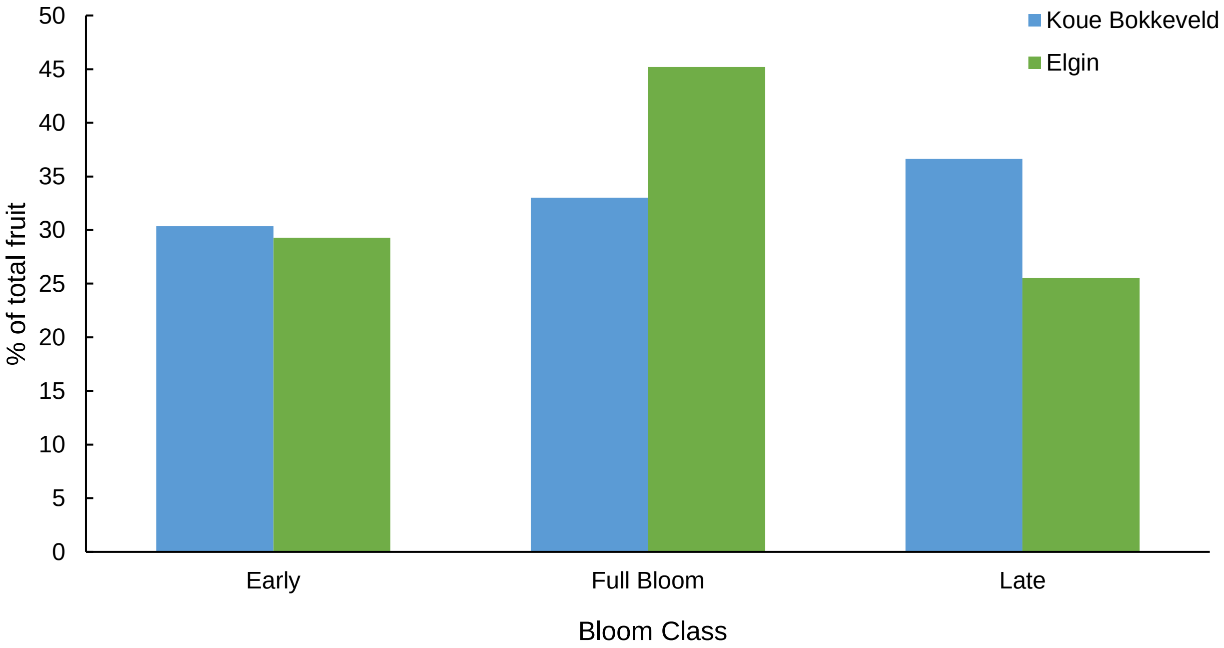

Figure 3: The percentage of fruit in each bloom class (Early, Full Bloom and Late).

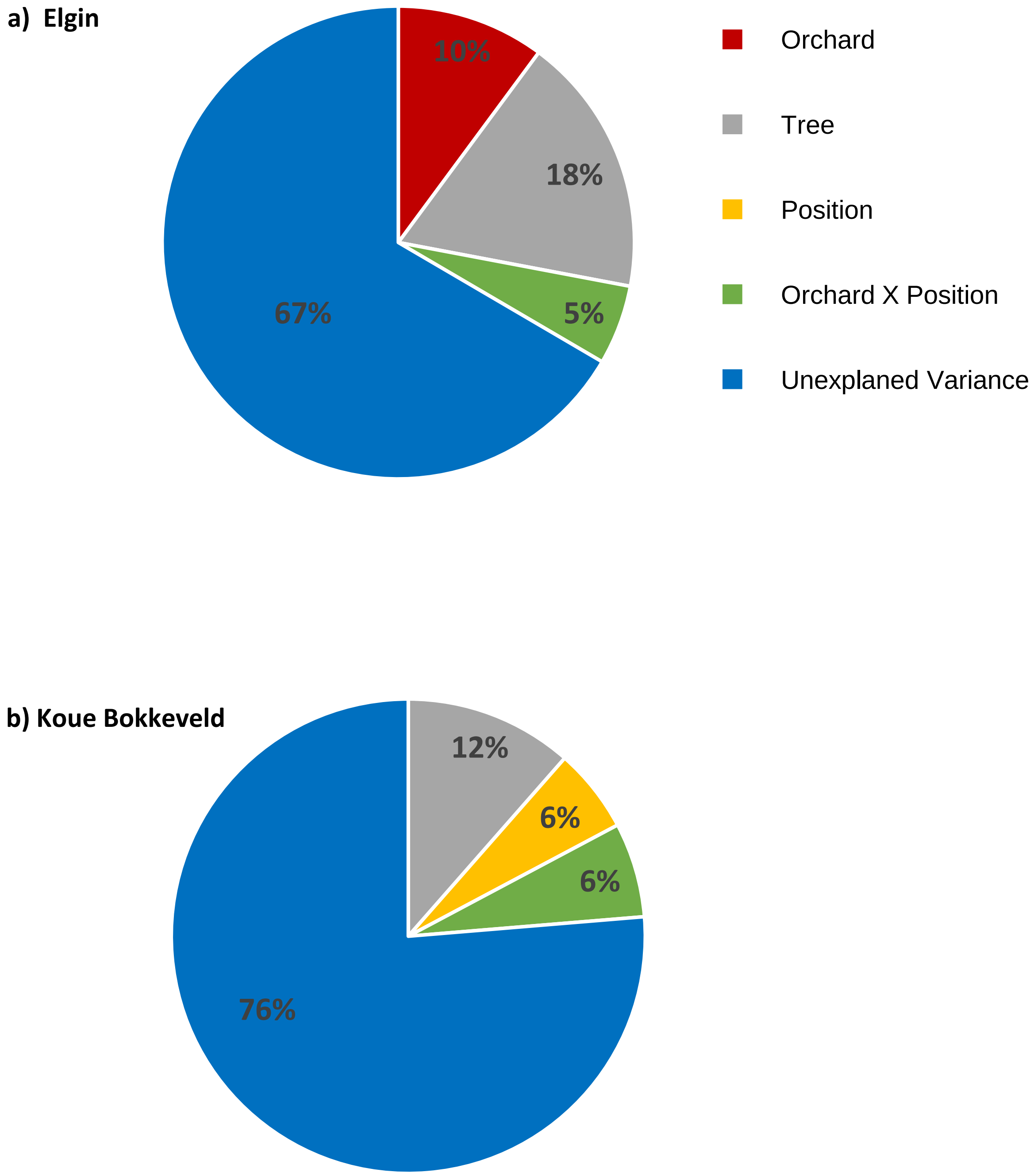

Figure 4*:* Pie chart showing the variance component analysis for Elgin (a) and the Koue Bokkeveld (b), indicating the percentage contribution of each component towards Relative Bloom Date as a variable.

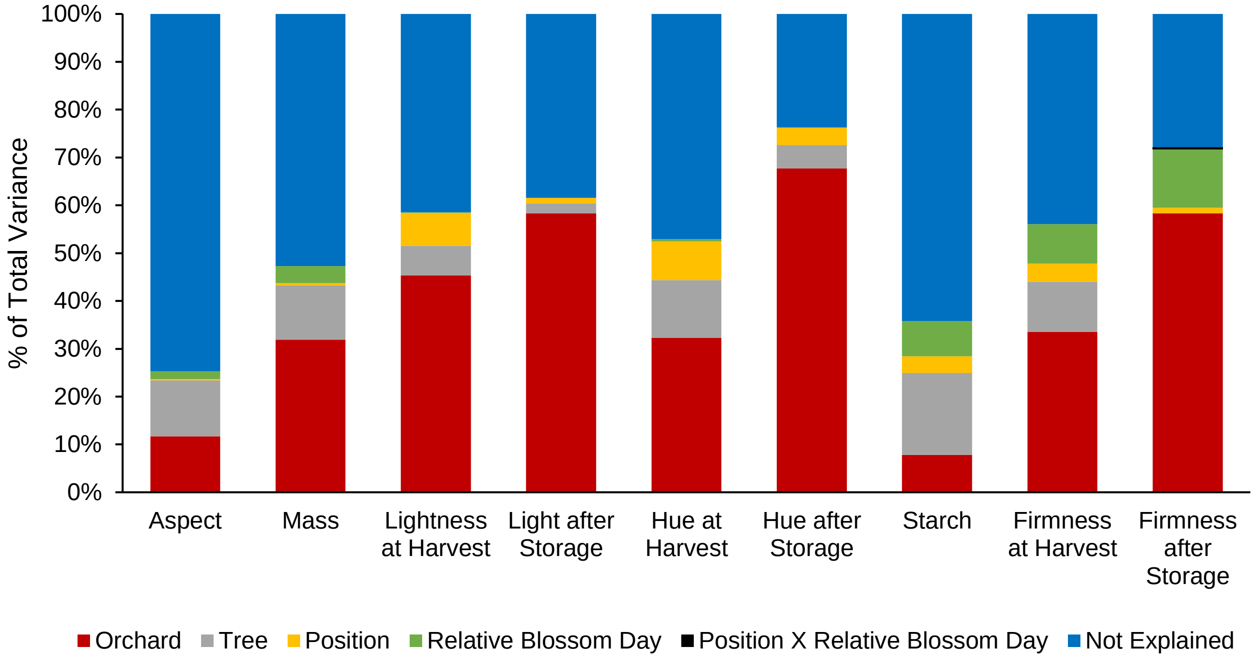

Figure 5: Variance component analysis for **Elgin**, indicating the percentage contribution of each component towards the measured variable.

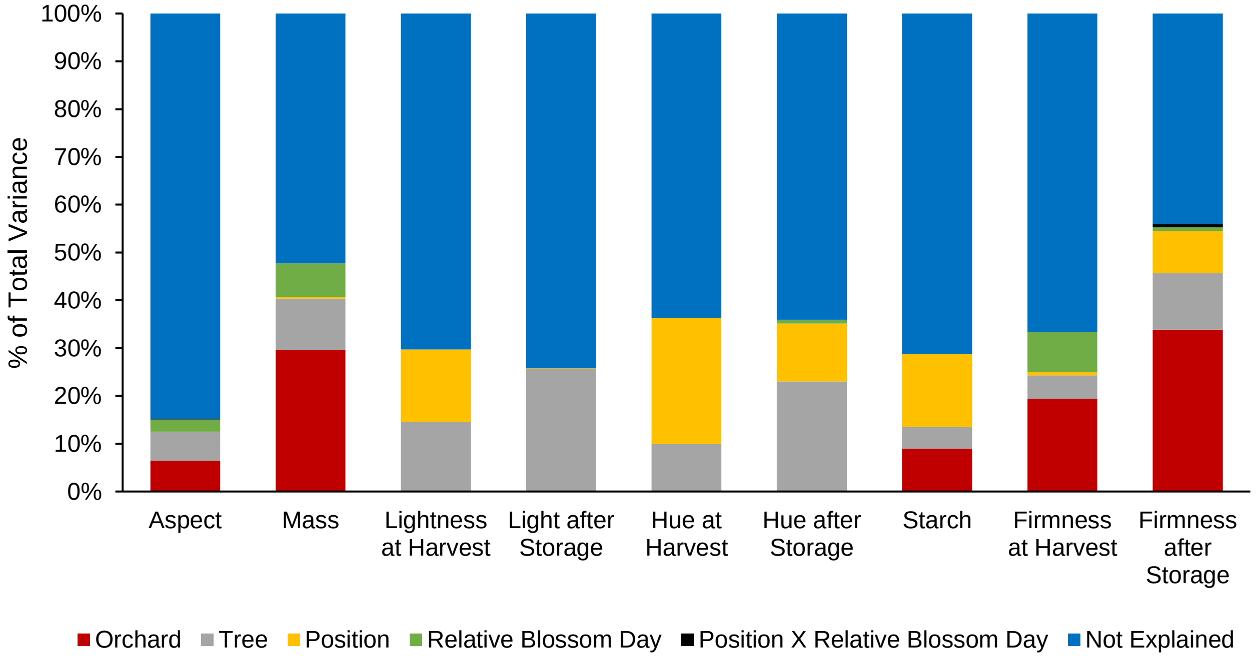

Figure 6: Variance component analysis for the **Koue Bokkeveld**, indicating the percentage contribution of each component towards the measured variable.

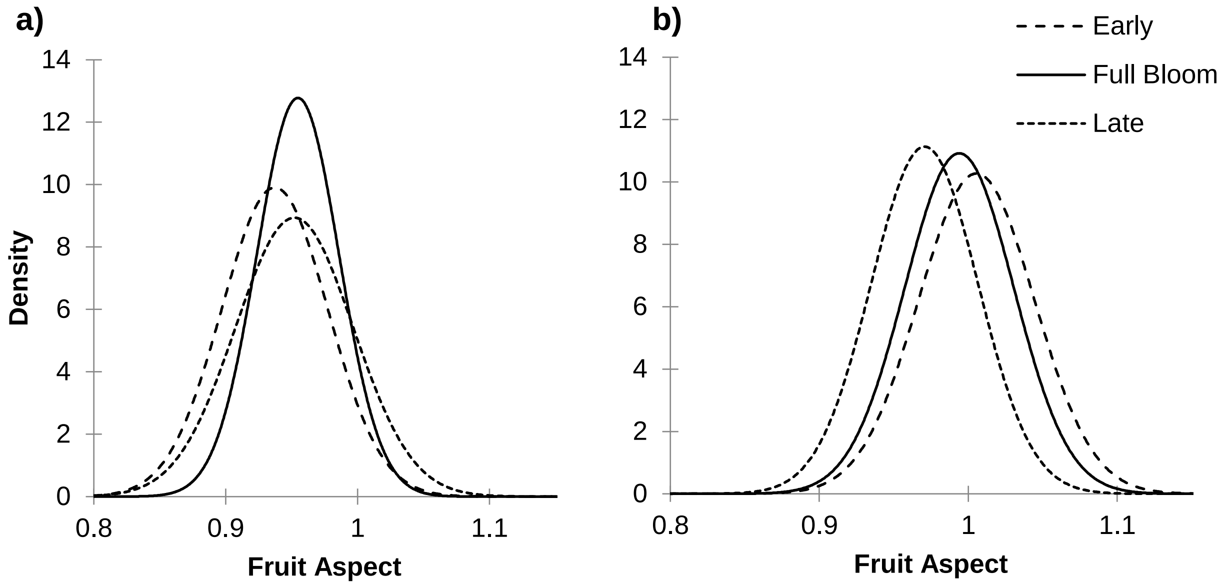

Figure 7: Density curves showing the distribution of **fruit aspect** for **Elgin** (a) and the **Koue Bokkeveld** (b) for each bloom class (Early, Full Bloom and Late).

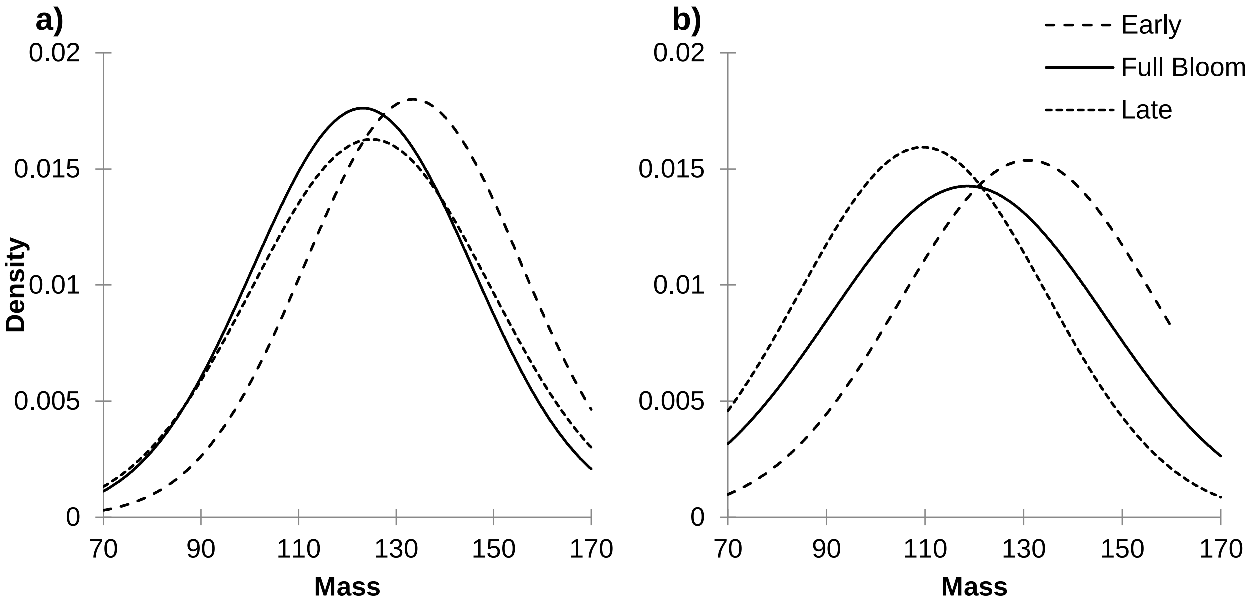

Figure 8: Density curves showing the distribution of **fruit mass** for **Elgin** (a) and the **Koue Bokkeveld** (b) for each bloom class (Early, Full Bloom and Late).

