

**Influence of various storage conditions on rind breakdown and quality parameters in tangelo (*Citrus reticulata* Blanco x *Citrus paradisi* Macf.)**

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## ABSTRACT

*Minneola tangelos* were obtained from different production areas in two seasons (1996: Western Cape and Swaziland, 1997: Western and Eastern Cape), and stored at two different temperature regimes simulating the shipping and storage treatments of fruits for overseas markets to determine if storage regimes have an influence on quality parameters. Samples were taken every two weeks and quality parameters such as fruit shape index, mean rind thickness, % juice, %TSS, and % acid were determined. An experiment to determine weight loss during storage time was done at the same time. Main significant differences across all the parameters were observed between fruits from the different production areas, while fruit shape, % juice, and occurrence of rind breakdown did not differ significantly. In 1996, the weight loss developed in storage only showed a significant difference after 8 weeks in storage. In 1997, there was no clear pattern, but the interaction between producers and storage temperatures differed significantly. Focusing on the producer from the Western Cape, there was a significant difference between storage temperatures, confirming that citrus fruits stored at a higher temperature (11 °C) will lose moisture faster than those stored at a lower temperature (4.5 °C). Even with the higher moisture loss, there were no differences between the fruit stored at the different temperatures, apart from the difference between the producers indicating that changes in quality of 'Minneola' fruit are mainly determined by the producers and apparently to a lesser extent by storage temperatures of 4.5 °C and 11 °C. With the high rate of over-maturity developing in storage, it is possible that the fruit might have been stored too long. Therefore, the marketing period for soft citrus types is apparently too long and problems with physiological ageing can be abundant at this storage length.

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## 1. General Introduction

During the 1993, 1994, and 1995 seasons, rind breakdown (RB) in 'Clementines' and 'Minneolas' caused substantial financial losses to producers, which had a detrimental effect on marketing, since the confidence in the cultivars suffered as a result of the problem. As OUTSPAN International exports a variety of soft citrus types to the overseas markets, these losses had a great impact on the revenue earned by the company. For example, in 1994 1.38 million cartons each of 'Clementines' and 'Minneolas' were exported, of which 8% suffered from RB, accounting for 110400 cartons per cultivar. The direct losses per cultivar were  $\pm$  R5.5M (Van Rensburg et al.,1995). The Minneola tangelo is not a fast-selling fruit on the overseas market and buyers have expressed an interest in storing 'Minneolas' for sale in Europe through September. According to Du Toit Pelser and Lesar (1994), 'Minneolas' develop chilling injury (CI) or a pitting of the rind when stored at 4.5°C continuously for 10 weeks or more. It is, therefore, necessary to investigate temperature storage regimes in order to store the fruit for 16 to 18 weeks while limiting decay development and RB or CI.

Although postharvest rind disorders of citrus fruits cause severe economic losses, information on the physiological development of these disorders is not readily available. The occurrence of post harvest rind disorders displays an erratic pattern, since it may occur during one part of the season but not in another and only on fruit from some production areas or groves. Conditions in the grove, such as general climate, local weather conditions, soil, rootstock, cultivar, cultural practices, handling and other conditions during picking play an important role in the predisposition of the fruit to many of these disorders. Some of these factors are not controllable. Postharvest practices also influence fruit quality and storage life and need to be optimal to minimize or prevent the development of rind disorders. External quality factors such as peel colour, incidence of blemishes and fruit shape, are significantly affected by climate (Eaks, 1969; Grierson and Ben-Yehoshua, 1986).

Citrus fruit has the advantage over deciduous fruit in that it can be stored effectively on and off the tree without loss of flavour. It is a common practice to extend the harvest period of many cultivars after they have reached maturity, to be able to optimize factors such as profitability, orderly marketing, weather, and available manpower. However, postbloom storage can severely limit the potential for postharvest cold storage and reduce the next year's yield (Grierson and Ben-Yehoshua, 1986).

When studying the optimum conditions for storage it is important to know what the major hazards during the storage period might be. A significant portion of the crop might not even reach the

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consumer, because of losses due to postharvest decay or quality deterioration (Wenda and Kelly, 1987). According to Grierson and Ben-Yehoshua (1986) the commercial life span of harvested citrus may be reduced or terminated because of one or more of the following processes:

- decay
- weight loss and resulting shrinkage
- over-maturity resulting in off-flavour and undesirable colour changes
- softening
- physiological disorders in general
- chilling injury (CI) in cold storage.

Citrus fruit generally become unmarketable because of development of decay or peel shrinkage, even though the juice vesicles are still turgid and the fruit flavour remains normal. The postharvest life of fruit depends on interactions between the physiology of the fruit and its pathogens. These interactions need to be manipulated by various storage techniques to prolong the life span of stored fruit (Grierson and Ben-Yehoshua, 1986).

A few cultural practices have a direct effect on post harvest keeping quality. Fruit harvested early in the season is more resistant to decay but more susceptible to oleocellosis and other harvest induced blemishes. As the season progresses, the fruit also becomes more susceptible to decay. It is important to determine the optimal harvest time for prolonged storage for each variety (Grierson and Ben-Yehoshua, 1986). Careful harvesting is the single most critical factor in fruit storage. Fruit that has been abused during harvesting will have a very poor storage potential (Grierson and Ben-Yehoshua, 1986). Reported effects of fertilizer treatments on storage quality are highly inconsistent. Generally, high levels of nitrogen decrease storage potential while addition of potassium phosphate increases it (Grierson and Ben-Yehoshua, 1986; Reitz and Embleton, 1986). An absence or minimum doses of nitrogen fertilizers produces fruits with very thin skin. These fruit appear to be more subject to the physiological disorder called 'pitting', while maximum application of nitrogen produces fruit with a thick skin and notably inferior yield of juice (Reig, 1969). According to Wenda and Kelly (1987) very small or very large citrus fruit has a tendency to develop problems during post harvest storage. There are year-to-year changes in maturity and hence storage potential, which are probably primarily influenced by climatic factors. When the fruit reaches earlier maturity, an earlier harvesting by 2 to 4 weeks can be required. The storage potential is also influenced by the location of the groves. In Israel, fruit from coastal areas with light soils tends to develop more decay than fruit from the central parts of the country on heavy soils (Grierson and Ben-Yehoshua, 1986).

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Suitable climatic conditions and orchard treatments during fruit development are of importance in conditioning the fruit for better viability and longer life, both on the tree and in storage (Monselise, 1981). Climate has a dominant effect on fruit quality. All the fruit produced in a specific regional climatic area share a common set of quality characteristics. Areas with hot, dry summers and cool, more humid or wet winters (Mediterranean climate) generally produce fruit with brighter colour and coarser peel, than areas with more humid growing seasons and warmer winter nights. Cultural practices cannot completely overcome these climatic differences (Reitz and Embleton, 1986). Irrigation, aside from its benefits in setting a full crop had little effect on the external fruit quality, while high levels of nitrogen in orange trees are associated with thicker peel, lower percentage juice, coarser peel texture and greener fruit colour (Grierson and Ben-Yehoshua, 1986).