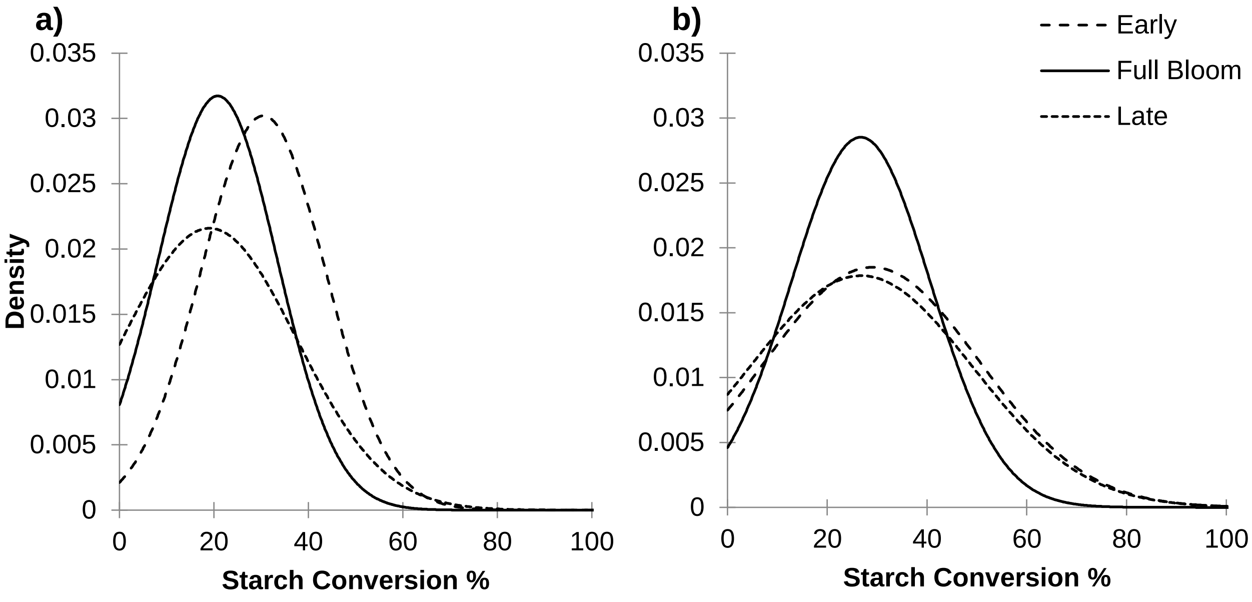

Figure 9: Density curves showing the distribution of **starch conversion percentage** for **Elgin** (a) and the **Koue Bokkeveld** (b) for each bloom class (Early, Full Bloom and Late).

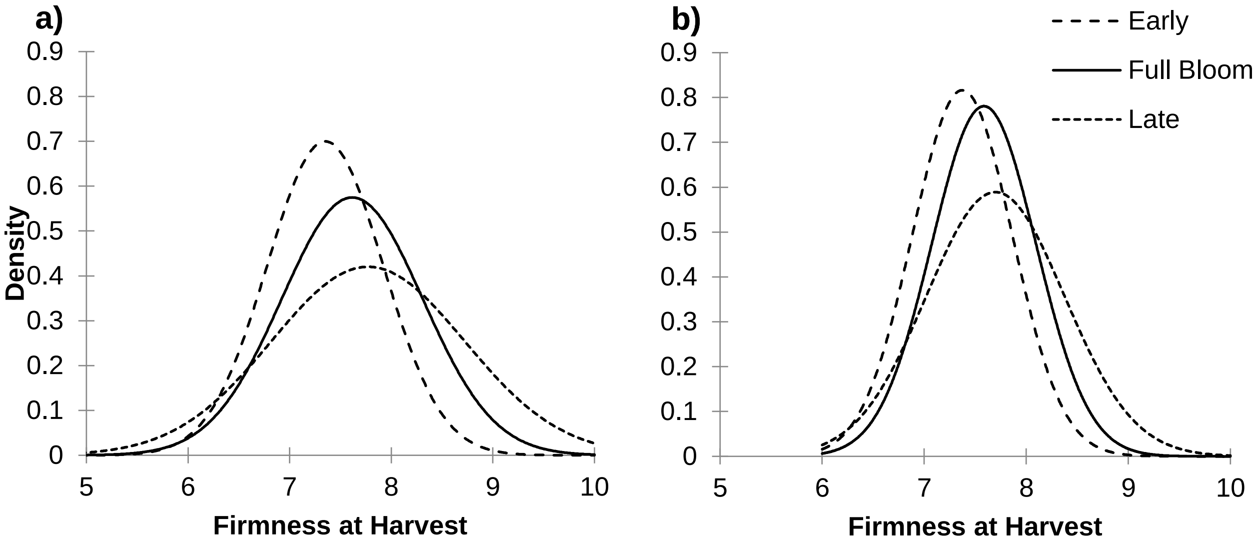

Figure 10: Density curves showing the distribution of **firmness at harvest** for **Elgin** (a) and the **Koue Bokkeveld** (b) for each bloom class (Early, Full Bloom and Late).

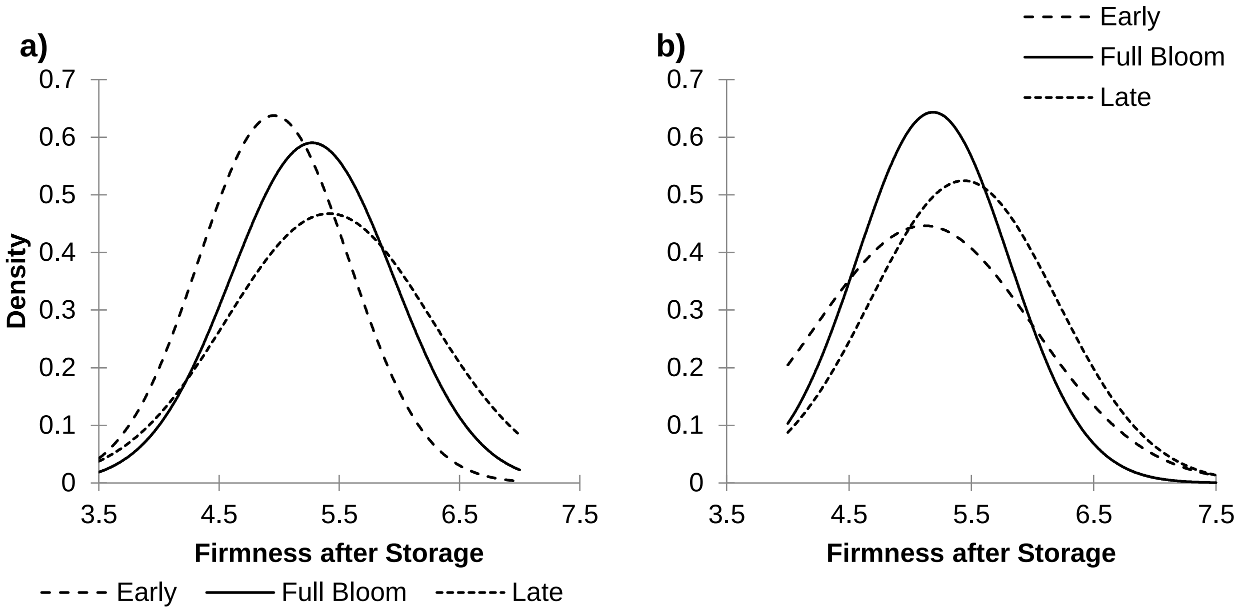

Figure 11: Density curves showing the distribution of **firmness after storage** for **Elgin** (a) and the **Koue Bokkeveld** (b) for each bloom class (Early, Full Bloom and Late).

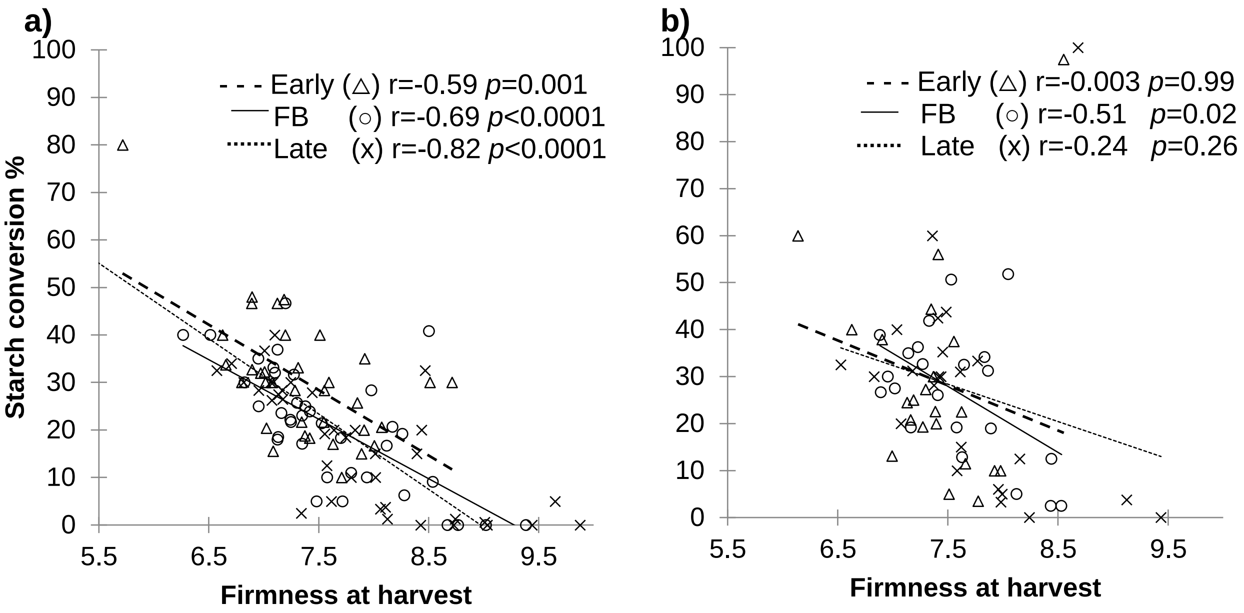

Figure 12: Scatter plot showing the correlations between **starch conversion percentage** and **firmness at harvest** for **Elgin** (a) and the **Koue Bokkeveld** (b) for each bloom class (Early, Full Bloom (FB) and Late).

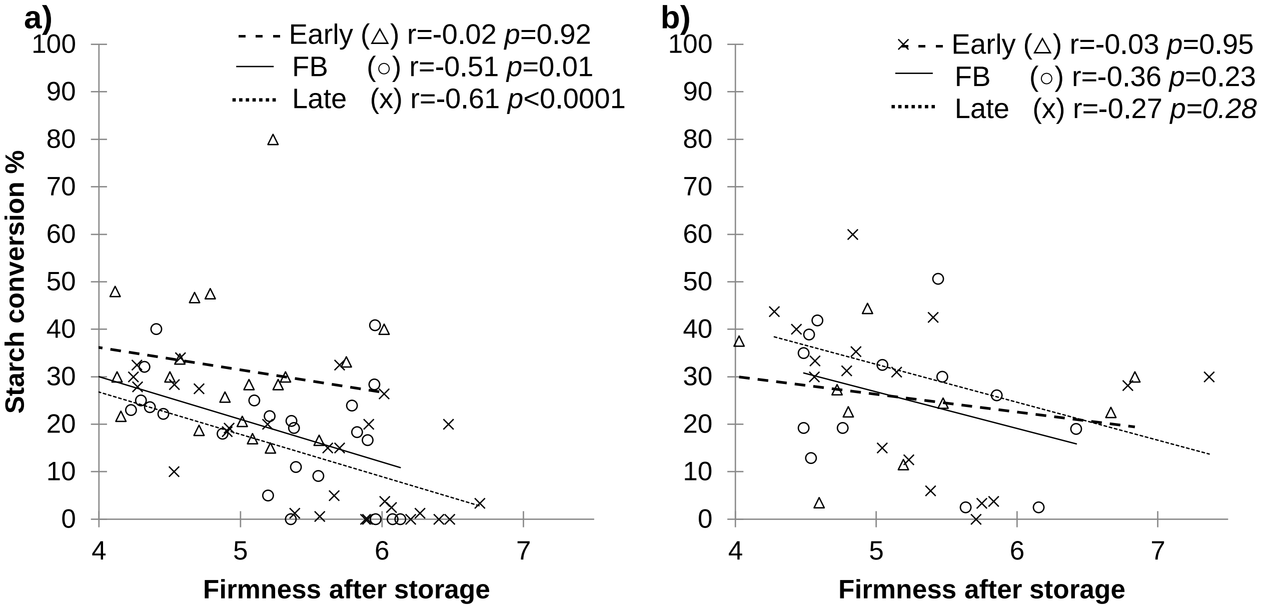

Figure 13*:* Scatter plot showing the correlations between **starch conversion percentage** and **firmness after storage** for **Elgin** (a) and the **Koue Bokkeveld** (b) for each bloom class (Early, Full Bloom (FB) and Late).

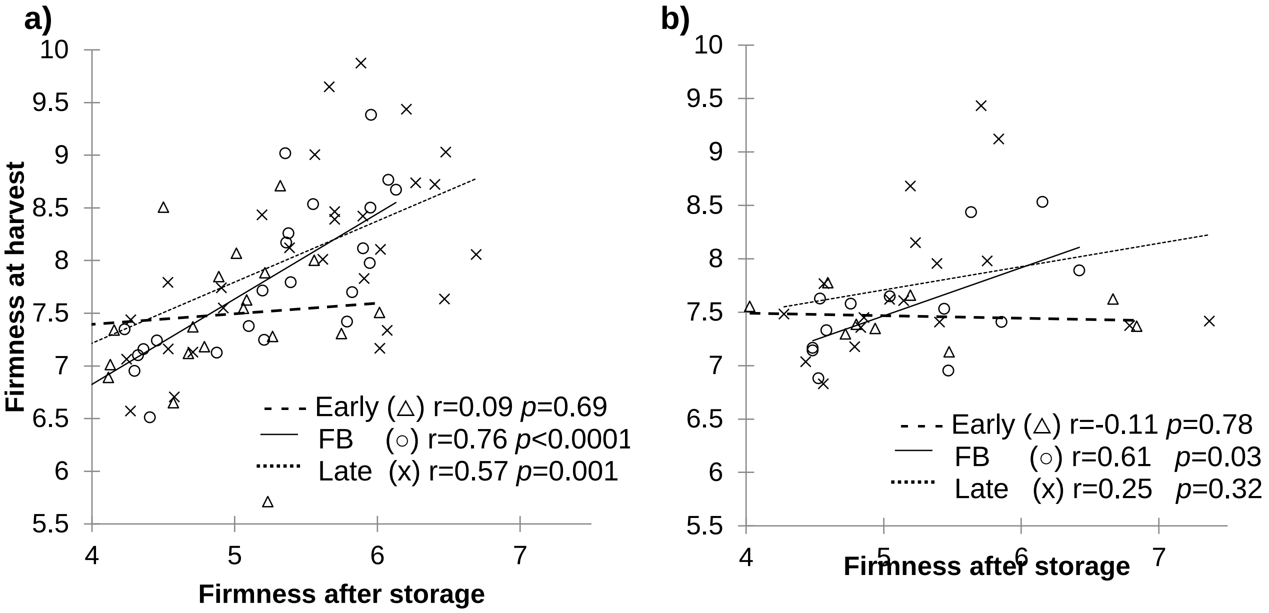

Figure 14: Scatter plot showing the correlations between **firmness at harvest** and **firmness after storage** for **Elgin** (a) and the **Koue Bokkeveld** (b) for each bloom class (Early, Full Bloom (FB) and Late).