According to Tugwell and Chvyl (1996a) storage temperatures during shipping of Australian citrus, (subjected to long sea voyages to reach lucrative northern hemisphere markets) are a compromise between the low temperature that will minimize mould wastage and weight loss and the higher temperatures that will minimize the risk of chilling related blemishes. The losses due to lengthy export shipments can be higher than those in relatively short domestic shipments (Tugwell and Chvyl, 1996a). Maintaining a near to optimum environment during transport to overseas markets is essential for loss prevention and retention of product quality. The total cost of marketing depends on refrigeration and other handling costs. The importance of quality maintenance and loss reduction increases as agricultural products approach the final stages of marketing, since the costs increase as the products move through the marketing system (Harvey and Houck, 1986).

As the reason for the development of RB is not clear, a study to evaluate the effect of storage temperatures on moisture content and physiological properties and the incidence of rind breakdown of the fruit after harvest will help to explain possible mechanisms of development. Another problem with RB is that there is no warning system for the industry to know if the RB in the season will be high or not. This could influence the marketing strategy of the fruit, as RB development is usually higher at the end of the season (Van Rensburg et al., 1995). Physiological parameters such as sugar and acid content, percentage juice, rind thickness and weight loss are used extensively by the citrus industry to determine harvesting dates and standards for marketing at overseas markets (Van Wyk, 1996). The correlation of one or more of these parameters with the incidence of RB would therefore create a possible warning system for the development of rind breakdown later in storage.

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## 2. Literature Review

### 2.1. Rind breakdown

Rind breakdown (RB) is a known physiological disorder in citrus fruit. Some researchers see rind breakdown as a chilling injury (CI) or as oleocellosis symptoms. Others see it as a totally different physiological disorder (Van Rensburg et al., 1995; Du Toit Pelser and Lesar, 1995; Eaks, 1969).

Van Rensburg et al. (1995) classified RB in two types: early season sensitivity of fruit (especially fruit from the inside of the tree) and RB caused by prolonged storage (more than the maximum protocol time). According to them many physiological disorders occur on a cyclical basis, mostly related to crop levels, known as "on" and "off" years. They deduced that RB is possibly not the same as the classical CI, but that the RB in 'Minneolas' is similar to that of 'Clementines' (Van Rensburg et al., 1995). Du Toit Pelser and Lesar (1994) described RB as rind spotting and stippling. Van Rensburg et al. (1995) described rind breakdown (RB) on a morphological level. RB pits in the initial stages are always associated with one or more collapsed oil cells. Unknown factors cause the destruction of membranes of the oil cells. The oil leaks from the cells into the spongy albedo-area and destroys the structure of these cells. The oil, containing various volatile compounds and enzymes, destroys the sub-surface albedo cells. This coincides with a visible browning of the areas between the oil cells. Later the outer cell layers of the exocarp are also destroyed, with the resulting emptying of these cells as well. This stage coincided with the blackening of the surface area between the oil cells (Van Rensburg et al., 1995).

They also described RB on a physiological basis according to the "Shewfelt model" (Shewfelt and Prussia, 1993) for damage to plant tissues by stress. This model assumes that it is always the weakest membranes in an organ, which will collapse first (lose integrity) under stress conditions (Van Rensburg et al., 1995). Van Rensburg et al. (1995) found that the cell membranes of the oil cells are the most sensitive and are therefore the first to collapse under stress conditions. The defense mechanism against stress-induced breakdown of membranes involves  $\beta$ -carotene. The lack of carotene limits a plant's defense mechanism against the destruction of membranes (Van Rensburg et al., 1995). Citrus fruit from the inside of the tree is usually considerably bleaker in colour because of less carotene. This can increase the sensitivity to RB, especially if stored longer than the protocol period (Van Rensburg et al., 1995). Poorly waxed fruit, stored for long periods at low temperature, also has a higher degree of carotene loss and micro collapse of oil cells (Van Rensburg et al., 1995).

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## 2.2. Factors influencing the occurrence of rind breakdown

There are many possible reasons given as to what causes RB. The amount of rind breakdown in storage can be influenced by the variety and season, the temperature and duration, and atmosphere during storage as well as postharvest treatments (Smoot, 1969). There is also a strong indication that RB is caused by preharvest factors, made worse by postharvest treatments (Van Rensburg et al. 1995). Apparently moisture and/or temperature can cause this disorder (Offers, 1987). A high humidity after heavy rainfall followed by low temperatures and moisture loss due to dry conditions and high temperatures in the orchard as well as low temperatures during packing, storage and shipping have also been associated with the occurrence of rind breakdown in soft citrus (Offers, 1987; Smoot, 1969). Du Toit Pelser and Lesar (1994) found that RB or rind spotting occurred in fruit stored at 2, 4.5 and 8°C for 7 weeks. They concluded that this was possibly caused by a combination of water stress and heat stress, resulting in a more senescent peel at the time when the fruit was harvested. Low moisture content of the peel, due to pre- and postharvest conditions, could also have played a role (Du Toit Pelser and Lesar, 1994), and green fruit was more prone to RB during cold storage in 1995 (Du Toit Pelser and Lesar, 1995). Chun et al. (1990) also found Florida 'Minneola' tangelos to be very susceptible to storage decays severely limiting their export abroad. Shipping 'Minneolas' at 13-15°C might help to reduce RB in drought years but will increase decay in storage (Du Toit Pelser and Lesar, 1994).

According to Eaks (1969) RB occasionally occurs in oranges and grapefruit as a result of precooling, usually when air temperatures are considerably below 0°C and high air velocities for cooling are used. He reported this disorder to be more severe on fruit near the air inlet and concluded that the high velocity of cold air has a desiccating effect upon the rind. This can primarily be because, as the air initially comes in contact with the fruit, it is warmed, thus reducing the relative humidity and increasing the vapor pressure deficit of the air, having a severe drying effect. Lower initial air temperatures that are not saturated with water vapor for warmer fruit will increase the vapor pressure deficit and thus be even more damaging (Eaks, 1969).

When fruit is left over weekends before packing, it also can cause stress resulting in a higher RB. Stress can also be caused if the fruit is overheated by the sun ("cooked") in trucks waiting at depots, or storage with forced air-cooling at -1°C and the storage of fruit outside the protocol period (Van Rensburg et al., 1995). Moisture condensation on citrus fruit, when fruit is pre-cooled and then delayed at port before export, does not lead to increased levels of RB and decay, even if the fruit is left

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less of an influence on the development of RB than length of cold storage while RB of navels appears after prolonged storage at 5°C but not at 10°C on fruit from the same location. Therefore they concluded that it appeared to be more associated with chilling injury and not stem-end rind breakdown (Du Toit Pelser and Lesar, 1995). A fairly acceptable eating quality is maintained until rind breakdown appears, when off-flavor can be detected (Eaks, 1969). Du Toit Pelser and Lesar (1994) gave a combination of water and heat stress, resulting in a more senescent peel at the time the fruit was harvested, as a possible reason for RB development. They also mentioned that a low moisture content of the peel, due to pre-harvest and post-harvest conditions could also have played a possible role in the development of RB (Du Toit Pelser and Lesar, 1994).

Van Rensburg et al. (1995) put an action plan for 1995 together after the substantial losses in 1993 and 1994. They fully investigated all possible reasons for RB, by identifying orchards, their nutritional status, fruit size and position on the tree, maturity-selective picking, transport to the packinghouse, packinghouse treatments, cold storage, shipping, age of fruit and overseas reports.

Van Rensburg et al. (1995) found that the following fruit parameters were involved in RB development as a result of this action plan:

- Fruit was more sensitive to cold storage pitting damage between 3 and 8°C and fruit stored at 11°C developed more RB than the fruit from -0.5°C (temperature of most apple chambers).
- Green fruit did not develop more rind breakdown.
- Fruit picked early in the season developed more RB than fruit picked later. The initially high levels of RB in overseas storage decreased later in the season, until a flare-up of RB during the late part of the season, which could indicate the maximum storage period.
- With the use of forced air-cooling at 4.5 or 11°C no significant difference between the latter and static air-cooling could be found.
- Results with boron treatments were not significant, as the applications did not solve RB
- There was a significant difference between the sensitivity to RB on fruit from inside and outside of the tree. Fruit picked from the inside of the tree, waxed and stored at 11°C, had significantly more RB than fruit picked from the outside.
- No differences in fruit sprayed with preharvest plant growth regulators were found (synthetic auxins and an unregistered BASF compound were applied when stored for 8 weeks at -0.5°C).

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- There was no difference in RB development in fruit with different physiological ages (3 week difference in fruit from the "first" and "last" flower) when stored at temperatures preventing RB.
  - RB occurred mainly in the area between the middle and the top of the fruit.
  - Injured fruit had thinner peels than healthy fruit.
  - There was a significant difference in the pulp texture. The area directly adjacent to the RB was slightly dried out. This could have been from the over-usage of sugars of other cell constituents by the peel or by direct effect of oil leaking from cells.

They concluded that cultural practices to eliminate the generally small, poorly coloured fruit should be used or the penetration of more light in the trees should be allowed. Cultural management practices, such as suppressing the flowering levels with winter GA sprays, winter pruning to stimulate formation of a green blossom, pruning in spring to eliminate the small inside borne flowers and allow more light, manipulation of the GA<sub>3</sub> spray to prevent the set of small inside fruit, or the use of plant growth regulators to reduce the set of small inside fruit, could be used (Van Rensburg et al., 1995).

The storage temperatures resulting in the least RB is more or less the same as the recommended storage temperatures for 'Dancy' tangerine of 4.5 - 7.3°C at 93% RH for commercial storage up to 4 weeks (Grierson and Ben-Yehoshua, 1986). Du Toit Pelsler and Lesar (1995) also found that there was a difference in RB occurrence on different fruit sizes packed at the end of April 1995. RB occurred more on small fruit (count 117-200) than larger fruit (count 69-99) and occurred at all storage temperatures (2 to 8°C) when 'Clementine' Nules from Schoemanskloof near Nelspruit were stored. The same results with fruit size were found with Novas from the same area (Du Toit Pelsler and Lesar, 1995). The RB on 'Minneola' tangelos increased as the duration of the storage at 11°C increased. RB can therefore be a result of rind senescence. Du Toit Pelsler and Lesar (1995) found that CI or RB tends to increase as the storage period was increased, but was similar in fruit stored at 2°C, 4.5°C or 8°C.

Du Toit Pelsler and Lesar (1994) used 'Satsumas' from Elands Rivier Valley, near Nelspruit, for a RB storage trial. The fruit was harvested on 25 March 1994. RB occurred after 12 weeks of storage, at which time decay development was excessive when stored 1 week at 20°C, shipping simulation at 11°C for 3 weeks, storage at 2,4.5 or 8°C for 3, 5 or 7 weeks and 1 week at 20°C. The most RB occurred when fruit was stored for 7 weeks at 2, 4.5 and 8°C, but not when stored for 3 or 5 weeks at the same temperatures. The best results were obtained when fruit was "shipped" at 11°C (3 weeks) and

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then stored for 3 weeks at 2°C. Similar results were obtained with fruit harvested 16 April 1994. RB started appearing after a total of 10 weeks in storage, and also when stored for 5 weeks at 2 and 4.5°C. When a trial was done with 'Clementine' Nules, RB occurred when the fruit was "shipped" at 11°C. With this experiment the best results (lowest decay percentage, best colour, best flavour, least RB) were obtained by "shipping" green Nules at 11°C (3 weeks) and storage at 8°C (3 weeks) for a total storage period of 8 weeks (Du Toit Pelser and Lesar, 1994).

Van Rensburg et al. (1995) also reported that shipping temperatures had an influence on RB. The best results were obtained with shipping the fruit at 11°C, then storing at lower temperatures. The shipping temperature should be based on the time of harvest after the start of the season. The fruit could even be stored at -0.5°C, but not for longer than 5 weeks (Van Rensburg et al., 1995). Storage of 'Clemlate', 'Minneola', 'Navel' and 'Navelate' fruit for 5 weeks at -0.5°C, followed by 11°C, resulted in perfect internal and external condition. The shipping of the fruit at low temperatures (arriving with the same colour as when packed), storing overseas at higher temperature to allow for colour development or ship at 11°C, then 2°C to slow respiration down can be used to prevent occurrence of late RB. Storage temperatures of 4.5°C resulted in more damage than 11°C (Van Rensburg et al., 1995).

When 'Minneola' fruit from Malelane Co-op. was stored in 1986 for 10 weeks at 4.5 °C, only 0.1% RB developed, but fruit from the same Co-op., stored for the same duration at the same temperature in 1987, developed 18.7% RB (Du Toit Pelser and Lesar, 1994). Storage at a low temperature (4.5°C) in 1992 had the highest RB incidence, while the highest RB in 1994 was caused by storage for 8 weeks at 4.5°C, 8 weeks at 11°C and 4 weeks at 11°C, 12 weeks at 4.5°C (Du Toit Pelser and Lesar, 1994). This can possibly be due to the climatic influences of the different seasons (Du Toit Pelser and Lesar, 1994). It can also be the reason why fruit from Karino, stored in 1992 and 1993 for 16 weeks at 11°C, developed approximately 1.0% RB, but in 1994, fruit from the same area developed more than 40% RB after 16 weeks of storage at 11°C (Du Toit Pelser and Lesar, 1994). It could be concluded that fruit from the relatively cool Karino area seemed not to be prone to RB in 1992 and 1993, while in 1994 the opposite was true.