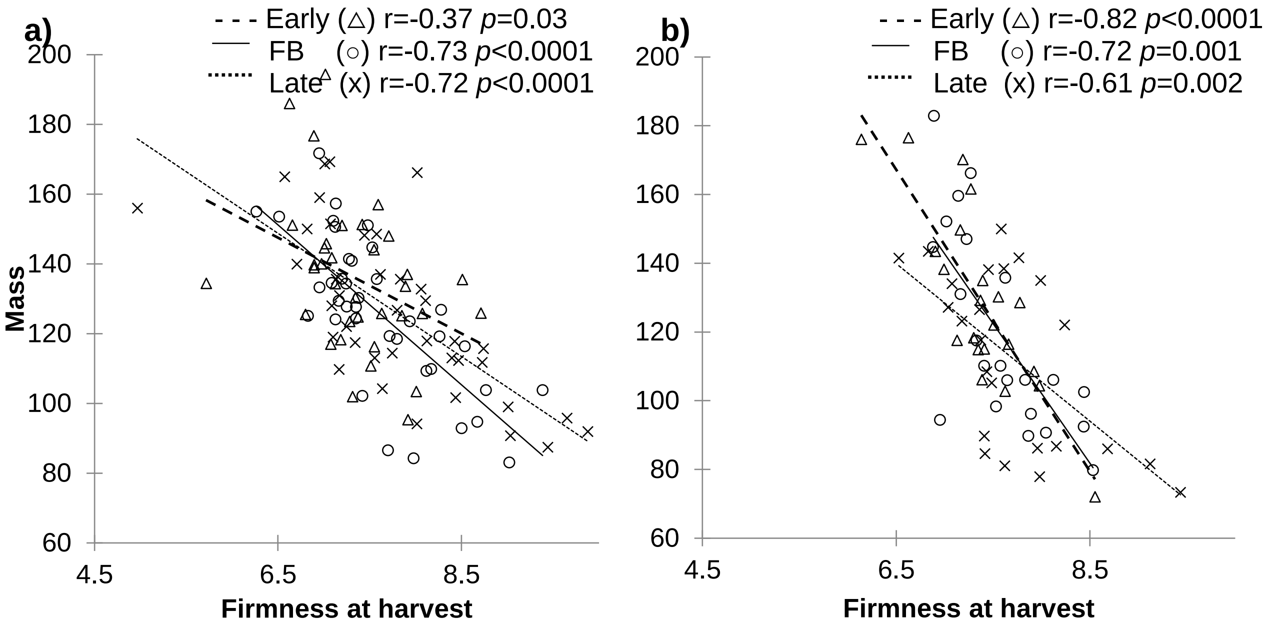

Figure 15: Scatter plot showing the correlations between **mass** and **firmness at harvest** for **Elgin** (a) and the **Koue Bokkeveld** (b) for each bloom class (Early, Full Bloom (FB) and Late).

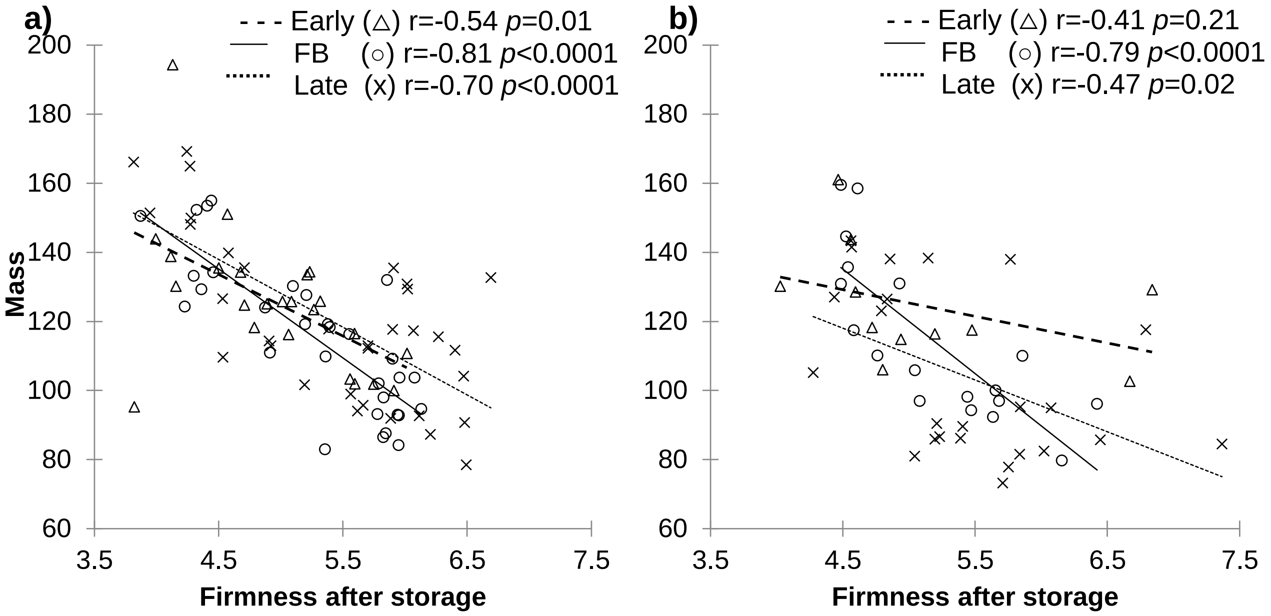

Figure 16: Scatter plot showing the correlations between **mass** and **firmness after storage** for **Elgin** (a) and the **Koue Bokkeveld** (b) for each bloom class (Early, Full Bloom (FB) and Late).

PAPER 3: The influence of bearing position and blossom date on apple flower quality.

Abstract

Originating in Central Asia, apple trees have developed complex physiological mechanisms to survive extended periods of sub-zero temperatures. These survival mechanisms become a stumbling block when cultivating apple orchards at lower latitudes where winter temperatures are too mild and the buds receive insufficient winter chill. The consequences of insufficient chill are typified by delayed, prolonged and insufficient bud break in spring, as well as basitonic branching. The aim of this study was to determine the influence of blossom time, canopy position and bearing position on flower quality. Flower quality was evaluated in two climatically distinct production sites in South Africa. Trial 1 investigated the effect of bloom time (early and late) and canopy position (branch height) on flower quality and Trial 2 investigated the effect of bearing position and spur age on flower quality (2-year-old spurs, 3-year-old spurs, old spurs, and shoots). Number of flowers per cluster, receptacle diameter, pedicel length and dry weight of flower clusters were measured as parameters of flower quality. Early flowers had a greater dry weight than late flowers on the colder site but no significant differences were found in the warmer site. Late blooming clusters generally exhibited poorer quality traits, such as lower dry mass and smaller receptacle diameters compared to early flowers in both sites. For bearing wood in the warmer site, 2-year-old spurs had the lowest dry flower mass, and while the effect of wood age in the colder site was less pronounced, old spurs produced the highest flower dry weight. The flower quality differences at bloom carried through to fruit mass at harvest where fruit from shoots were heaviest in the warmer site, while in the colder site fruit from spurs were heavier This showed that differences in flower quality throughout the canopy and across bloom time, gave rise to variation that persisted through to harvest. If the differences observed in this study were observed over seasons and across more sites, production practices may be modified to proactively remove old spurs in the milder winter region, while preserving it in the higher chill regions.

Keywords: fruit quality, variation, region, *Malus domestica* Borkh

1. Introduction

The apple is a temperate fruit crop originating from central Asia, specifically Kazakhstan (Forsline *et al*., 2003). To survive the characteristically cold winter where temperatures are below freezing for extended periods of time, temperate plants have developed mechanisms for protection (Horvath *et al*., 2003). The apple tree temporarily suspends all visible growth and enters a dormant state in early autumn that will continue until spring when growth continues as more favourable climatic conditions return.

Lang (1987) refined the definition of dormancy into three subclasses, namely: paradormancy, ecodormancy and endodormancy. Paradormancy refers to inhibitory signals from other structures within the plant, for example apical dominance exerted on the axillary buds. During ecodormancy, growth and development of a plant part is inhibited by environmental factors such as low temperature or a lack of nutrients and water. Endodormancy is the inhibition of growth by physiological signals or mechanisms within the bud meristem itself (Lang, 1987). The period of endodormancy is essential to the survival during the cold winters of the apple's origin. In their review, Horvath *et al*. (2003) expound on the mechanisms responsible for dormancy regulation in plants, and highlight, that in comparison to paradormancy and ecodormancy, there are gaps in understanding the physiological and molecular aspects of endodormancy.

A certain period of cold is required to break, or alleviate, the endodormant state of the buds; this is referred to as the chilling requirement (Fishman *et al*., 1987) and differs between apple cultivars. Cultivars are often referred to as low, medium or high chill-requiring based on the amount of chill they need. For example, cultivars with a medium chill requirement, such as 'Granny Smith' and 'Royal Gala' need <800 Utah chill units while higher chill cultivars, 'Fuji' and 'Golden Delicious', need more than 1000 Utah units to satisfy their chilling requirement (Tharaga *et al*, 2021). Cultivation of apple orchards are typically in the mid to high latitudes (35-60º) where the winter is sufficiently cold; however, with modified practices, production is possible at lower latitudes (Jackson, 2000). Most of South African apple production at 33-34ºS takes place under suboptimal chilling and the cold requirement for the buds is not fully met (Cook *et al*., 2017). When receiving insufficient cold to satisfy the chill requirement, the percentage bud break is reduced and there is an extended bloom period (Cook and Jacobs, 2000).

Insufficient winter chill also affects the quality of the flowers that emerge in spring (Mahmood *et al.,* 2000). Flower quality in turn, is important as it is determinant in fruit set and final fruit quality (Lauri *et al.,*1996). Mahmood *et al*. (2000) showed that greater winter chill produced larger sweet cherry flowers (method of flower size measurement was not defined) with longer pedicels. In apples, Petri and Leite (2003) reported that with higher chill exposure, pedicel length increased and fruit were heavier at harvest, but no flower size or mass measurements were taken. Feucht (1976) reported that very short or very long pedicel lengths in apple were indications of poorly developed flowers that are unlikely to set. Reginato *et al*. (2019) found that winter cold had a direct influence on ‘Gala’ apple fruit size when grown between 34 and 38ºS latitude, with lower average maximum temperatures during the winter being a stronger determinant of fruit size than season length.

The branching habit of apple trees are affected by insufficient chilling with the delayed foliation, basitonic branching and bare necks resulting in atypical canopy structure and basal dominance (Cook and Jacobs, 2000). Shoots and spurs may vary in their potential to produce high quality fruit (Hirst and Ferree, 1995) and the architectural modification associated with lack of winter chill, may influence the bearing potential of different plant parts. Maggs (1975) showed that an apple’s potential weight decreases by 2 grams for every day’s delay in flowering after full bloom and suggested that if later flowers could be stimulated to open earlier this would improve fruit size at harvest. However, this does not take into account initial flower weight or whether late opening flowers are already deficient in some way.

In this thesis, Paper 1 and 2 focused on final fruit quality and described the spatial and temporal effects of various influencing factors. It was seen in Paper 1 that spatial position within a canopy effects fruit maturity parameters with fruit from the upper canopy having higher flesh firmness and lower starch conversion. Paper 2 showed that relative blossom date had an influence on the starch conversion percentage in Elgin, with early clusters having the highest starch breakdown but there were no differences in the Koue Bokkeveld. To determine if these differences were already present at bloom time, the current paper aims to compare flower quality of early and late flowers on upper and lower canopy branches (Trial 1), as well as the influence of bearing position on flower quality (Trial 2).

2. Materials and methods

2.1. Plant material and sites

During the 2017/2018 season, ‘Golden Delicious’ flowers were collected in the climatically contrasting sites in the Koue Bokkeveld (33.4° S, 19.5° E, 945 m.a.s.l and annual avg. of ±1400 Utah chill units) and Elgin (34.2°S, 19.0°E, 305 m.a.s.l. and annual avg. ±700 Utah chill units), similar sites as described in Paper 1. It is known that bloom dates of both regions can differ, with the Koue Bokkeveld often blooming up to ten days earlier compared to orchards from Elgin. It has also been observed that the lower and upper portion of the tree can have asynchronous flowering as was the case for both regions in this study (Table 1).

One orchard from each area was selected based on the same criteria mentioned in Paper 1. The orchard in the Koue Bokkeveld was on Nooitgedacht (planted 1997), and the orchard in Elgin was on Disseldraai (planted 1999). Because only one site was included in each area, regional comparisons cannot be made and we rather refer to a “colder” or “warmer” site.. Two separate trials were conducted to study flower quality. Trial 1 examined the effect of tree position (top vs bottom) and time (early vs late) of flowering on flower quality and Trial 2 investigated the effect of bearing wood (shoots, 2-year-old spurs, 3-year-old spurs, and old spurs) on flower quality.

2.2. Flower cluster collection

To determine the effect of flowering time on flower quality (Trial 1), 15 trees were randomly selected in each orchard and to accommodate for asynchronous flowering an upper and lower scaffold branch were selected on the eastern side of each tree. “Early” flower clusters were collected randomly from each branch when 30% of the flower clusters had opened on a branch and “Late” flower clusters were sampled once 70% of the flower clusters had opened on a branch (a cluster was considered as “open” once the third flower within the cluster had opened). Top and bottom branches flowered asynchronously and were sampled separately. The dates of the cluster collections are shown in Table 1. On each date 30 flower clusters were collected per category.

To investigate the effect of bearing wood on flower quality (Trial 2), 12 trees were randomly selected in each orchard. Flower clusters were collected at “full bloom”, defined as 20% petal drop or when the greatest number of recently opened flowers were present on the tree. Four flower clusters were collected from each tree, corresponding to different bearing positions, viz. one-year-old shoots, spurs on two-year-old wood, spurs on three-year-old wood, and spurs on older wood (> three-year-old). In this paper two-year-old spurs, three-year-old spurs and old spurs will refer to the categories above. Sampling was done in the colder and warmer sites on the 18th and 26th of October, respectively.

In both trials, all flower clusters were collected from the adaxial side of the branch when all the flowers in the cluster were open. Each cluster was placed in a separate paper bag and kept at -0.5°C until measurements were taken.

2.3. Flower measurements

For both trials the leaves and flowers from each cluster were separated and the flowers in each cluster were tallied. The pedicel lengths and receptacle diameter of each flower was measured (Fig. 1) using an electronic calliper (Model EC799, Starret, USA). Pedicel length measurements were grouped as means per cluster, while for receptacle diameter, the king and lateral flowers were measure separately. The flower parts from each cluster (spur leaves removed) were placed in an oven and dried for 24 hours at 70°C or until mass remained unchanged. The dried flower mass was then determined using the electronic scale (Model C131AM, Hygeco, France). The king flower was weighed separately and the rest of the flowers in the cluster (lateral flowers) were weighed together to the nearest milligram.

2.4. Spur leaf measurements

Spur leaf area is a useful metric for determining flower quality. Procter and Palmer (1991) showed that early spur leaf removal resulted in poor fruit set and reduced calcium levels in apple fruit. Volz *et al*. (1994) also demonstrated that reduced spur leaf area resulted in the production of smaller apples. Spur leaves were collected in this study, but due to a sample mix-up in the laboratory, spur leaf area could not be assessed.

2.5. Statistical analysis

An analysis of variance (ANOVA) was performed at tree level (trees were considered as block reps) for the data set of both trials per production site using the GLM procedure in SAS, followed by Fisher's least significant difference (LSD) test when the significance level was <0.05. The overall variability within each of the quality parameters was determined using a box and whiskers plot to depict the mean, median and distribution of each variable where possible.

3. Results

3.1. Trial 1: Effect of bloom time and tree position on flower quality

A summary of the Trial 1 ANOVA results is presented in Table 2 for data from both the cold and warm sites. Only the significant results (in bold) will be discussed in detail.

3.1.1. Number of flowers

Neither the tree position x time of bloom interaction, nor the individual main effects (tree position and time of bloom) had a statistically significant effect on number of flowers per cluster in either site (Table 2). Box and whiskers plots could not be drawn for the number of flowers per cluster as a range of 4 to 6 integers would not produce a sensible box plot. The mean number of flowers in a cluster in the colder site was 4.65 and 5.28 in the warmer site.

3.1.2. Receptacle diameter

The receptacle diameter box plots for the warmer site showed late flowers from the bottom branch having the largest interquartile range (0.36 mm), while late flowers from the top branch had the smallest (0.21 mm; Fig. 2). Skewness was not consistent in the warmer site, with receptacle diameters negatively skewed for late flowers from the top branches, while other categories were positively skewed (Fig. 2). In the colder site, receptacle diameter was most variable in top early flowers which an interquartile range of 0.45 mm and least variable in late flowers from the bottom branch (0.11 mm; Fig. 2). Receptacle diameter of all categories in the colder site were negatively skewed but early flowers from the top branch were the most negatively skewed.

Both sites showed a significant tree position x blossom time interaction with a *p*-value of <0.0001 (Table 3). The mean receptacle diameters of flowers on the bottom branch in the warmer site, showed early flowers (2.27 mm) being smaller that late flowers (2.56 mm) (Table 6). The opposite was true on the top branches, where early flowers, with a mean receptacle diameter of 2.55 mm, were significantly larger than the mean 2.25 mm of the late flowers (Table 6). Receptacle diameters of early flowers from the bottom branch, and late flowers from the top branch did not differ from one another; likewise, bottom late and top early flowers had similar receptacle diameters (Table 6).

In the colder site, receptacle diameters of flowers on the bottom branch did not differ between early and late flower clusters, but on the top branch, the mean receptacle diameter of early flowers (2.69 mm) were significantly larger than the 1.45 mm of late flowers (Table 3). Comparing the bloom time within branch height, receptacle diameter of early flowers did not differ between top and bottom branches, but late flowers on the bottom branch had a greater receptacle diameter than top-late flowers (Table 3).

3.1.3. Pedicel length

The box plots for pedicel lengths showed that the top branches in the warmer site had a greater interquartile range for the late clusters (5.24 mm) than that of the early clusters (3.68 mm; Fig. 3). The opposite was observed on the bottom branches where the interquartile range of early clusters (4.36 mm) was greater than that of the late clusters (3.59 mm). Distributions were negatively skewed for early and late clusters on the bottom branch, as well for pedicel length of late clusters on the top branch, but was positively skewed for early clusters from the top branches (Fig. 3).

For the colder site the box plots showed a greater interquartile range for early (4.24 mm) and late (4.10 mm) clusters from the top branch than that of early (3.34 mm) and late (3.47 mm) clusters from the bottom branch (Fig. 3). Both early and late clusters showed pedicle length being skewed to the right, while late clusters from the bottom were normally distributed and early clusters from the bottom were positively skewed.

The interaction between the position and bloom time did not significantly influence pedicel length for either site (Table 2). The mean pedicel length of flower clusters in the warmer site showed significant differences for both the main effects, branch height and bloom time, with *p*-values of <0.0001 and 0.024, respectively. In the colder site only tree position showed significant differences (p<0.0001; Table 2).

In terms of tree position, mean pedicel length in the colder site was greater on the bottom branches (28.28 mm) than the top branches (22.51 mm), and the same was true for flowers clusters from bottom (31.82 mm) and top (25.31 mm) branches in the warmer site (Table 4). The time of bloom in the warmer site showed greater mean pedicel length for late flower clusters (29.47 mm) compared to early clusters (27.67 mm; Table. 5).

3.1.4. Dry weight of flower parts

The box plots of the dry weight of entire clusters showed in the warmer site the most variable category was early clusters from the bottom branch (interquartile range of 0.054 g) and the most uniform was early clusters from the top branch (interquartile range of 0.027 g; Fig. 4a). Weight of early clusters from the bottom branch was negatively skewed while all other categories in the warmer site were positively skewed (Fig. 4a).

In the colder site, early clusters from the bottom branches had the most variable mass with an interquartile range of 0.030 g and early flowers from the top branch were the most uniform having an interquartile range of 0.021 g (Fig. 4a). The data for all the categories in the colder site were negatively skewed.

In the warmer site, the dry weight of the king flowers had the largest interquartile range from early clusters on the bottom branch (0.007 g) with the smallest from early clusters on the top branch (Fig. 5). The dry weight of king flowers from early clusters on the top branch was negatively skewed while the other categories in the warmer site were positively skewed (Fig. 4b). The dry weight of king flowers from the early clusters on the bottom branch were again the most variable in the colder site with an interquartile range of 0.007 g (Fig. 4b). The narrowest interquartile range (0.003 g) was from late clusters on the bottom branch (Fig. 4b). King flower mass from early clusters on the bottom branch was negatively skewed the other categories in the colder site were positively skewed (0.005 g; Fig. 4b).

Fig. 4c shows the box plots of the dry weight of lateral flowers. Early clusters on the bottom branch in the warmer site had the largest interquartile range (0.047 g), while early clusters from the top branch had the smallest (0.026 g; Fig. 4c). Dry weight from early clusters on the bottom branch was negatively skewed but positively skewed for all other categories in the warmer site (Fig. 4c). In the colder site the most variable category was late clusters from the bottom branch (interquartile range of 0.024 g), while early clusters from the top branch were the most uniform (interquartile range of 0.017 g; Fig. 4c). Skewness was positive for early clusters on the top branch and negatively skewed for the other categories (Fig. 4c).

The ANOVA results showed that in the warmer site the effects of bloom time, tree position and their interaction were not significant for all dry weight measurement categories (dry weight all flowers, dry weight king flower, dry weight lateral flowers; Table 2).

For the dry weight of all flowers in a cluster, king flowers, and lateral flowers in the colder site, neither the bloom time x tree position interaction nor the position main effect was significant. Only the main effect of bloom time was significant for all three categories (all flowers p=0.009; king flowers p<0.0001; lateral flowers p=0.039; Table 2).

The differences between mean mass of the dry weight of all flowers in the colder site showed that early clusters (0.157 g) were heavier than late clusters (0.141 g; Table 5). The same was true for dry weight of king flowers with early clusters (0.036 g) being heavier than late clusters (0.031 g; Table 5). With the dry weight of all flowers less the king flowers we again seen early clusters (0.121 g) being heavier than late clusters (0.110 g; Table 5).

3.2. Trial 2: Effect of bearing position and spur age on flower quality

3.2.1. Number of flowers

For Trial 2 there was a significant difference in the mean numbers of flowers per cluster for bearing wood (*p*=0.009) in the colder site but there were no significant differences in the warmer site (Table 6). In the colder site, flower clusters borne on shoots (5.18) and old spurs (5.17) had a greater number of flowers per cluster compared to clusters on two-year-old (4.56) and three-year-old (4.58) spurs (Table 6).

3.2.2. Receptacle diameter

In the warmer site, receptacle diameters from 3-year-old spurs were the most variable (interquartile range of 0.34 mm) while those from shoots were the least (interquartile range of 0.14 mm; Fig. 5). Receptacle diameter data from old spurs were positively skewed while diameters from shoots, 2-year-old spurs and 3-year-old spurs were negatively skewed (Fig. 5).

The greatest interquartile range in the warmer site, was from 2-year-old spurs (0.22 mm) and the smallest from shoots (0.15 mm; Fig. 5). Receptacle diameters from shoots and 3-year-old spurs were positively skewed while those from 2-year-old spurs and old spurs were negatively skewed (Fig. 5).

The ANOVA results showed significant differences in the warmer site receptacle diameters for bearing wood (*p*<0.0001; Table 6), but not in the colder site. In the warmer site , flowers borne on shoots (2.63 mm) had the largest mean receptacle diameter, but it was not significantly larger than those of three-year-old spurs (2.56 mm; Table 6). Two-year-old spurs had receptacle diameters smaller (2.47mm) than shoots but did not differ from three-year-old spurs (Table 6). Old spurs produced flowers with the smallest receptacle diameters (2.10 mm; Table 6).

3.2.3. Pedicel length

In the warmer site, pedicel length from old spurs were the most variable (interquartile range of 6.56 mm), and that of 2-year-old spurs was the most uniform (interquartile range of 3.04 mm; Fig. 6). Data from old spurs and 2-year-old spurs were positively skewed and data from shoots and 3-year-old spurs were negatively skewed (Fig. 6).

Pedicel lengths of old spurs in the warmer site was the least uniform with an interquartile range of 6.60 mm compared to the 2-year-old spurs which was the most uniform (interquartile range of 3.07 mm; Fig. 6). Data from shoots and 2-year-old spurs were negatively skewed while data of 3-year-old spurs were positively skewed. Data from old spurs were normally distributed (Fig. 6).

Bearing wood did not show significant differences in the colder site but in warmer site, with a *p*-value of 0.013, differences of mean pedicel length were significant among the different bearing positions (Table 6). Mean pedicel length of clusters on spurs in the warmer site did not differ from one another, but those of shoots (30.96 mm) were longer compared to flowers on two-year-old (27.37 mm) and three-year-old spurs (26.46 mm; Table 6).

3.2.4. Dry weight of flower parts

The dry weight of flowers clusters (all flowers included) in the warmer site from 3-year-old spurs had the greatest interquartile range (0.45 g), while 2-year-old spurs had the smallest (0.014 g; Fig. 7a). Data from old spurs was normally distributed while that of shoots, 2-year-old spurs and 3-year-old spurs was positively skewed (Fig. 7a). The colder site had the most variation of dry mass on 2-year-old spurs (interquartile range of 0.050 g) and the least on 3-year-old spurs (interquartile range of 0.022 g; Fig. 7a). Data from shoots and 2-year-old spurs were positively skewed while data from old spurs were negatively skewed. Dry weight from 3-year-old spurs was normally distributed (Fig. 7a).

The ANOVA showed bearing wood had a significant effect on dry weight of all flowers in a cluster in both the warmer (p=0.001) and the colder (p=0.001; Table 6) sites. The dry mass of clusters from the warmer site were heaviest on shoots (0.184 g) and was significantly heavier than clusters from 2-year-old (0.143 g) and 3-year-old spurs but not different from clusters on old spurs (Table 6). In the colder site, dry weight of all flowers was greatest on old spurs (0.186 g) and the weight of 2-year-old spurs (0.156 g), 3-year-old spurs (0.162 g) and shoots (0.158 g) did not differ (Table 6).

The dry weight of king flowers in the warmer site had the greatest interquartile range on 3-year-old spurs and the least on 2-year-old spurs (Fig. 7b). Data from 2-year-old spurs were positively skewed while data from shoots, old spurs and 3-year-old spurs were negatively skewed (Fig. 7b). In the colder site king flower dry weight was most variable on 3-year-old spurs and least on 2-year-old spurs with an interquartile range of 0.008 g and 0.004 g, respectively (Fig. 7b). Data from shoots, old spurs and 2-year-old spurs were positively skewed while data from 3-year-old spurs were normally distributed (Fig. 7b).

Bearing wood did not have a significant effect on king flower dry weight in either of the sites (Table 6).

The variance of lateral flowers in a cluster is illustrated with box plots in Fig. 7c. For the warmer site the greatest interquartile range was seen on 3-year-old spurs (0.043 g) and the smallest on shoots (0.025 g; Fig. 7c). Data from old spurs were negatively skewed and on 3-year-old spurs it was positively skewed. Shoots and 2-year-old spurs had normally distributed data Fig. 7c). The most variation in the colder site is seen on 2-year-old spurs (interquartile range of 0.043 g) and the least on shoots (interquartile range of 0.025 g; Fig 7c). Data from shoots and 2-year-old spurs were normally distributed while data from 3-year-old spurs and old spurs were normally distributed (Fig. 7c).

For all lateral flowers in a cluster, the effect of bearing position was significant in both sites (warmer p=0.001; colder p=0.002; Table 6). Lateral flowers on shoots in the warmer site had the greatest dry mass (0.154 g; Table 6). Flowers from 3-year-old spurs (0.134 g) and old spurs (0.137 g) were lighter than those of shoot but did not significantly differ from one another (Table 6). In the colder site, the dry weight of lateral flowers was greatest on old spurs (0.150 g) and the weight of 2-year-old spurs (0.121g), 3-year-old spurs (0.126g) and shoots (0.127) did not differ (Table 6).

Discussion

Apple flowers clusters have three to seven flowers per cluster but typically for commercially important cultivars it ranges from four to six (Pratt, 1988) as was the case for ‘Golden Delicious’ in this study. The number of flowers per cluster was not influenced by bloom time or branch height within the canopy but an effect of bearing wood was observed in the colder site with 2 and 3-year-old spurs having the lowest number. For ‘Cox Orange Pippin’, Robbie and Atkinson (1994) showed that younger wood had fewer flowers per cluster and that these clusters had a lower chance of setting fruit, but the number of flowers in a cluster was not determinant of flower quality or final fruit quality (if they did indeed develop to maturity).

Little early work could be found that correlates receptacle diameter with final fruit size, and Palmer and Johnson (2019) showed only a weak correlation between flower receptacle diameter of ‘Gala’ apples and final fruit size. They later emphasize the use of dry matter content as a better metric to indicate fruit quality (Palmer and Johnson, 2019). Denne (1963) reported on apple blossom time and receptacle diameter, and showed that the earliest clusters to open had the greatest receptacle diameter. This (early cluster having a greater receptacle diameter than late clusters) was also the case for the top branches in both sites of this study. At harvest, Paper 2 also showed heavier fruit for early clusters compared to late. For the bottom branches in the warmer site, the late clusters had larger receptacle diameters than early clusters and in the colder site there was no difference. Denne (1963) explained that the best quality flower buds were the first to open. While the flowers that open early during bloom have larger receptacles, this is not due to their earlier opening but rather to their inherent better quality, which may relate to other factors such as their date of initiation, position, etc. In this study, the top and bottom branches flowered asynchronously and in the warmer site, bottom late flowers and top early flowers were ready to be sampled on the same day. This suggests that chronological date may have more relevance than dates relative to full bloom. If relative bloom date was disregarded, the top early clusters and bottom late clusters opened on the same day and resulted in no size differences between the two. The bottom early clusters and top late clusters in the warmer site, had the smallest receptacle diameters, suggesting that any extremities in bloom time is undesirable (too early or too late). The lack of this effect is difficult to explain for the colder site because the bloom period was of a similar length for both sites in 2018 (Paper 2).

The genetic makeup of a cultivar determines the bearing position that has the highest quality flower or fruit development potential (Parisi *et al*., 2014), and it was different across sites for ‘Golden Delicious’. One site being colder region suggests that climate may influence the bearing potential of different position in a tree. Not only the type of bearing position, but the age thereof may influence bearing potential. Robbie and Atkinson (1995), who examined inflorescence quality on various ages of ‘Cox Orange Pippin’, reported smaller clusters of lower weight on younger bearing wood but in their study, the trees used for the wood age trial were young (4-years-old), and they explained that since vegetative growth was high the previous season, growth cessation was delayed and inadequate time for reserve accumulation resulted in the poor quality on the 1-year-old wood. Also studying the effect of wood age on apple fruit quality, Volz *et al*. (1994) showed that on ‘Braeburn’ and ‘Granny Smith’, 2-year-old spurs produced the heaviest fruit, and fruit from 1-year-old laterals and spurs older than 3 years producing the smallest fruit. In this study bearing wood and spur age did not influence receptacle diameter in the colder site but in the warmer site it had a pronounced effect. In Paper 1 spurs and shoots were compared without considering spur age, and there it was seen than fruit from shoots were heavier than fruit from spurs in the warmer site, while in the colder site the spurs produced the heavier fruit. This is contrary to Ferree *et al*. (1997) where for ‘Royal Gala’ and ‘Jonagold’ flowering on three different sites, they reported no differences in flower quality even though differences in final fruit quality was observed. A possible explanation for our differences in quality could be that our colder site was exposed to an adequately cold winter and the comparison of a suboptimal winter site in this study produced a difference. The symptoms of insufficient winter chill include delayed foliation, basitonic branching and the formation of bare necks, i.e., two-year-old shoots without spurs or lateral shoots (Cook and Jacobs, 2000). This modification results in atypical canopy structure and may have an influence on phenotypic expression within the same cultivar. Evidence of this would be that in the cooler climate, no differences were seen between bearing positions, while old spurs in the warmer winter site showed signs of poor flower quality according to dry weight.

Using pedicel length as a qualified parameter to measure flower quality is challenging. Feucht (1976) reported that any extremes in length, large deviations in length from the mean, indicated poor flower quality and a reduced set potential. Besides the flowers with anomalous pedicel lengths, longer pedicels will be considered an indication of better quality for the purpose of this study. This is motivated by Buszard and Schwabe (1995) who found shorter pedicel length in a season following a large crop load. Large crop loads are associated with deleterious effects on return bloom and fruit set for the following season (Serra *et al.,* 2016). The association of crop load with shorter pedicels in a season following a large crop allows inference that shorter pedicels are an indication of deficient flower quality. Petri and Leite (2003) also reported that pedicel length increased with greater chill and associated greater pedicle length with fewer stigma and anther anomalies, and an increased number of pollen grains on anthers, all indicators of high flower quality.