The length of the storage period is important. Even tougher outside fruit developed RB at the end of the storage period. A possible explanation for this could be the following: respiration, requiring a substrate energy source, takes place in citrus fruit after harvest. As limited amounts of sugars are available in the fruit for utilization in the form of energy for respiration, sugars are moved from the

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pulp (high in soluble sugars) to the peel, which is the major interface where respiration takes place (the lenticels through which the gas exchange - also needed for respiration - are situated in the peel). As long as enough energy (sugars) is supplied from the pulp to the peel, the metabolic processes take place normally. However, over longer periods of storage, the energy source directly next to the peel might become exhausted ("drying out" of the pulp next to the RB peel). The membrane systems are then at risk and collapse, possibly causing RB (Van Rensburg et al., 1995).

Van Rensburg et al. (1995) also found that a preharvest Ethrel treatment had no effect on RB and postharvest ethylene degreening caused higher levels of RB. This could be because the temperature of postharvest degreening treatment was substantially higher than the specified range of 21°C, which could result in an earlier onset of RB (Van Rensburg et al., 1995).

### **2.3. Other physiological disorders of stored citrus fruit and possible causes**

Apart from RB citrus fruit storage can also be influenced by other physiological disorders. These physiological disorders are any non-parasitic maladies of citrus fruits - regardless whether caused by weather, nutrition, physical handling operations, or postharvest ambient conditions (Grierson, 1986). Physiological rind disorders can be classified in two categories. The first is associated with harvesting and the packinghouse operations and preparing the fruit for storage or the market (e.g. oleocellosis and rind staining). The second occurs during precooling or storage resulting in pitting or general rind breakdown (Eaks, 1969). Physiological disorders produced by low temperatures in citrus fruit cause a decrease in the commercial value of the fruit due to the loss of its organoleptic as well as visual properties (Rodriguez et al., 1981). Although Grierson and Ben-Yehoshua (1986) reported that most physiological disorders affecting fruit storage tend to be related to water loss or to chilling injury, a clear classification cannot be made between the different disorders and their causes. Because of this a study was made of most of the physiological disorders affecting the storage of citrus fruits. Although not all the causes are the same as that for rind breakdown, an intensive study will show what factors influence the development of the main physiological disorders and will give a better understanding of the physiology of citrus fruit.

Chilling injury (CI) can be described as storage pitting in which discrete areas of the peel collapse forming sunken lesions (Grierson, 1981). Cold pitting and oleocellosis together with penicillium decay used to be the main factors determining the storage life of oranges (Pratella et al., 1969). Surface pitting and rind scald is usually typical of chilling injury, which can be observed as dark, depressed necrotic tissue (McDonald et al., 1991; Murata, 1981; Rodriguez et al., 1981).

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(1988) described pitting as when oil cells in the deeper parts of the outer mesocarp or flavedo burst and the oil is forced in the surrounding flesh. The upper epidermis is not damaged, but the oil causes plasmolysis of the cells in the flavedo. These cells collapse and cause a depression in the peel over the damaged cells. It is mainly the deeper, younger oil cells that burst first (Pratella et al., 1969). In the case of citrus in Japan, pitting is observed at storage temperatures from 0-30°C, with the highest rate at room temperature. The chilling sensitivity is important in CI development (Murata, 1981). Minimum temperatures needed to induce symptoms of CI and susceptibility to injury vary depending on the time of harvest, colour or maturity, region of the tree from which fruit is harvested, their post-harvest handling and preharvest conditions (Aljuburi and Huff, 1985).

Calcium oxalate crystals appear in fleshy tissue of 'Persian' limes before cold damage in epidermal cells is detected. The crystal concentration increases the longer the fruit is stored at low temperatures. The appearance of crystals should be noted as it can be an indication before damage occurs (Bertolini et al., 1986). Cohen et al. (1994) also reported the development of microscopic cracks in citrus peel due to exposure to temperatures that cause chilling injury.

Membranosis is a type of CI that occurs in lemons at temperatures between 2 and 5°C and is slowed by temperatures of more than 10°C or in the region of 0°C (Bertolini et al., 1986). Contrary to the findings of other researchers, no significant differences were noted in the properties of the juice, which was detectable by chemical analysis or organoleptic tests on lemons affected by membranosis. The weather conditions of the individual years and the microclimates found in the areas in which lemons are grown can have a significant impact on the occurrence of the disorder. This physiological disorder spreads rapidly during the first 30 days of storage. An analysis of the mineral composition of the membranes showed no direct relationship to its susceptibility to CI (Bertolini et al., 1986).

Squalene, a triterpene is present in the epicuticular wax of citrus fruit and the squalene level was reported by Bertolini et al. (1992) to have an inverse effect on CI. A prestorage conditioning of 15°C favoured the biosynthesis of squalene. The postharvest application of squalene also reduced CI. 'Valencia' oranges were treated after harvest with squalene and stored at 1°C and 5°C. At both temperatures squalene drastically reduced the incidence and severity of rind pitting in the first 60 days of storage. At the final control, after 90 days, the efficacy of the treatment was sensibly reduced. It had no detrimental effect on taste or appearance (Bertolini et al., 1992). Postharvest applications of squalene can also reduce peel pitting for the first 60 days of 'Valencia' fruit stored at 1 and 5°C. After 90 days the efficacy was reduced (Zaragoza et al., 1996).

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Tugwell and Chvyl (1996b) found that during the 1993 season less than 3% of fruit from a first shipment of Navels stored at 5 or 8°C developed CI. Negligible chilling injury also occurred on fruit stored at higher temperatures. Significant CI in the form of Navel end collapse and rind blemish developed on 5.6% of fruit stored at 2°C in 1994. Rind blemish due to CI was almost non-existent when 'Navel' oranges were stored at 10°C for two weeks followed by 5°C. Chilling increased to 3% at 5°C and 5% at 2°C, but mould wastage and weight loss were reduced at the lower temperatures (Tugwell and Chvyl, 1996b).