Flowers from both sites showed longer pedicels in the lower branches, but in terms of blossom time, differences were only seen in the warmer site where late fruit had the longest pedicels. If longer pedicels are considered to indicate better quality flowers, this is contrary to the observations made for receptacle diameter. It would be more reasonable to assume, based on Feuchts (1976) that these pedicels (from bottom branch flower clusters in Elgin) were abnormally long. The increased length was unlikely due to increased chill as indicated by Petri and Leite (2003), but rather that the pedicels in the warmer site (which were longer than pedicels in the colder site) were an indication of poor quality of late opening flower clusters.

Ferree *et al*. (1997) showed dry weight of flower parts had a stronger influence on final harvest weight than receptacle diameter and pedicel length, and this could explain some of the inconsistencies between receptacle diameter and pedicel length results in this study. At the higher chilling site, the time of blossom influenced dry matter content of the flower clusters but this was not apparent in the warmer site. Paper 2 showed that the differences in dry weight of flowers carried through to harvest for the Koue Bokkeveld (colder) with late clusters producing the lightest fruit. The effect was also present at harvest in Elgin (warmer).

Bearing wood and spur age was an influencing factor in both sites, but not for king flowers. Palmer and Johnson (2019), reported that the greatest variation in 'Gala' flower dry weight came from between the different clusters and not from individual flowers within a cluster. In the warmer site, flower cluster dry weight was greatest on shoots, but the results for spurs were not consistent with that of receptacle diameter. Using receptacle diameter as the metric, old spurs had the worst quality, while for dry weight, 2-year-old spurs were the worst. It is important to note that quality on shoots was consistently better in the warmer site for both parameters. At the colder site the best flower quality was found on old spurs. This difference in flower quality translated directly to fruit mass at harvest as seen in Paper 1 where shoots produced the heaviest fruit in Elgin (warmer) and spurs (not split into age for Paper 1) produced the heaviest fruit in the Koue Bokkeveld (colder).

5. Conclusion

For pedicel length in the warmer site and dry weight in the colder site, late blooming clusters exhibited poorer quality traits than early flowers at both sites. Flower quality in relation to bloom time was similar between the sites for dry weight in the top branches but differences were seen for bearing wood with the best quality flowers in the warmer site being produced on shoots and the best in the colder site on old spurs. In terms of dry weight, early flowers were of better quality in the colder site but this was not observed in the warmer site. The influence of bearing wood was apparent for dry weight in the warmer site, with the highest mass found on shoots and 3-year-old spurs but in the colder site old spurs produced the heaviest flowers. In Paper 1, this difference was seen to translate to final fruit mass with the heaviest fruit in Elgin (warmer) being borne on shoots and heaviest fruit in the Koue Bokkeveld (colder) produced on spurs.

If the differences found in this study were to persist between seasons and in more sites, region specific studies on bearing sites yielding the greatest quality would benefit the industry. Pruning practise could further be refined for region specific goals such as judicious removal of old spurs in Elgin (warmer) orchards while preserving them in the Koue Bokkeveld (colder). If the effect is found to be directly dependent on winter chill, further adjustments to pruning practice could be made during a warmer winter.

Some anomalies in pedicel length were found when evaluating quality of early and late flower clusters and future work should collect samples more frequently to discern if a progressive change in flower quality is observed over chronological dates. Here it would be beneficial to log temperature and humidity data to account for weather irregularities during flowering. Furthermore, it would add value if the results from this study were to be confirmed using spur leaf number and area as a metric.

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Tables

Table 1: Trial 1. Collection dates (2017/2018 season) of flower cluster samples (30 per category) in relation to flower time, tree position and site.

Site	Branch position	Time	Date
Warmer site	Top	Early	26th Oct
		Late	28th Oct
	Bottom	Early	21st Oct
		Late	26th Oct
Colder site	Top	Early	16th Oct
		Late	18th Oct
	Bottom	Early	11th Oct
		Late	15th Oct

Table 2: ANOVA summary of **Trial 1** indicating the significance between means from the number of flowers (per cluster), pedicel length, receptacle diameter, dry weight of the king flower, dry weight of lateral flowers, and the dry weigh of all the flowers in cluster. F- and p-values are indicated for the main effects of branch position, blossom time and the branch x time interaction in the warmer and colder sites. Numbers in bold indicate significant *p*-values.

		Warmer site		Colder site	
Variable	Source	F-value	*p*-value	F-value	*p*-value
Number of flowers	Position	0.56	0.457	3.15	0.083
	Time	1	0.322	1.51	0.226
	Position x Time	1.57	0.218	2.25	0.141
Receptacle diameter (mm)	Position	0.07	0.793	87.87	**<0.0001**
	Time	0.01	0.920	159.97	**<0.0001**
	Position x Time	34.45	**<0.0001**	148.70	**<0.0001**
Pedicel length (mm)	Position	71.89	**<0.0001**	79.61	**<0.0001**
	Time	5.53	**0.024**	1.42	0.241
	Position x Time	0.09	0.764	0.01	0.921
Dry weight of all flowers (g)	Position	0.35	0.555	0.65	0.425
	Time	0.11	0.748	7.42	**0.009**
	Position x Time	0.97	0.329	0.25	0.622
Dry weight of king flower (g)	Position	0.40	0.529	1.86	0.179
	Time	0.54	0.466	26.85	**<0.0001**
	Position x Time	3.51	0.068	0.59	0.445
Dry Weight Lateral Flowers (g)	Position	0.31	0.582	0.43	0.514
	Time	0.23	0.638	4.53	**0.039**
	Position x Time	0.62	0.435	0.48	0.495

Table 3: Trial 1. Receptacle diameter means for the tree position x bloom time interaction for the warmer and the colder site as well as the LSD, F and *p*-values. Significance level of <5% in bold.

Variable		Receptacle Diameter(mm)	
Position		Warmer site	Colder site
Top	Early	2.55 a	2.69 a
	Late	2.25 b	1.45 c
Bottom	Early	2.27 b	2.55 ab
	Late	2.56 a	2.53 b
LSD		0.14	0.14
F-value		34.45	148.7
p-value		**<0.0001**	**<0.0001**

Table 4: Trial 1. Pedicel length means for bloom time in both sites as well as the LSD, F and *p*-values. Significance level of <5% in bold.

Variable	Pedicel Length(mm)	
Position	Warmer site	Colder site
Top	25.31 b	22.51 b
Bottom	31.82 a	28.28 a
LSD	1.55	1.31
F-value	79.61	71.89
p-value	**<0.0001**	**<0.0001**

Table 5: Trial 1. Pedicel length, dry weight entire cluster, dry weight king flower and dry weight lateral flowers for bloom time the two sites as well as the LSD, F and *p*-values. Significance level of <5% in bold. NS means no significant differences at 5% confidence interval.

Variable	Pedicel Length (mm)		Dry Weight Entire Cluster (g)		Dry Weight King Flower (g)		Dry Weight Lateral Flowers (g)	
Timing	Warmer	Colder	Warmer	Colder	Warmer	Colder	Warmer	Colder
Early	27.67 b	25.78 NS	0.185 NS	0.157 a	0.033 NS	0.036 a	0.153 NS	0.121 a
Late	29.47 a	25.01 NS	0.183 NS	0.141 b	0.033 NS	0.031 b	0.150 NS	0.110 b
LSD	1.55	1.31	0.014	0.012	0.002	0.002	0.013	0.011
F-value	5.53	1.42	0.11	7.42	0.54	26.85	0.23	4.53
p-value	**0.024**	0.241	0.748	**0.009**	0.466	**<0.0001**	0.638	**0.039**

Table 6: Trial 2. Number of flowers, receptacle diameter**,** pedicel length, dry weight entire cluster, dry weight king flower and dry weight lateral flowers for bearing position in the two sites as well as the LSD, F and *p*-values. Significance level of <5% in bold.

Variable	Number of Flowers		Receptacle Diameter (mm)		Pedicel Length (mm)		Dry Weight Entire cluster (g)		Dry Weight King Flower (g)		Dry Weight Lateral Flowers (g)	
Bearing wood	Warmer	Colder	Warmer	Colder	Warmer	Colder	Warmer	Colder	Warmer	Colder	Warmer	Colder
2ySpur	5.08 NS	4.56 b	2.47 b	2.49 NS	27.37 b	27.17 NS	0.143 c	0.156 b	0.028 NS	0.036 NS	0.115 c	0.121 b
3ySpur	5.33 NS	4.58 b	2.56 ab	2.48 NS	26.46 b	27.66 NS	0.162 b	0.162 b	0.028 NS	0.036 NS	0.134 b	0.126 b
Old Spur	5.17 NS	5.17 a	2.10 c	2.47 NS	28.37 ab	29.38 NS	0.168 ab	0.186 a	0.031 NS	0.037 NS	0.137 b	0.150 a
Shoot	5.67 NS	5.18 a	2.63 a	2.45 NS	30.96 a	26.15 NS	0.184 a	0.158 b	0.030 NS	0.032 NS	0.154 a	0.127 b
LSD	0.44	0.46	0.14	0.14	2.73	2.55	0.018	0.016	0.004	0.004	0.016	0.015
F-value	2.82	4.53	23.09	0.14	4.21	2.31	7.61	6.61	1.34	2.38	8.53	6.32
p-value	0.054	**0.009**	**<0.0001**	0.938	**0.013**	0.094	**0.001**	**0.001**	0.278	0.088	**<0.0001**	**0.002**

Figures

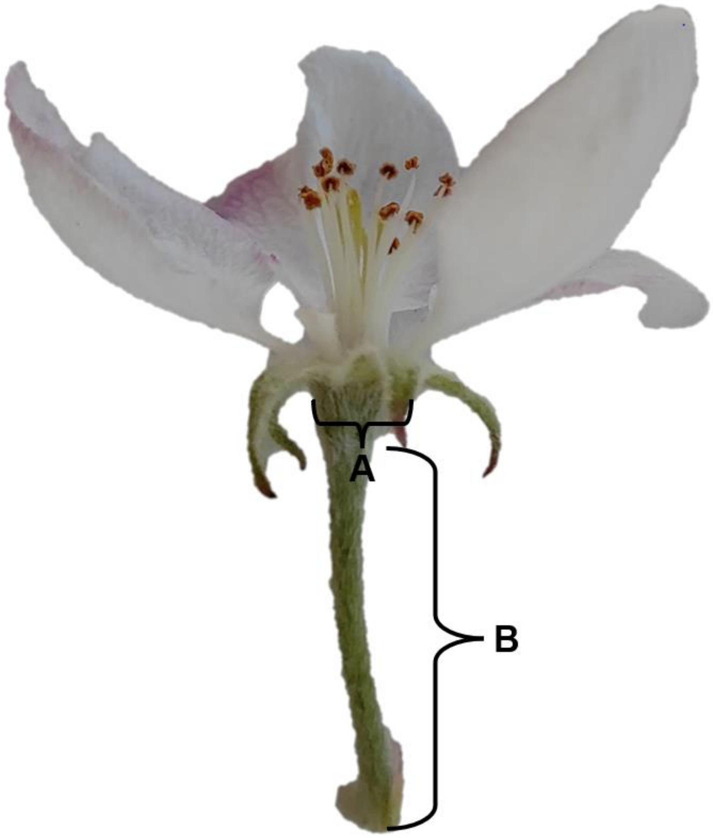

Figure 1: Diagram of the flower parts of a ‘Golden Delicious’ flower indicating the A**:** receptacle diameter and **B:** pedicel length that were used as indicators of flower quality.

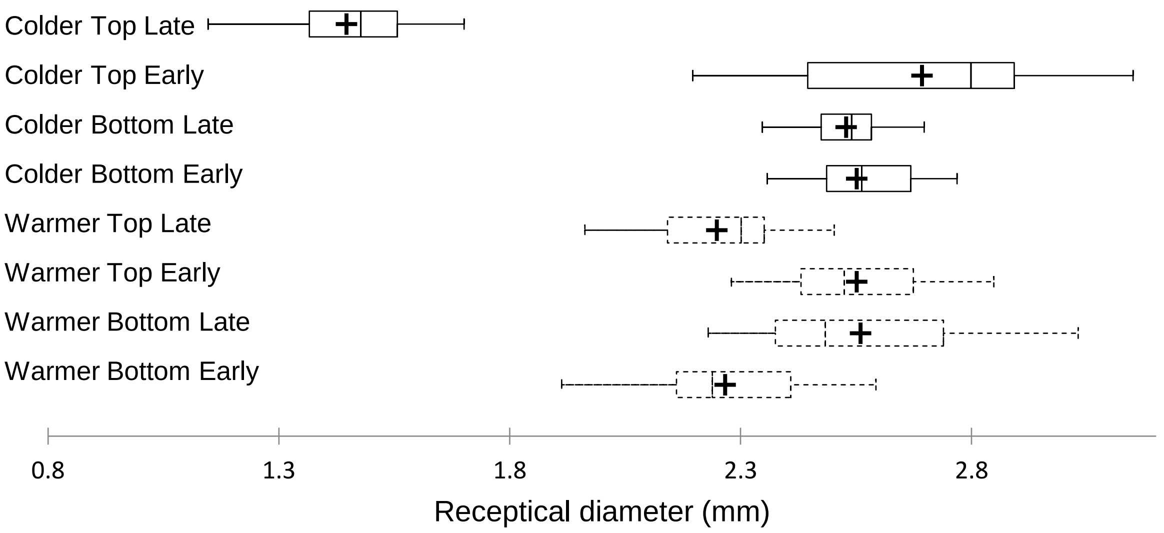

Figure 2: Trial 1. Box plots of receptacle diameter for time of bloom within branch position for both the colder (—) and warmer (E) (- -) sites. The box plot splits the data in four quartiles with an equal number of data points in each. The vertical line in the box is the median and the + indicates the mean.

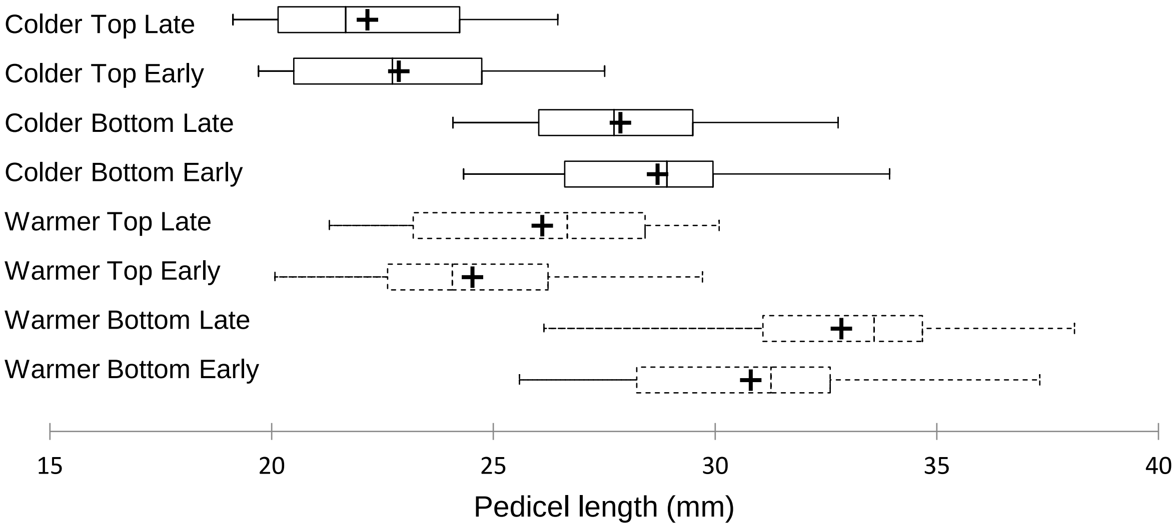

Figure 3: Trial 1. Box plots of pedicel length for time of bloom within branch height for both the colder (—) and the warmer (- -) sites. The box plot splits the data in four quartiles with an equal number of data points in each. The vertical line in the box is the median and the + indicates the mean.

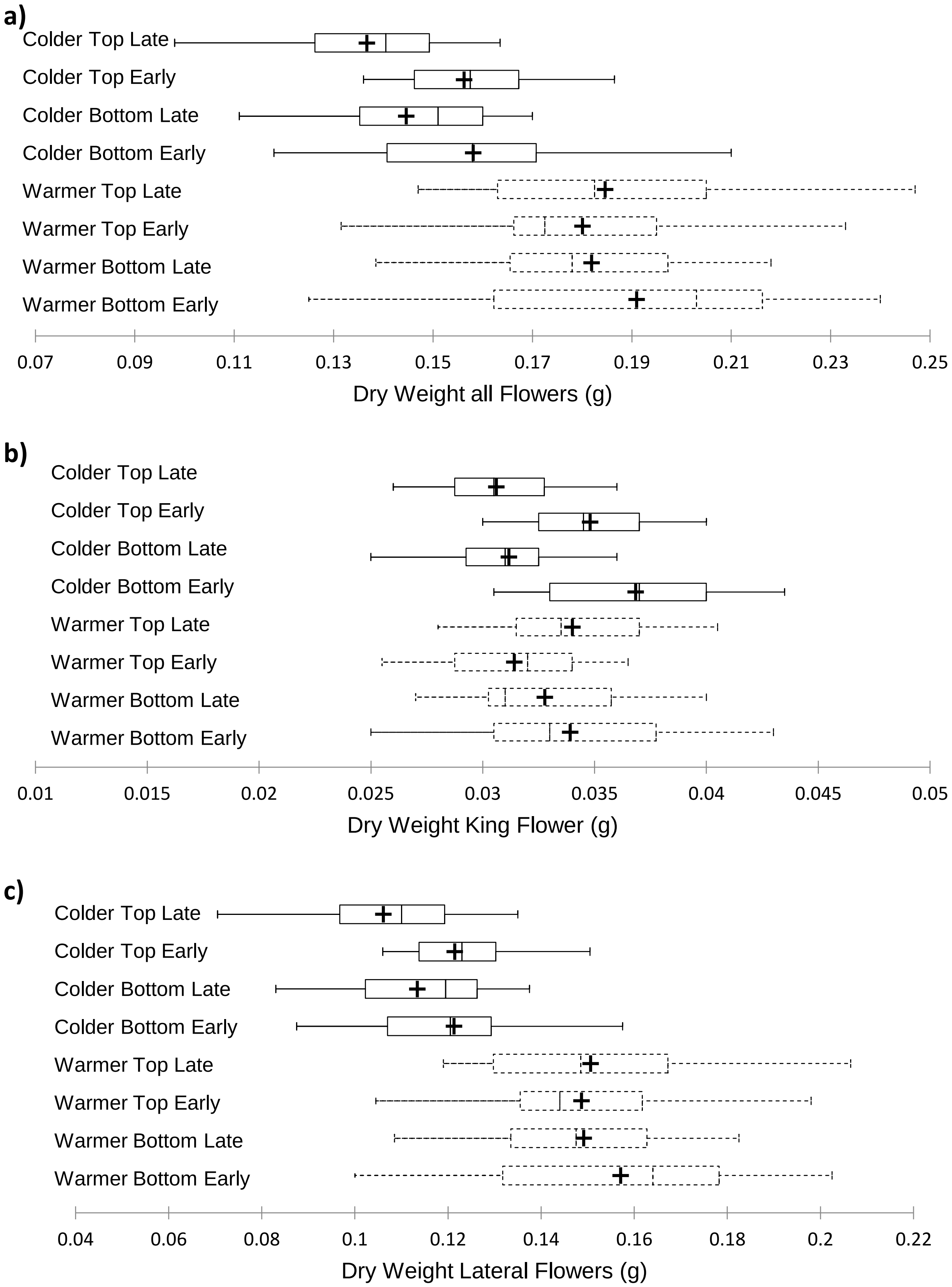

Figure 4: Trial 1. Box plots of the dry weight of (a) all flowers, (b) king flowers, and (c) lateral flowers for time of bloom within branch height for both the colder (—) and the warmer (- -) sites. The box plot splits the data in four quartiles with an equal number of data points in each. The vertical line in the box is the median and the + indicates the mean.

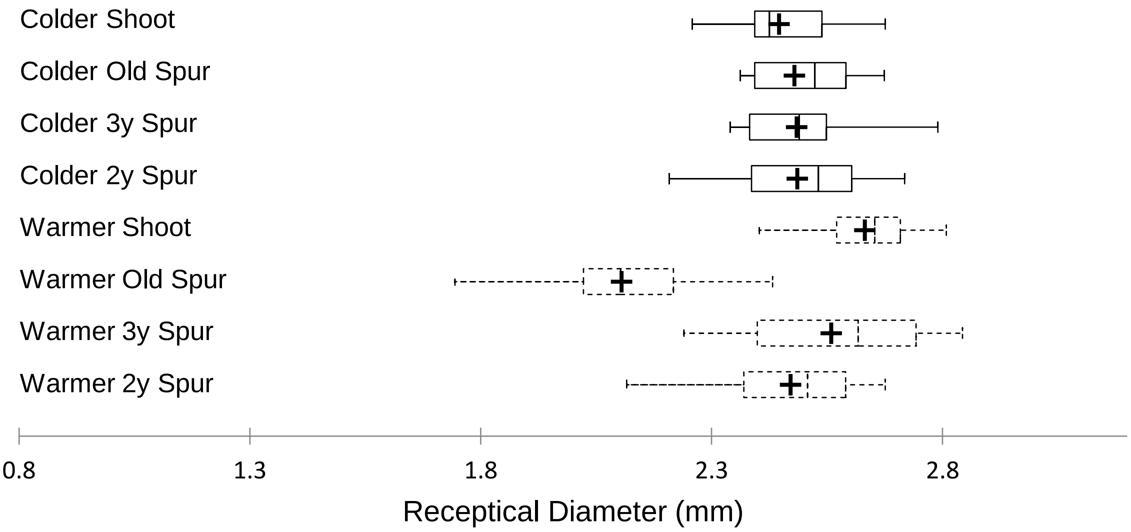

Figure 5: Trial 2. Box plots of receptacle diameter for bearing position in terms of spur age and shoot type for both the colder (—) and warmer (- -) sites. The box plot splits the data in four quartiles with an equal number of data points in each. The vertical line in the box is the median and the + indicates the mean.

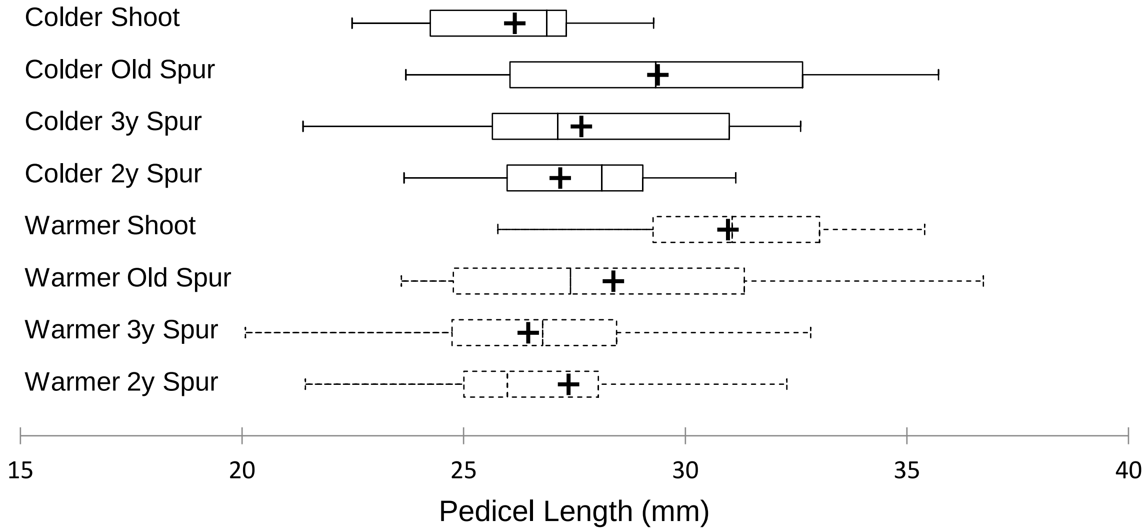

Figure 6: Trial 2. Box plots of pedicel length for bearing position in terms of spur age and shoot type for both the colder (—) and the warmer (- -). The box plot splits the data in four quartiles with an equal number of data points in each. The vertical line in the box is the median and the + indicates the mean.

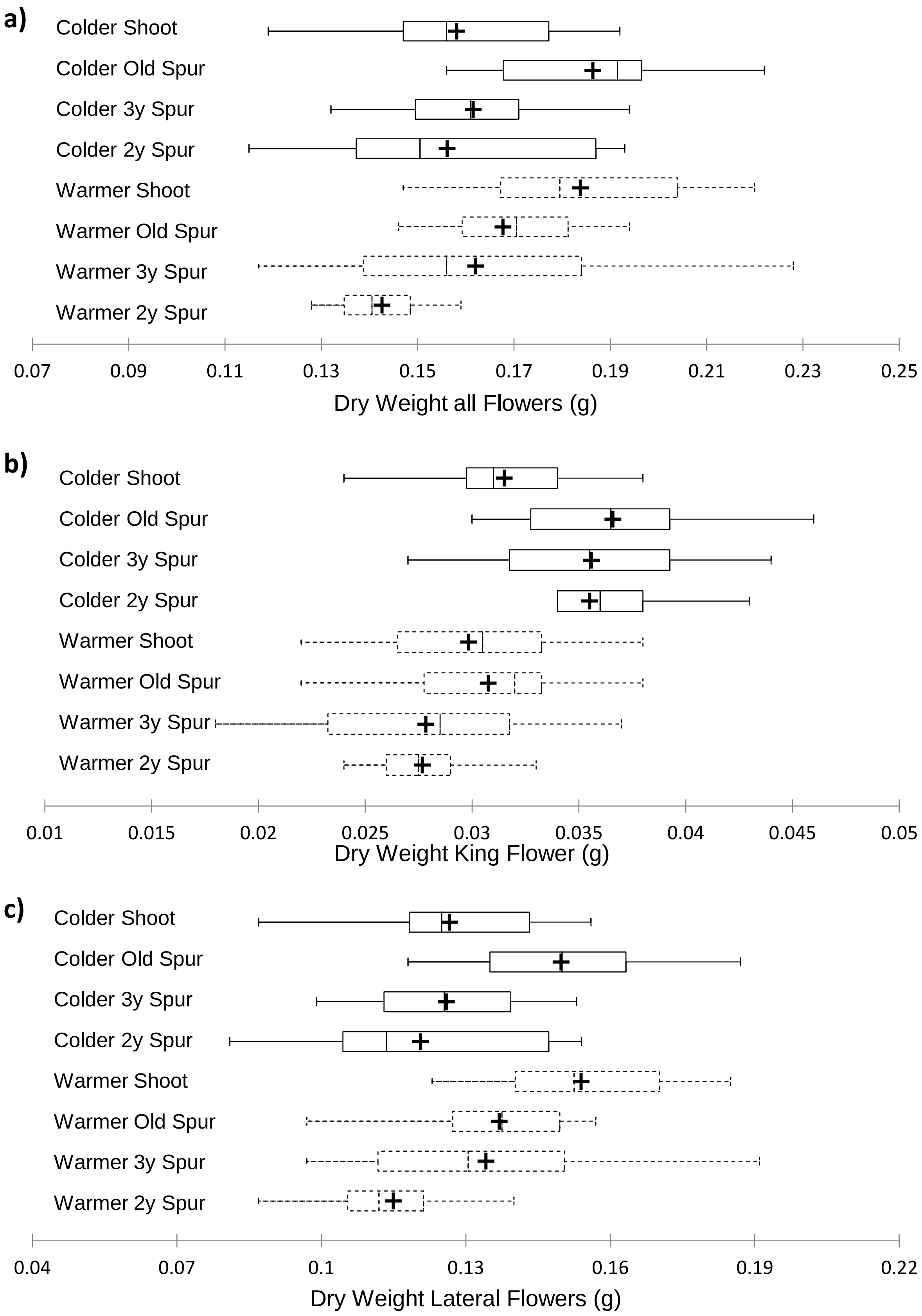

Figure 7: Trial 2. Box plots of the dry weight of (a) all flowers, (b) king flowers, and (c) lateral flowers for bearing position in terms of spur age and shoot type for both the colder (—) and the warmer (- -). The box plot splits the data in four quartiles with an equal number of data points in each. The vertical line in the box is the median and the + indicates the mean.

GENERAL DISCUSSION AND CONCLUSIONS

Using ‘Golden Delicious’ as a case study, this research aimed to identify the sources of variation and quantify their effect on specific external and internal fruit quality characteristics. To achieve this, ‘Golden Delicious’ apples were sampled in orchards from a mild winter region (Elgin) and a colder winter region (The Koue Bokkeveld) across multiple positions (vertical canopy position, sun exposure, and bearing wood) within a tree (Paper 1). Relative bloom date (RBD) was also investigated as a possible source of variation (Paper 2). Further examination of flower quality at bloom was conducted to determine how much variation was present at the start of the growing season (Paper 3).

Paper 1 showed that extensive variation in apple fruit quality and maturity are present at harvest. It was expected that the regional contribution to variation would be more apparent and that the insufficient winter chill, and subsequent erratic bloom in Elgin, would result in more variation in fruit quality and maturity at harvest than fruit from the Koue Bokkeveld. This was not the case, and instead orchard-to-orchard variation and tree-to-tree variation were the largest contributors. A high degree of tree-to-tree variation in fruit mass and yield was also reported by Aggelopoulou *et al*. (2010), who conducted a study mapping yield and quality across an orchard, but they found relatively little variation in quality. In contrast, Paper 1 and 2 showed a great deal of variation in quality among trees (except sunburn).

It was anticipated that including relative bloom date (RBD), as a source of variation, would explain more of the variation but again orchard and tree differences remained larger contributors. It is generally accepted that Western Cape soils are heterogenous over small areas and it is possible that soil conditions influenced the variation. In a study on terrain and management practice on soil variation, Umali *et al*. (2012) suggested more intensive soil mapping and detailed amelioration to mitigate the effect of “pocket zones” in apple orchards.

The orchards used in this study were standardized for rootstock, but errors during planting can directly introduce tree-to-tree variation. Li *et al*. (2015) showed that rootstock planting depth can have an influence on the rootstock’s effect on the scion. It is often the case in South African orchards, where little care has been taken to plant rootstocks at consistent depth, that the graft union is sometimes buried beneath the soil resulting in scion rooting and the vigour of such trees are greatly increased (personal observation).

Surprisingly, large unexplained variation was present in Paper 1 and Paper 2. It is important to note that the methodology of this work could influence the variability that remained unaccounted for. In orchards, commercial practise in hand thinning was followed, where labourers were tasked with removing the smallest fruit in each cluster. Also, the classification of sun exposed and shaded fruit was made at harvest, meaning fruit that were sun exposed at the start of the season but were shaded by shoot growth later would be classified as shaded. This effect of changing sun exposure was shown to effect colour development in 'Granny Smith' where fruit that transitioned from sun exposed to shaded during the season resulted in greener fruit compared to those shaded from the start of the season (Fouché *et al.,* 2010). Variation in our shaded category would then include fruit from both types and variation in green colour, and likely other qualities, would be higher. The same would apply for shaded fruit that became exposed to sunlight later during fruit development as branches bent under the weight of fruit.

Variation in maturity parameters increased from at harvest to after storage (Paper 1). Storing fruit of mixed maturities (immature, optimal and overmature fruits) within one cold store can lead to postharvest disorders. Manseka and Vasilakakis (1992) showed that 'Granny Smith' apples were more susceptible to superficial scald development when harvested immature. Over maturity can also cause issues such as internal browning in 'Braeburn' (Lau, 1998). The correlation between fruit colour and maturity parameters were moderate to weak, meaning that sorting 'Golden Delicious' fruit according to colour at harvest may to some extent create more uniform batches for storage. Colour parameters had strong correlations at harvest that were also present after storage. Greener 'Golden Delicious' fruit are more desirable and provide better income to south African producers (Adams 2021, personal communication, 22 November), and by colour sorting at harvest, it would be possible to prioritize marketing of batches expected to yellow quickly. Effort is made in industry to address the orchard-to-orchard variation, with fruit harvested from orchards that have yellower apples being grouped in storage separately from those with greener apples.