Castro-Lopez (1992) found that 'Persian' limes stored at 10°C for 20 days showed no CI, whereas other stored for a similar period at 7°C developed 23% CI. The longer the fruit was stored the more CI developed. When stored for 30-35 days at 10°C stylar-end rind breakdown, chilling injury and the loss of commercial green colour increased. A cold treatment can also be used as a quarantine treatment against Mediterranean fruit fly (Chalutz et al., 1985b). There was no difference in the incidence of CI between 'Marsh' Grapefruit treated at 0°C for 10 days or at 2.2°C for 16 days as regulations require. The cold treatment enhanced decay development during long-term storage of the fruit at 11°C. Mould rots developed on the CI peel pitting, and their incidence increased from 1.7% to 3.5% during a storage period of 12 weeks. The presence of Thiabendazole (TBZ) in the wax treatment reduced the incidence of CI up to 50%. Delayed cooling, that is keeping the freshly harvested packed fruit for 6 days at 17°C prior to the initiation of the cold treatment, reduced the incidence of CI by the same extent. By combining TBZ with delayed cooling, CI can be reduced, and cold treatment can be practiced with a low risk of CI and subsequent decay development (Chalutz et al., 1985b). McDonald et al. (1991) found that fruit dipped in fungicides had less CI than fruit dipped in water alone. Imazalil was more effective in reducing CI than TBZ. Grapefruit could be stored at 5°C for 6 weeks with an Imazalil treatment (McDonald et al., 1991).

Grapefruit conditioned for at least 7 days at 10, 16 or 21°C before storage at 1°C had significantly less CI than fruit stored continuously at 1°C or conditioned for 2 days. Intermittent warming reduced CI in 'Marsh' grapefruit when stored at 4°C. Treatments of 10°C and 16°C gave better results than 21°C (Hatton et al., 1981). However the technology of intermittent warming for a reducing effect on CI, has limited commercial use (Pratella et al., 1992). The gradual lowering of temperature to 1°C can also reduce CI (Hatton et al., 1981).

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Waxing the fruit also has a reducing effect on the incidence of cold pitting (Pratella et al., 1969). The susceptibility to CI varies according to species and cultivar. Grapefruit, lemon and lime are much more susceptible to CI than orange and mandarin. Orange cultivars grown in Florida are reported to be less sensitive to CI than those grown in California and Arizona (Kader and Arpaia, 1992). Chalutz et al. (1985a) drew a comparison between the storage temperatures (0-17°C) of grapefruit, 'Shamouti' and 'Valencia' oranges and lemon. No CI was found at 12°C or higher, and the highest CI incidence was found at 6°C in 'Shamouti' and 'Valencia' and at 2°C in lemon. Grapefruit and 'Shamouti' oranges exhibited the highest susceptibility to CI. Chalutz et al. (1985a) concluded that an increased susceptibility of the cultivar to CI could be correlated with an increase in ethanol content at low temperatures. They did experiments with each cultivar, with fruit picked at various dates during the harvest season, from different growing regions and in several years. There were some differences from year to year. Most CI developed only after 4 weeks storage at the chilling inducing temperature. Peel pitting is usually followed by increase in decay development mostly during the shelf-life period (Chalutz et al., 1985a). Early season fruit is more susceptible to CI than fruit harvested later (Harvey and Houck, 1986).

'Washington' Navels harvested in Australia by Tugwell and Chvyl (1996b) during June and July showed rind disorders such as oleocellosis and rind spotting, especially during cold wet conditions. Storage at 10°C for a voyage time of 6 weeks resulted in weakened rind, which increased the risk of developing chilling related rind blemishes. To determine the most suitable temperature to minimize rind blemish caused by chilling, moulds and weight loss, fruit were stored at 2, 5, 8, 10°C and a step down temperature of 10°C for 2 weeks followed by 5°C. Storage times were 5, 7 and 9 weeks. After a week at 20°C to simulate retail conditions fruit was assessed for rind blemishes, mould wastage and internal quality (TSS and Acid). Rind blemish attributed to chilling injury was almost non-existent when Navel oranges were stored at 10°C or 10°C for 2 weeks followed by 5°C. Chilling increased to 3% at 5°C and 5% at 2°C but mould wastage and weight loss were reduced at the lower temperatures (Tugwell and Chvyl, 1996b).

Several other techniques are available to reduce CI. These include temperature conditioning, step-down temperature regimes, periodic warming, controlled atmosphere storage, chemical treatments (fungicides benomyl and thiabendazole) and applications of plant growth regulators (2,4-dichlorophenoxyacetic acid (2,4-D) and Gibberellic acid (GA<sub>3</sub>)). Pre-storage vapour heat treatments were also investigated (Wright et al., 1996; Aljuburi and Huff, 1985). Low temperature in conjunction with controlled atmosphere (CA) storage is not used commercially for citrus because it is not very

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superior to the ordinary storage procedure, but has a great influence on the cost of storage (Manago, 1981). Curing for a week at 14°C before storage at 6°C yielded good results and gave a 30% reduction in damage (Bertolini et al., 1986). In the adaptation of citrus to chilling stress, flavedo fatty acids play a significant role (Mulas et al., 1996). Total fatty acid content was greatly influenced by curing. The efficacy of the curing (3 days at 37°C in a water saturated atmosphere) treatment to avoid CI appears to be associated with the induction of an increase in fatty acid biosynthesis (Mulas et al., 1996). Slutzky et al. (1981) found that the ethanol and acetaldehyde contents are both higher in CI damaged fruit, especially ethanol.

Although the influence of some mineral elements, particularly calcium, magnesium and potassium, on the development of chilling injury has been established in the case of other plant species, research carried out on citrus has not produced conclusive results (Bertolini et al., 1986; Van Rensburg et al., 1995). Slutzky et al. (1981) found that healthy flavedo tissue had a significantly lower Ca content than tissues in diseased grapefruit. There was an increase in Ca content of juice during storage, especially in fruit staying healthy until the end of storage. The mineral composition of grapefruit and limejuice may be related to pitting during cold storage (Slutzky et al., 1981). Significant differences in Ca, Mn, Fe, Zn and in some cases Mg in juice from healthy or damaged fruit were observed and the juice from damaged fruit showed a higher Ca content than that of healthy fruit. A positive relationship was found between low Ca juice content and a higher percentage of damaged limes under cold storage. Due to the influence over the mineral composition of juice, rootstocks could regulate CI damage of lime in cold storage (Slutzky et al., 1981).

Fruit position on the tree and ripening stage influences fruit sensitivity to peel pitting. Fruits located on the North Western external position on the tree are more affected than those located on the opposite position in Spain (Zaragoza et al., 1996). They found coloured fruit to be more sensitive than green fruit, although the sensitivity disappeared after colouring. These two types of fruit only differ in citicolic permeability. Applications of calcium nitrate and natural and synthetic antitranspirants reduced peel-pitting incidence on 'Fortune' mandarin under field conditions. These applications also reduced citicolic permeability. These effects were probably due to a reduction of water loss from peel tissue (Zaragoza, et al., 1996). Substances capable of reducing peel transpiration were tested by Vercher et al. (1994). Applications gave an irregular response, depending on the orchard, the year and the storage temperature regime. The effectiveness of the applications increased as incidences of peel-pitting increased. Montana wax, Emultrol and beeswax were the most effective. Effectiveness was linked to the effect in reducing citicolic permeability. Depression of the rind surface caused by peel

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pitting caused a loss of crystalline wax structures (Vercher et al., 1994). Chalutz et al. (1981) found that different cultivars had different responses to the different temperatures. Grapefruit suffered the most from CI, while 'Shamouti' and 'Valencia' suffered little decay in cold storage but much more during shelf life. When stored at 12°C, there was no evidence of CI in any of the cultivars. This breakdown during shelf life might be associated with microscopic peel injuries, which formed during cold storage. These injuries (which can be made visible by dipping fruit in a 0.5% water solution of indigo carmine) probably enabled fungus to penetrate the peel and caused a high incidence of mold rot (Chalutz et al., 1981).