Region had a consistent influence on fruit aspect but not on fruit size. Although aspect ratio has a strong genetic component in its determination, Petri and Leite (2003) showed that warmer winters produced fruit with a smaller aspect ratio. This was confirmed in our study where both seasons showed taller fruit in the Koue Bokkeveld compared to those from Elgin. For fruit mass in 2017, a strong region effect was apparent and fruit from the Koue Bokkeveld were heavier. This was consistent with Reginato *et al*. (2019) who showed that winter climate has a direct influence on fruit mass with winters with a lower average daily maximum temperature producing larger fruit. In the 2018 season of this study though, no mass differences were observed between the areas. Using chill unit (CU) accumulation as a possible explanation, the Koue Bokkeveld was colder in both seasons but the difference in accumulated units was smaller in 2018 than in 2017 (2018: 234 CU difference; 2017: 516 CU difference). Winter chill almost certainly does have an effect on final fruit size, but in the second season where the difference in cold was smaller, other causal effects on fruit size may have been more pronounced such as pollination efficacy, seed number and growing season temperatures.

Light environment had a marked effect on appearance parameters. Sun exposure notably interacted with position within a tree (top and bottom). Wünsche and Lakso (2000) found that increased light interception led to increased yields and that individual parts of the tree that received more light produced heavier fruit. The distribution of light through the canopy is also important, and the review by Corelli-Grappadelli and Lakso (2007) explained that areas in the tree oversaturated with light overwhelmed the photosystems in leaves and carbon assimilation was reduced. They also described shaded sections of trees receiving inadequate light fixing too little carbon for adequate fruit growth and development. The upper branches in our study produced slightly larger fruit but considering that a solaxe training system (the case for all orchards in this study) can have 3.7 times the illumination in the upper canopy compared to the lower canopy (Buler and Mika, 2009), it is more likely that light exposure of upper branches was the driving factor to the difference.

Peel colour results also showed the influence of light exposure and fruit position within a tree, with yellower fruit found on the upper branches and at sun exposed sites. It is generally the case that ‘Golden Delicious’ becomes yellower during storage (Blažek *et al*., 2003). Our results confirm this and fruit that were greener at harvest were less yellow than those that entered storage with a yellower peel. It was interesting that fruit from shoots in our study was greener than fruit from spurs. Literature to explain this phenomenon could not be found but since higher nitrogen levels produce greener fruit (Hansen, 1980), it may be that fruit from these positions could have received more nitrogen from the tree. It may also be the case, as with the work by Fouché *et al*. (2010) discussed above, that fruit on shoots were initially exposed to the sun and as the fruit gained mass during the season, they weighed down the shoot and moved it into a shaded position. In this case the fruit would be protected from photobleaching in the late part of the season before harvest.

Light environment had a relatively smaller effect on the maturity parameters (starch conversion percentage and firmness). Ju *et al*. (1999) showed for ‘Fuji’ apples that light environment did not influence maturity at harvest. In our study though, we found that fruit from sun exposed positions had a lower starch conversion percentage (SC%) (were less mature) than fruit from shaded positions. The effect of light exposure on SC% was stronger in the upper branches but inconsistent in the lower branches where cases of no significant difference were observed. Since total light in the lower canopy of a solaxe trained orchard can be less than in the upper canopy (Buler and Mika, 2009), it may be that the difference of light received by sun exposed and shade fruit on the bottom branch is smaller than in the upper branch. This could explain why the effect was smaller and sometimes absent in the lower branches. The upper and lower branches showed stark differences in SC%, with the upper branches having a lower SC% than the bottom branches. The observed interaction between light exposure and position within a tree had a marked effect on maturity, but time of bloom within these positions likely contributed to the differences seen. The date of bloom, recorded in Paper 3, showed flower clusters from bottom branches opening earlier than those in top branches. The effects of bloom date are discussed further below.

Although using SC% as a measure of maturity is commonplace, Blanpied and Silsby (1992) emphasised some drawbacks such as light crop loads falsely showing a low SC%. Duly, it is sensible to evaluate the SC% results in conjunction with fruit flesh firmness. Literature has shown that sun exposed fruit has a higher firmness than shaded fruit on a tree (Blanpied *et al*., 1978; Klein *et al*., 2001). The results from our study confirm that the sun exposed fruit were firmer and the difference was larger after storage than at harvest. Klein *et al*. (2001) also found a marked increase in firmness after storage, but that the difference then dissipated after a 10-day shelf-life period. In this study a 7-day shelf-life period was used and we could not determine if this was the case in our trials. The contribution of RBD to overall variation was smaller than expected but had some effect on the quality parameters measured, most notably on fruit mass, SC% and firmness. Richardson *et al*. (2019) showed that for kiwi fruit, bloom date strongly influences fruit mass and fruit that developed from the early flowers were larger than from late flowers. In our study RBD presented similar results to their work, with early flowers producing the heaviest fruit in both regions, but with relatively small differences compared to in kiwi (Richardson *et al*., 2019). Studying the effect of prolonged flowering in apples, Maggs (1975) reported that for every day that bloom was delayed, final fruit mass was 2 grams lighter. Even though the differences observed in this study were small, they would equate to significant economic gain to producers if the bud break was more condensed.

RBD had a greater contribution to variation in maturity (SC% and firmness) than it did on the other parameters measured. Early blooming flowers would begin development first, and it would be expected that if fruit is strip picked at harvest, as is typically the case for ‘Golden Delicious’, these would be the ripest fruit on the tree. The SC% results in Elgin followed the above assumption, with each successive bloom class having less mature fruit in it (lower SC%). In the Koue Bokkeveld however, SC% did not differ between bloom classes. For firmness both regions showed a progressive decline in firmness for each successive bloom class, but the effect was smaller in the Koue Bokkeveld. It may have been that on-tree development (or season length) had a greater effect in Elgin than the Koue Bokkeveld. Growing season temperatures also need to be considered as the root of why the response to RBD differed between the areas, but this was unfortunately not recorded during our study.

Season length differences between the bloom classes alone does not suffice as an explanation for the effects observed. Denne (1963) reported that the earliest flowers to open on apple trees were generally of better quality. Paper 3 was aimed at determining if the differences in fruit quality were already present at bloom time by considering the influence of spatial and temporal factors from the previous papers on flower quality.

Recent work by Palmer and Johnson (2019) showed a correlation, albeit weak, between flower receptacle diameter and final fruit size of ‘Gala’ apples. Our study showed that the early clusters from the top branches had the greatest receptacle diameters; this carried through to harvest with early clusters producing heavier fruit than late clusters. Our finding on receptacle diameter also concurs with Denne (1963) who also found larger receptacle diameter on early clusters. The bottom branches in this study did not react similarly with late clusters in the warmer site having larger receptacle diameters than those from early clusters. It may have been that lower amounts of light in the bottom of the canopy produced weaker flower buds, overriding the temporal effects (Barritt *et al*., 1991). It should also be noted that the top and bottom of the tree flowered asynchronously, and in the warmer site late flowers on the bottom branch were sampled on the same day as early flowers from the top branch. This points to chronological date possibly being more relevant than date relative to full bloom.

Bearing wood type (shoots and spurs) has an influence on the potential of a flower bud to yield good quality fruit, and being genetically determined, varies between cultivars (Parisi *et al*., 2014). The spur age may also influence the final fruit quality (Robbie and Atkinson, 1995; Volz *et al.,* 1994). Flower cluster dry weight in the warmer site was greatest on shoots while in the colder site the greatest dry weight was on old spurs. This difference was present at harvest where the heaviest fruit in Elgin (warmer) were found on shoots, while the old spurs in the Koue Bokkeveld (colder) produced the heaviest fruit. Insufficient winter chill modifies branching habit, causing delayed foliation, basitonic branching and “bare necks” (Cook and Jacobs, 2000). This phenomenon shows that phenotypic expression differed between the areas, and a difference in winter chill accumulation was likely a strong determinant.

Variation is present from bloom through to harvest and there are a number of practises that would be able to address this. For established orchards, the use of either aminoethoxyvinylglycine (AVG; Venburg *et al*., 2008) or 1-methylcyclopropene (1-MCP; Varanasi *et al*., 2013) would reduce the variation in maturity at harvest. Judicious pruning and the removal of branches which grow too vigorously would improve the light distribution through the canopy and reduce some of the variation due to light environment described in this paper. These practices are applied in the majority of currently producing orchards and were applied to the orchards in this study, with the exception of 1-MCP and AVG which were not sprayed on the trial orchards. Current industry research is being done on ‘Big Bucks’, a full red ‘Gala’ mutation, where cytokinin (6-BA and CPPU) applications are being trialled in conjunction with conventional rest breaking applications and shortly after green tip to stimulate a more uniform blossom. The stark differences in maturity between top and bottom branches of the tree, warrants selective picking for ‘Golden Delicious’ which is typically strip picked in South Africa. Having been informed of these results, some growers in the 2021 season picked their top and bottom portions of the tree separately (personal observation). Fruit from the upper canopy was at optimal picking maturity, while fruit from the lower canopy was overmature even though both portions of the tree were picked on the same day (personal observation). This allowed for separation between the two batches of fruit in storage.

For the establishment of new orchards, growers should aim at “light friendly” plantations, i.e., orchards with good light interception and distribution. This can be achieved with higher planting density, which allows for slender spindle tree architecture or a more formal two-dimensional “fruiting wall”. With these plantings, the difference in wood age on a tree would typically be less, eliminating another portion of the variation. To reduce variation between trees, these new orchards should also have a constant planting depth and avoid burying the trees above the graft union. Additionally, the use of shade netting in combination with reflective mulch could create a far more uniform light environment. Aggelopooulou *et al*. (2013) suggested using a combination of soil characteristics, yield and fruit quality to create “management zones” within orchards to enable production practices to be adapted for each zone type. Their methods in defining the zones unfortunately required much time and specialize skill. Sun *et al*. (2019) showed that drone imaging could accurately map apple orchards, and it should be possible for the management zones defined above, to be generated by bulk data from remote sensing instead of intensive measurements.

'Golden Delicious' was used as a case study, but additional work on other cultivars would be useful to determine if these results hold true. Our work revealed large variation in fruit quality and maturity at harvest. If this variation were to be addressed and consequentially reduced, it could lead to direct economic benefit to South African apple producers. Future work on apple fruit quality in regions with insufficient winter chill would benefit by accounting for both the dormant season temperatures as well as the growing season temperatures. Repeating this type of work on high density planting would also be beneficial to the field.

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APPENDIX A

Table 1: ANOVA summary of **fruit aspect** ratio, showing F and *p*-values for the 2017 and 2018 seasons.

Source	2017		2018	
	F-value	*p*-value	F-value	*p*-value
Region	1567.09	**<0.0001**	166.05	**<0.0001**
Orchard	38.20	**<0.0001**	15.83	**<0.0001**
Tree	4.11	**<0.0001**	3.19	**<0.0001**
TP	0.10	0.750	2.12	0.147
LE	0.23	0.634	2.76	0.099
TP x LE	0.01	0.907	1.37	0.244
BP	20.02	**<0.0001**	2.07	0.152
TP x BP	0.16	0.685	1.25	0.265
LE x BP	8.83	**0.003**	1.05	0.307
TP x LE x BP	1.51	0.220	0.45	0.505
R x TP	0.50	0.478	1.10	0.296
R x LE	7.21	**0.007**	1.24	0.266
R x TP x LE	3.98	**0.047**	0.31	0.578
R x BP	0.19	0.661	0.12	0.732
R x TP x BP	4.78	**0.029**	1.18	0.279
R x LE x BP	1.88	0.171	0.36	0.547
R x TP x LE x BP	2.01	0.156	1.58	0.211

Table 2: ANOVA summary of mass showing F and *p*-values for the 2017 and 2018 seasons.

	2017		2018	
Source	F-value	*p*-value	F-value	*p*-value
Region	892.03	**<0.0001**	36.34	**<0.0001**
Orchard	49.16	**<0.0001**	50.72	**<0.0001**
Tree	8.36	**<0.0001**	5.57	**<0.0001**
TP	7.35	**0.007**	9.30	**0.003**
LE	21.52	**<0.0001**	4.04	**0.046**
TP x LE	13.55	**0.001**	0.16	0.691
BP	15.29	**0.001**	0.21	0.651
TP x BP	3.87	**0.049**	1.27	0.261
LE x BP	0.29	0.589	0.74	0.389
TP x LE x BP	0.00	0.967	0.32	0.570
R x TP	29.44	**<0.0001**	2.00	0.160
R x LE	6.14	**0.013**	0.48	0.488
R x TP x LE	6.54	**0.011**	0.49	0.486
R x BP	0.88	0.349	13.09	**0.001**
R x TP x BP	1.92	0.166	2.82	0.095
R x LE x BP	1.11	0.292	2.21	0.139
R x TP x LE x BP	0.03	0.867	0.01	0.940

Table 3: ANOVA summary of hue angle at harvest showing F and *p*-values for the 2017 and 2018 seasons.

	2017		2018	
Source	F-value	*p*-value	F-value	*p*-value
Region	99.75	**<0.0001**	6.59	**0.011**
Orchard	38.25	**<0.0001**	25.65	**<0.0001**
Tree	5.41	**<0.0001**	3.47	**<0.0001**
TP	460.56	**<0.0001**	96.70	**<0.0001**
LE	521.91	**<0.0001**	64.07	**<0.0001**
TP x LE	1.31	0.254	1.70	0.194
BP	0.18	0.668	0.90	0.345
TP x BP	6.25	**0.013**	0.00	0.997
LE x BP	0.70	0.404	3.73	0.055
TP x LE x BP	0.01	0.903	0.17	0.678
R x TP	17.99	**<0.0001**	1.23	0.269
R x LE	0.71	0.400	0.03	0.860
R x TP x LE	1.32	0.251	0.08	0.783
R x BP	6.06	**0.014**	1.08	0.300
R x TP x BP	0.01	0.926	0.97	0.327
R x LE x BP	3.04	0.082	0.09	0.762
R x TP x LE x BP	1.44	0.230	0.12	0.731

Table 4: ANOVA summary of hue angle after storage showing F and *p*-values for the 2017 and 2018 seasons.

	2017		2018	
Source	F-value	*p*-value	F-value	*p*-value
Region	2.11	0.147	0.30	0.587
Orchard	42.36	**<0.0001**	32.18	**<0.0001**
Tree	5.87	**<0.0001**	6.11	**<0.0001**
TP	222.32	**<0.0001**	58.80	**<0.0001**
LE	136.03	**<0.0001**	35.64	**<0.0001**
TP x LE	0.04	0.850	0.54	0.465
BP	7.68	**0.006**	0.74	0.395
TP x BP	7.28	**0.007**	0.35	0.555
LE x BP	6.09	**0.014**	0.48	0.493
TP x LE x BP	0.24	0.628	0.00	0.994
R x TP	37.38	**<0.0001**	1.77	0.190
R x LE	0.57	0.452	3.45	0.069
R x TP x LE	0.08	0.777	0.65	0.425
R x BP	3.56	0.060	0.27	0.605
R x TP x BP	0.31	0.577	2.04	0.160
R x LE x BP	1.85	0.175	0.01	0.924
R x TP x LE x BP	0.77	0.380	0.01	0.936

Table 5: ANOVA summary of lightness at harvest showing F and *p*-values for the 2017 and 2018 seasons.

	2017		2018	
Source	F-value	*p*-value	F-value	*p*-value
Region	3.66	0.056	48.31	**<0.0001**
Orchard	100.61	**<0.0001**	30.80	**<0.0001**
Tree	8.92	**<0.0001**	5.01	**<0.0001**
TP	172.67	**<0.0001**	55.72	**<0.0001**
LE	169.81	**<0.0001**	56.30	**<0.0001**
TP x LE	3.03	0.082	1.77	0.185
BP	45.37	**<0.0001**	4.60	0.033
TP x BP	0.58	0.446	0.22	0.638
LE x BP	0.55	0.458	0.81	0.368
TP x LE x BP	0.12	0.734	1.31	0.254
R x TP	79.60	**<0.0001**	1.18	0.278
R x LE	6.06	**0.014**	0.46	0.498
R x TP x LE	0.28	0.597	0.02	0.891
R x BP	7.55	**0.006**	1.38	0.242
R x TP x BP	0.45	0.502	2.30	0.132
R x LE x BP	0.28	0.599	1.10	0.296
R x TP x LE x BP	1.19	0.276	0.06	0.805

Table 6: ANOVA summary of lightness after storage showing F and *p*-values for the 2017 and 2018 seasons.

	2017		2018	
Source	F-value	*p*-value	F-value	*p*-value
Region	0.78	0.376	1.33	0.254
Orchard	72.53	**<0.0001**	19.23	**<0.0001**
Tree	8.39	**<0.0001**	4.39	**0.001**
TP	23.67	**<0.0001**	6.92	**0.011**
LE	21.33	**<0.0001**	10.24	**0.002**
TP x LE	0.43	0.514	1.29	0.262
BP	36.10	**<0.0001**	2.07	0.156
TP x BP	0.34	0.559	0.20	0.659
LE x BP	1.83	0.177	0.01	0.905
TP x LE x BP	0.18	0.673	0.06	0.812
R x TP	46.90	**<0.0001**	0.00	0.956
R x LE	0.86	0.355	0.66	0.419
R x TP x LE	0.06	0.811	1.38	0.247
R x BP	6.37	**0.012**	0.00	0.955
R x TP x BP	2.51	0.115	0.88	0.353
R x LE x BP	1.61	0.206	0.29	0.592
R x TP x LE x BP	0.09	0.763	0.15	0.698

Table 7: ANOVA summary of sunburn showing F and *p*-values for the 2017 and 2018 seasons.

	2017		2018	
Source	F-value	*p*-value	F-value	*p*-value
Region	12.64	**0.001**	54.16	**<0.0001**
Orchard	7.40	**<0.0001**	5.32	**<0.0001**
Tree	1.50	**0.001**	1.04	0.426
TP	234.66	**<0.0001**	64.62	**<0.0001**
LE	1595.28	**<0.0001**	386.24	**<0.0001**
TP x LE	47.59	**<0.0001**	1.89	0.171
BP	3.90	**0.049**	4.94	**0.028**
TP x BP	3.32	0.069	3.35	0.069
LE x BP	1.34	0.247	0.99	0.320
TP x LE x BP	0.69	0.406	0.03	0.874
R x TP	9.74	**0.002**	7.33	**0.008**
R x LE	0.27	0.603	7.83	**0.006**
R x TP x LE	8.68	**0.003**	0.26	0.610
R x BP	1.58	0.209	0.57	0.451
R x TP x BP	0.02	0.877	4.56	**0.034**
R x LE x BP	1.59	0.207	4.17	**0.043**
R x TP x LE x BP	0.01	0.928	4.58	**0.034**

Table 8: ANOVA summary of starch conversion percentage showing F and *p*-values for the 2017 and 2018 seasons.

	2017		2018	
Source	F-value	*p*-value	F-value	*p*-value
Region	2.73	0.099	13.59	**0.001**
Orchard	66.24	**<0.0001**	21.71	**<0.0001**
Tree	17.22	**<0.0001**	6.75	**<0.0001**
TP	222.68	**<0.0001**	64.23	**<0.0001**
LE	4.96	**0.027**	1.68	0.201
TP x LE	3.68	0.056	0.17	0.685
BP	10.73	**0.001**	0.11	0.737
TP x BP	0.07	0.790	0.72	0.399
LE x BP	2.00	0.158	1.45	0.234
TP x LE x BP	0.25	0.616	0.00	0.963
R x TP	44.27	**<0.0001**	20.09	**<0.0001**
R x LE	3.71	0.055	17.36	**0.001**
R x TP x LE	4.27	**0.040**	18.01	**<0.0001**
R x BP	0.00	0.991	0.01	0.918
R x TP x BP	0.12	0.731	0.70	0.406
R x LE x BP	0.25	0.619	0.04	0.837
R x TP x LE x BP	0.66	0.418	6.73	**0.012**

Table 9: ANOVA summary of firmness at harvest showing F and *p*-values for the 2017 and 2018 seasons.

	2017		2018	
Source	F-value	*p*-value	F-value	*p*-value
Region	8.29	**0.004**	0.51	0.479
Orchard	9.55	**<0.0001**	24.25	**<0.0001**
Tree	4.17	**<0.0001**	4.38	**<0.0001**
TP	15.80	**<0.0001**	9.09	**0.004**
LE	1.62	0.205	2.36	0.130
TP x LE	0.25	0.615	2.72	0.105
BP	19.48	**<0.0001**	11.45	**0.001**
TP x BP	0.58	0.445	0.34	0.563
LE x BP	1.14	0.286	0.99	0.324
TP x LE x BP	1.33	0.250	1.58	0.215
R x TP	9.36	**0.002**	3.78	0.057
R x LE	0.09	0.760	1.86	0.178
R x TP x LE	0.40	0.525	3.21	0.079
R x BP	2.25	0.135	4.99	**0.030**
R x TP x BP	0.27	0.607	1.65	0.205
R x LE x BP	0.71	0.400	0.00	0.969
R x TP x LE x BP	0.48	0.488	1.29	0.260

Table 10: ANOVA summary of firmness after storage showing F and *p*-values for the 2017 and 2018 seasons.

	2017		2018	
Source	F-value	*p*-value	F-value	*p*-value
Region	3.96	0.048	6.79	**0.012**
Orchard	23.66	**<0.0001**	47.73	**<0.0001**
Tree	7.33	**<0.0001**	3.33	**0.002**
TP	59.94	**<0.0001**	18.2	**<0.0001**
LE	100.95	**<0.0001**	25.2	**<0.0001**
TP x LE	4.31	**0.039**	5.59	**0.022**
BP	16.20	**<0.0001**	15.15	**0.001**
TP x BP	1.79	0.182	0.01	0.929
LE x BP	0.02	0.897	0.64	0.426
TP x LE x BP	0.10	0.752	0.56	0.456
R x TP	0.11	0.741	7.58	**0.008**
R x LE	0.06	0.805	2.96	0.092
R x TP x LE	0.68	0.411	0.47	0.495
R x BP	0.01	0.931	1.75	0.192
R x TP x BP	0.05	0.831	0.29	0.591
R x LE x BP	0.25	0.615	0.13	0.725
R x TP x LE x BP	0.20	0.657	0.16	0.689

APPENDIX B

Table 1: Summarised ANOVA results of Elgin and Koue Bokkeveld, showing the LSD, F and *p*-values of Lightness at harvest, Lightness after storage, Hue at harvest and Hue after storage with flower class as a factor. '*' indicates a non-significant *p*-value at a 5% confidence level.

Parameter	Region	Early flowers	Full Bloom	Late flowers	LSD	F-value	*p*-value
Lightness at Harvest (value)	Elgin	72.50 ab	72.37 b	72.82 a	0.33	3.56	0.029
	Koue Bokkeveld	71.92 b	70.91 b	71.90 a	0.40	13.03	<0.0001
Light after Storage (value)	Elgin	74.58 b	74.75 b	75.65 a	0.56	7.88	<0.0001
	Koue Bokkeveld	74.83 b	75.05 b	75.90 a	0.74	5.4	0.0048
Hue at Harvest (degrees)	Elgin	113.13 *	113.29 *	113.07 *	0.24	2.01	0.134
	Koue Bokkeveld	113.38 b	113.62 a	113.31 b	0.19	6.06	0.0024
Hue after Storage (degrees)	Elgin	106.97 a	107.10 a	105.78 b	0.55	13.1	<0.0001
	Koue Bokkeveld	106.68 a	106.70 a	105.99 b	0.59	5.48	0.0045

Table 2: Pearson correlation matrix for all variables in **Elgin** for fruit from **Early** flowers. Values in bold indicate a significant correlation at a 5% confidence level.

Variable	Fruit aspect	Mass harvest	Lightness harvest	Lightness storage	Hue-angle harvest	Hue-angle storage	Starch conversion	Firmness harvest	Firmness storage
Fruit aspect	**1.00**	-0.02	-0.18	-0.02	0.20	-0.02	0.07	-0.07	-0.10
Mass	-0.02	**1.00**	0.13	-0.22	**-0.39**	-0.12	0.08	**-0.37**	**-0.54**
Lightness harvest	-0.18	0.13	**1.00**	**0.87**	**-0.79**	**-0.86**	-0.02	0.19	0.40
Lightness storage	-0.02	-0.22	**0.87**	**1.00**	**-0.68**	**-0.86**	-0.12	0.27	**0.43**
Hue angle harvest	0.20	**-0.39**	**-0.79**	**-0.68**	**1.00**	**0.87**	-0.01	-0.06	-0.06
Hue angle storage	-0.02	-0.12	**-0.86**	**-0.86**	**0.87**	**1.00**	0.15	-0.19	-0.17
Starch conversion	0.07	0.08	-0.02	-0.12	-0.01	0.15	**1.00**	**-0.59**	0.02
Firmness harvest	-0.07	**-0.37**	0.19	0.27	-0.06	-0.19	**-0.59**	**1.00**	0.09
Firmness storage	-0.10	**-0.54**	0.40	**0.43**	-0.06	-0.17	0.02	0.09	**1.00**

Table 3: Pearson correlation matrix for all variables in **Elgin** for fruit from **Full Bloom** flowers. Values in bold indicate a significant correlation at a 5% confidence level.

Variable	Fruit aspect	Mass harvest	Lightness harvest	Lightness storage	Hue-angle harvest	Hue-angle storage	Starch conversion	Firmness harvest	Firmness storage
Fruit aspect	**1.00**	**0.30**	-0.29	-0.15	-0.05	0.01	0.31	**-0.35**	-0.17
Mass	**0.30**	**1.00**	0.00	0.06	-0.25	-0.21	**0.44**	**-0.73**	**-0.81**
Lightness harvest	-0.29	0.00	**1.00**	**0.85**	**-0.76**	**-0.86**	**-0.35**	**0.43**	**0.38**
Lightness storage	-0.15	0.06	**0.85**	**1.00**	**-0.66**	**-0.88**	-0.14	0.39	0.23
Hue angle harvest	-0.05	-0.25	**-0.76**	**-0.66**	**1.00**	**0.87**	0.10	-0.14	-0.10
Hue angle storage	0.01	-0.21	**-0.86**	**-0.88**	**0.87**	**1.00**	0.13	-0.30	-0.15
Starch conversion	0.31	**0.44**	**-0.35**	-0.14	0.10	0.13	**1.00**	**-0.69**	**-0.51**
Firmness harvest	**-0.35**	**-0.73**	**0.43**	0.39	-0.14	-0.30	**-0.69**	**1.00**	**0.76**
Firmness storage	-0.17	**-0.81**	**0.38**	0.23	-0.10	-0.15	**-0.51**	**0.76**	**1.00**

Table 4: Pearson correlation matrix for all variables in **Elgin** for fruit from **Late flowers**. Values in bold indicate a significant correlation at a 5% confidence level.

Variable	Fruit aspect	Mass harvest	Lightness harvest	Lightness storage	Hue-angle harvest	Hue-angle storage	Starch conversion	Firmness harvest	Firmness storage
Fruit aspect	**1.00**	**0.50**	0.14	-0.27	**-0.31**	0.20	**0.57**	**-0.58**	**-0.55**
Mass	**0.50**	**1.00**	-0.05	-0.22	-0.23	-0.01	**0.50**	**-0.72**	**-0.70**
Lightness harvest	0.14	-0.05	**1.00**	**0.85**	**-0.69**	**-0.81**	-0.04	0.08	**0.55**
Lightness storage	-0.27	-0.22	**0.85**	**1.00**	**-0.58**	**-0.86**	**-0.65**	0.35	**0.51**
Hue angle harvest	**-0.31**	-0.23	**-0.69**	**-0.58**	**1.00**	**0.82**	**-0.42**	**0.33**	-0.24
Hue angle storage	0.20	-0.01	**-0.81**	**-0.86**	**0.82**	**1.00**	**0.51**	-0.16	**-0.43**
Starch conversion	**0.57**	**0.50**	-0.04	**-0.65**	**-0.42**	**0.51**	**1.00**	**-0.82**	**-0.61**
Firmness harvest	**-0.58**	**-0.72**	0.08	0.35	**0.33**	-0.16	**-0.82**	**1.00**	**0.57**
Firmness storage	**-0.55**	**-0.70**	**0.55**	**0.51**	-0.24	**-0.43**	**-0.61**	**0.57**	**1.00**

Table 5: Pearson correlation matrix for all variables in the **Koue Bokkeveld** for fruit from **Early flowers**. Values in bold indicate a significant correlation at a 5% confidence level.

Variable	Fruit aspect	Mass harvest	Lightness harvest	Lightness storage	Hue-angle harvest	Hue-angle storage	Starch conversion	Firmness harvest	Firmness storage
Fruit aspect	**1.00**	-0.12	-0.18	-0.27	0.26	0.15	-0.27	0.41	**-0.60**
Mass	-0.12	**1.00**	0.13	-0.39	-0.10	0.22	-0.13	**-0.82**	-0.41
Lightness harvest	-0.18	0.13	**1.00**	**0.65**	**-0.68**	-0.51	**-0.69**	-0.23	0.39
Lightness storage	-0.27	-0.39	**0.65**	**1.00**	-0.25	**-0.86**	-0.63	0.15	0.04
Hue angle harvest	0.26	-0.10	**-0.68**	-0.25	**1.00**	0.30	**0.58**	0.16	**-0.69**
Hue angle storage	0.15	0.22	-0.51	**-0.86**	0.30	**1.00**	0.47	-0.21	-0.17
Starch conversion	-0.27	-0.13	**-0.69**	-0.63	**0.58**	0.47	**1.00**	0.00	-0.03
Firmness harvest	0.41	**-0.82**	-0.23	0.15	0.16	-0.21	0.00	**1.00**	-0.11
Firmness storage	**-0.60**	-0.41	0.39	0.04	**-0.69**	-0.17	-0.03	-0.11	**1.00**

Table 6: Pearson correlation matrix for all variables in the **Koue Bokkeveld** for fruit from **Full Bloom flowers**. Values in bold indicate a significant correlation at a 5% confidence level.

Variable	Fruit aspect	Mass harvest	Lightness harvest	Lightness storage	Hue-angle harvest	Hue-angle storage	Starch conversion	Firmness harvest	Firmness storage
Fruit aspect	**1.00**	0.10	**-0.47**	-0.46	**0.45**	**0.55**	0.09	-0.05	-0.06
Mass	0.10	**1.00**	-0.06	-0.28	-0.03	0.00	0.19	**-0.72**	**-0.79**
Lightness harvest	**-0.47**	-0.06	**1.00**	**0.72**	**-0.85**	**-0.76**	**-0.52**	-0.05	0.06
Lightness storage	-0.46	-0.28	**0.72**	**1.00**	**-0.62**	**-0.85**	-0.34	0.34	0.13
Hue angle harvest	**0.45**	-0.03	**-0.85**	**-0.62**	**1.00**	**0.81**	**0.63**	-0.13	0.01
Hue angle storage	**0.55**	0.00	**-0.76**	**-0.85**	**0.81**	**1.00**	0.34	-0.18	0.03
Starch conversion	0.09	0.19	**-0.52**	-0.34	**0.63**	0.34	**1.00**	**-0.51**	-0.36
Firmness harvest	-0.05	**-0.72**	-0.05	0.34	-0.13	-0.18	**-0.51**	**1.00**	**0.61**
Firmness storage	-0.06	**-0.79**	0.06	0.13	0.01	0.03	-0.36	**0.61**	**1.00**

Table 7: Pearson correlation matrix for all variables in the **Koue Bokkeveld** for fruit from **Late flowers**. Values in bold indicate a significant correlation at a 5% confidence level.

Variable	Fruit aspect	Mass harvest	Lightness harvest	Lightness storage	Hue-angle harvest	Hue-angle storage	Starch conversion	Firmness harvest	Firmness storage
Fruit aspect	**1.00**	**0.46**	0.03	-0.26	0.23	0.02	-0.19	0.03	**-0.57**
Mass	**0.46**	**1.00**	-0.15	-0.34	0.20	-0.03	0.10	**-0.61**	**-0.47**
Lightness harvest	0.03	-0.15	**1.00**	**0.77**	**-0.72**	**-0.82**	-0.27	0.19	0.19
Lightness storage	-0.26	-0.34	**0.77**	**1.00**	**-0.48**	**-0.72**	-0.24	0.27	0.03
Hue angle harvest	0.23	0.20	**-0.72**	**-0.48**	**1.00**	**0.63**	0.33	-0.19	**-0.58**
Hue angle storage	0.02	-0.03	**-0.82**	**-0.72**	**0.63**	**1.00**	0.20	-0.06	-0.12
Starch conversion	-0.19	0.10	-0.27	-0.24	0.33	0.20	**1.00**	-0.24	-0.27
Firmness harvest	0.03	**-0.61**	0.19	0.27	-0.19	-0.06	-0.24	**1.00**	0.25
Firmness storage	**-0.57**	**-0.47**	0.19	0.03	**-0.58**	-0.12	-0.27	0.25	**1.00**