Apart from CI and RB there are quite a few other physiological disorders affecting citrus fruit. The physiological disorders in different varieties of lemon, seen during cold storage in normal and controlled atmosphere, were studied by Rodriguez et al. (1981). The parameters they considered were the period of storage between 1 to 2 months, temperature between 10°C and 14°C, relative humidity between 85% and 99%, and in controlled atmosphere the level of O<sub>2</sub> between 3-5% and 13-16% and CO<sub>2</sub> between 0% and 7-8%. Red blotch or peteca and membranosis were identified in different stages of intensity. Red blotch appeared more frequently when the temperature was higher, but peteca and membranosis appeared more frequently when the temperature was lower, and olleocellosis generally appeared when temperature was higher. Peteca and membranosis were observed most commonly at relative humidities approaching 100%. Relative humidity was more influential than controlled atmospheres in the development of these physiological disorders (Rodriguez et al., 1981). Membranosis of lemons also increased with time of storage, especially when stored at 5°C for 60 days and longer, but not when stored at temperatures lower than 5°C, when pitting was evident. Susceptibility to membranosis varies with the microclimates of different production areas (Pratella et al., 1992). Artes Calero et al. (1981) described Peteca in lemons as a physiological disorder that appeared as a deep sinking of the fruit's surface often with disorganized tissues. Sometimes it turned brown but might also stay white. The flavedo appeared normal at first, but later it dried and collapsed and secondary organisms might enter. Sometimes the disorder appeared in fruit sprayed with heavy oils before picking. Low temperatures in the storage chamber, excessive waxing and bad ventilation also aggravated it (Artes Calero et al., 1981). The postharvest development of peteca rind pitting in 'Meyer' lemons (*Citrus meyeri* (Y.) Tan) studied by Wild (1991) was aggravated by citrus wax application and fruit brushing. An increased brushing time increased the incidence of the disorder. Large mature fruit on the east side of the tree were more susceptible (Australia). The disorder also increased when fruit was brushed immediately after arriving from the orchard. Fruit should be stored 3-4 days before being waxed (Wild, 1991).

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According to Grierson (1981) rind staining is maturity related. It happens when the peel is so physiologically over-mature that the epidermal wax has softened so that handling causes reddish-brown blemishes. Rind staining can be prevented by the use of gibberellin sprays to delay peel senescence (Grierson, 1981). Harvest-induced physiological disorders like stem-end rind breakdown (SERB) can be initiated by an ill-defined imbalance in N and K nutrition, but its development as an economic hazard depends on handling procedures between picking and packing although the lesions usually develop until after shipment. The development of SERB in susceptible crops can be prevented or greatly diminished by never letting the fruit dry out in the period between picking and waxing (Grierson, 1981). There is a strong need to codify the interactions of weather, cultural programs, varietal characteristics, harvesting methods and postharvest conditions involved in these occasionally costly failures of citrus fruits (Grierson, 1981).

Pitting in 'Pineapple' oranges develops after harvest particularly if fruit are held at low humidity. It can also be associated with the size of the crop on the tree. The heavier the crop, the greater the tendency towards pitting (Grierson, 1986). Zebra skin of tangerines occurs when albedo and flavedo cells rupture because of a sudden excess of soil water taken up by tangerine trees that had been in a state of moderate to severe water stress. If fruit is picked 5-7 days after the rain or irrigation that caused the problem, normal handling will rupture the distended epidermal cells, causing a blackening of the peel over the individual fruit segments. Fruit very often responds in the same way for different handling practices, for example the injuries caused by irradiation treatments are usually indistinguishable from those due to more traditional treatments, such as chilling injury. The association of particular symptoms with a variety of causes will continue to complicate diagnosis of the physiological disorders of citrus (Grierson, 1986).

Another physiological disorder described by Hasegawa and Iba (1981) is Kohansho. This is the pitting on the stem end or on the periphery of the fruit. It can be described as relatively small sunken spots that gradually discolour and finally become brown. The size of the spots tends to increase. Kohansho seems to be induced by more or less the same factors as rind breakdown. This can be because it is a different physiological disorder, or the same disorder described by a different name, although Kohansho develops 2-3 days after transfer, while rind breakdown only develops later in storage. Its occurrence and symptoms vary among varieties and fluctuate year by year (Hasegawa and Iba, 1981). Hasegawa and Iba (1981) and Hasegawa and Yano (1992) investigated the cause and established a way to control the occurrence of Kohansho. Fruit was pricked with a needle 1mm deep to simulate the injury caused by the green leafhopper (*Empoasca* spp.). They found that the injuries did not respond to

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storage in the same manner as Kohansho. The susceptibility to Kohansho varied among varieties of 'Satsumas', Navels and lemons. Kohansho seemed to be more related to CI. An ethylene treatment reduced the rate of Kohansho in Navels (Hasegawa and Iba, 1981). The peel of Kohansho fruit had lower levels of sugar especially reducing sugars. Respiration of flavedo tissue isolated from fruit with Kohansho was higher than that of sound fruit. The ethylene production resembled that of wounding, disease, radiation and treatment with certain chemicals. To control Kohansho, fruit must be cold stored below 10°C. Individual wrapping with polyethylene film (0.02mm) and coating with wax can effectively suppress Kohansho (Hasegawa and Yano, 1992; Hasegawa and Iba, 1981). 'Kyomi' tangor (*Citrus unshiu* Marc. x *Citrus sinensis* Osb.) is particularly susceptible to Kohansho during storage at 10 to 20°C and after transferring to room temperature from storage at a low temperature (5°C). Hasegawa and Yano (1992) found that Kohansho appeared two to three days after shipping. High temperature curing of 20°C at ethylene degreening before storage reduced Kohansho. This seemed to advance the maturity of the peel. A treatment with wax and Thiabendazole (TBZ) effectively suppressed Kohansho.

Hasegawa and Yano (1992) also found that dropping of the fruit produced similar results as Kohansho. The unsaturated fatty acid contents of the rind decreased in Kohansho fruit, but calcium content was not affected by the occurrence of Kohansho. 'Kiyomi' tangor stored at 5°C for 2 months, then transferred to 10°C, 15°C and (ambient) 10-18°C, developed Kohansho 2-3 days after transfer. It was higher at higher temperatures and the most at ambient temperature with a relative humidity (RH) of 60%. Occurrence of Kohansho varied among trees, fields and years (Hasegawa and Yano, 1992). It was concluded by Hasegawa and Iba (1981) that humidity during storage, mineral nutrition, soil moisture, tree vigor and handling after harvest might affect the occurrence of Kohansho.

The thickness of the rind of 'Satsuma' mandarin increases under high humidity due to the increasing of volume of the oil glands. This causes a physiological disorder called puffy fruit, where the rind is detached from the segments (Murata, 1981). Rind puffing of 'Satsuma' mandarin is caused by water absorbance by the peel, destruction of albedo structure and reduced water content of the pulp. This leads to an increase in gas volume in the fruit. Rind puffing is accelerated by high temperature and high humidity in late maturing fruit and during storage. Storage of 3-5°C and 85% RH will minimize puffing (Manago, 1981).

Another physiological disorder occurring on citrus fruit is oleocellosis, which is caused by the rupturing of the oil cells during harvest (Esterhuizen, 1988). The oil is phytotoxic and causes the

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flavedo cells surrounding the ruptured oil cells to collapse and die. Visible oleocellosis lesions appear 1-4 days after damage as necrotic lesions on the rind surface of the fruit (Esterhuizen, 1988; Gilfillan, 1996). There is a close relationship between oleocellosis incidence and the relative humidity at the time of picking. The normal recommendations to citrus growers in South Africa for avoiding oleocellosis losses are to stop irrigation in time for the soil to dry before harvest, and to delay picking each morning until the fruit is dry following overnight dew. Damage is more likely to occur when the fruit is fully turgid in the morning (Esterhuizen, 1988). Oleocellosis incidence in South Africa is far greater in close-planted orchards or where the row orientation is not North to South. This happens more in the Eastern-Cape where picking starts before the sun rise above the horizon in winter and the vapour pressure deficit (VPD) between the fruit and the surrounding air is slow to rise to acceptable levels after irrigation. In the warmer and drier "Transvaal" production areas, oleocellosis is not nearly such a problem as in the Cape (Esterhuizen, 1988). Gilfillan (1996) described it as a grade-reducing factor to which fruit from some orchards are more susceptible than others. Two procedures that should both be carried out for accurate predictions of fruit susceptibility are the determination of VPD and rind turgidity or Rind Oil Release Pressure (RORP). Oleocellosis incidence is reduced to a minimum when VPD reaches 3 mm Hg. The proportion of fruit with RORP values below 3kg is related to the potential oleocellosis incidence following commercial picking, provided there is no vegetative flush on the trees. When vegetative flush is present on the trees, oleocellosis incidence is higher than predicted by the RORP test (Gilfillan, 1996). Highly significant differences in the levels of oleocellosis were found according to the time of day of harvest. The best harvest time for reducing the incidence of this injury was at 14h00. Harvesting should take place in low humidities. The harvesting method and time in the field after harvest did not influence the appearance of oleocellosis significantly (Castro-Lopez et al., 1981).

## **2.4. Fruit quality parameters**

### **2.4.1. Physiology of fruit in storage.**

The physiology of citrus fruit is a very important factor when considering storage of the fruit. The citrus fruit is a hesperidium, a specialized berry (Kader and Arpaia, 1992; Davies and Albrigo, 1994). Citrus fruits are non-climacteric, lacking both a ripening cycle and a well-defined abscission period, typical for climacteric fruit, such as apples. Therefore post harvest ripening cannot occur and the fruit has to be at the correct ripening stage before harvesting (Soule and Grierson, 1986). Unlike fruit of the climacteric class, showing increased ethylene evolution during ripening, citrus fruit normally produce very little ethylene (Thomas, 1986). Citrus is low in starch reserves and thus undergoes very slow

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changes in internal quality during storage. Protracted storage does decrease acid content converting it to sugars and CO<sub>2</sub> used in respiration (Davies and Albrigo, 1994).

Citrus fruit has a leathery peel surrounding the edible portion of the fruit. The fruit peel consists of an outer coloured exocarp (flavedo) and an inner white spongy mesocarp (albedo). The flavedo consists of the epicure proper, hypodermis, outer monocarp and oil glands. It can be described as a multi-layered "protective skin" of complex origin, structure and development over the epicure and usually separated from the latter by a pectin layer (Soule and Grierson, 1986; Davies and Albrigo, 1994). This leathery rind protects the fruit from damage during handling and desiccation during storage (Davies and Albrigo, 1994). The edible portion (endocarp) comprises of the inner portion of the carpals, expanding into segments containing juice vesicles and seeds. Citrus fruits are unique in having juice vesicles (sacs) emanating from the carpellary membranes (Davies and Albrigo, 1994). Changes in external colour can also continue after harvesting (Grierson and Ben-Yehoshua, 1986).

#### **2.4.2. Sugar and acid metabolism of stored fruit**

Fruit quality is usually determined by measuring the total soluble solids (TSS) and percentage of organic acids in the fruit. Total acidity is also important in overall juice quality and in determining the optimum harvest time. Mostly the ratio of TSS and titratable acids (TA) determine when fruit can be harvested (Davies and Albrigo, 1994; Van Wyk, 1996). TSS of citrus fruit accounts for 10-20% of fruit fresh weight (Davies and Albrigo, 1994). The TSS consists mainly of carbohydrates, such as monosaccharides (glucose, fructose), oligosaccharides (sucrose), and polysaccharides (cellulose, starch, hemicelluloses, pectins). Sucrose is the primary non-reducing sugar and is the major translocatable carbohydrate. Fructose and glucose are the major reducing sugars and are present at about one-half to equal quantities of sucrose in most citrus juices. Small quantities of mannose and galactose have also been found in citrus juice. All these carbohydrates represent approximately 70 - 80% of the total content of TSS (Davies and Albrigo, 1994).

Organic acids contribute significantly to overall juice acidity (Davies and Albrigo, 1994). The primary organic acid accumulating in citrus juice is citric acid, followed by malic and oxalic acids and free amino acids plus nitrogenous bases and glutathione (70-90%). The remaining components are inorganic ions, vitamins (2.5%), flavenoids and volatiles (1.2%), and lipids (1.2%) (Soule and Grierson, 1986; Davies and Albrigo, 1994). An analysis of the composition of different citrus types shows typical differences among cultivars and species. The free amino acid content also varies both

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quantitatively and qualitatively during fruit development (Soule and Grierson, 1986; Davies and Albrigo, 1994).

Organic acid levels generally decrease seasonally as citrus fruit matures. TSS levels increase as fruit size increases, becoming nearly constant or slightly increasing during the last stage of development (Davies and Albrigo, 1994). Echeverria and Ismail (1987) stored four citrus varieties ('Hamlin' orange, 'Marsh' grapefruit, 'Robinson' tangerine, and 'Palestine' lime) at 15°C and 95% RH for different periods of time and analyzed it weekly for TSS (°Brix), sucrose, fructose, glucose, citric and malic acid. In 'Hamlin' oranges, 'Robinson' tangerines and 'Palestine' limes TSS increased during storage, while it remained unchanged in 'Marsh' grapefruit. A significant decrease in citric acid was observed in all the varieties. The increase in TSS can be attributed to an increase in sucrose content. Relating changes of stored carbohydrates to the enzymes involved in their metabolism would provide a framework for understanding the metabolic processes that take place during the post harvest life of citrus fruits (Echeverria and Ismail, 1987).

Various enzymes control the processes whereby organic acids are changed and sugars are oxidized. The relationship between activities of gluconeogenic and glycolytic enzymes and postharvest changes in sugars and organic acids during storage of 'Valencia' oranges at 15°C was investigated by Echeverria and Valich (1989). Activities of the enzymes of acid metabolism (malic enzyme, isocitrate dehydrogenase/aconitase, and alcohol dehydrogenase) either increased during the first 3 weeks (malic enzyme) or remained constant during storage. Activities of enzymes involved in sugar catabolism (hexokinase, sucrose synthase, UDPG pyrophosphorylase and PPI-dependant phosphofruktokinase) increased during storage. These enzymes are necessary for organic acid use and for the subsequent oxidization of sugars in harvested sweet oranges (Echeverria and Valich, 1989).

The decline in organic acids has been linked to the use as energy substrates and to translocation to the peel. The activity of glukokinase is higher at pH 7.0 and higher than that of fructokinase. Organic acids can also serve as building blocks for the synthesis of sugars. Decline in acid content of harvested citrus fruit is at least in part due to the use of organic acids for energy production and alcoholic fermentation. Increase in acid-metabolizing enzymes is in accordance with this idea (Echeverria and Valich, 1989).

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### 2.4.3. Effect of water relations on rind disorders

One of the relatively unknown factors in the study of rind breakdown in citrus fruits is the role of water relations during fruit development and its effect on postharvest performance. One of the factors controlling fruit growth is the water supply through the roots while the rind texture is controlled by relative air humidity (RH). Daily variations in fruit size may be, therefore, depressed if soil moisture decreases or very low air humidity prevails. Since the peel is a visible marketing feature, peel appearance is just as important or even more important than the flavour of the pulp. Fruits serve as a storage organ for water and the ultimate fruit size is increased by irrigation and/or rainfall. Leaves on a detached fruitless twig will wilt within hours, while those on a detached twig with fruit will remain turgid for many hours. The majority of the water translocated to the leaves is stored in the fruit peel (Davies and Albrigo, 1994).

Water can be withdrawn from the peel with relatively more ease than from the pulp due to a lack of direct vascularisation of the vesicle stalks. This is one of the causes for the lack of turgor of the peel at times of water stress and the development of peel softness after prolonged storage (Grierson and Ben-Yehoshua, 1986). Most of the water vapour and other gases that move out of the detached citrus fruit, flow freely throughout the spongy parenchyma of the mesocarp (albedo) and the central axis, diffuse through the exocarp (the coloured portion of the rind - flavedo), and evaporate on the surface (Grierson and Ben-Yehoshua, 1986). The edible pulp, containing the juice vesicle, stalk and sac is sealed off from the ambient atmosphere by the peel (flavedo and albedo). Within the flesh (endocarp), the surfaces of the juice vesicle, stalk and sac have waxy cuticles. Because of this, vapour movement occurs primarily along the stalks and not between the vesicles. The vesicles are relatively impermeable and have a high osmotic pressure, so that the pulp retains its turgidity and juices long after the peel has become brittle and dry (Grierson and Ben-Yehoshua, 1986). Fruit size also affects water loss. The transpiration rate is greater for smaller fruit, which has a larger surface per area unit weight or volume (Grierson and Ben-Yehoshua, 1986). Fruit size also has a highly significant inverse relationship with the peel puncture resistance (PPR) before storage. Fucik (1981) found that the PPR is higher after 30 days storage at 21°C and 60% RH because of the leatherlike quality the peel assumes with desiccation. Some of the conditions and treatments that reduce water loss during storage are high RH, polyethylene emulsions, waxes and film wraps (Aljuburi and Huff, 1985). Water should never precipitate on the fruit during storage, whether due to inefficient humidifiers or to temperature fluctuations at very high humidity. In moisture-proof containers (e.g. wooden pallet boxes or plastic crates) humidity should be as high as possible. In non-moisture-proof containers (e.g. unwaxed carton) humidity should be as high as the containers can stand (Grierson and Ben-Yehoshua, 1986).

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#### **2.4.4. Oil glands and volatile compounds**

As rind breakdown is a disorder affecting the outer rind layer of citrus fruit, a study of the anatomy of this layer will help to achieve a better understanding of the development of disorders. The outer rind layer is coated with wax formulations, consisting mainly of esters (66%), alkanes, alkenes, and some alcohols. Essential oils of the terpenoid family are contained in specialized glands, called oil glands embedded mainly in the outer rind, but also in extremely small amounts in the juice vesicles (Monselise, 1986).

Monselise (1986) reported that a microscopic section through the peel would show a cuticle covered with extruded wax and a relatively small number of stomata (20-40 mm<sup>-2</sup>), sometimes occluded at maturity by a corky plug, followed by flavedo layers, consisting of relatively small cells. Multicellular, schizolysigenic oil glands (containing specific essential oils) and chromoplasts (containing drops of oil-dissolved carotenoids) are found in this layer. The albedo layers become progressively spongier centripetally and consist of deeply lobed cells with very large intercellular spaces and scattered vessels (Monselise, 1986). Holtzhausen (1975) also showed that in Valencia oranges the oil glands are located in the exocarp or flavedo, and that the oil cells enlarge from flowering until fruits are ripe at apparently the same rate, while new oil glands develop during the season and enlarge until maturity.

Phenolic compounds, under the action of associated enzymatic systems, are responsible for most of the browning reactions of fruit tissue (Santana et al., 1981). The most frequent injury in citrus is known as pitting, which takes the form of brown depressions in the flavedo. Flavedo tissue contains an abundant amount of phenolic compounds. The role played by these compounds and the related enzyme systems in chilling injury (CI) of citrus is under investigation and still not well known. Preliminary results indicate a relationship between CI and the behaviour of phenolic compounds in the flavedo during cold storage of 'Valencia' oranges at 6°C for 2.5 months (Santana et al., 1981).

#### **2.5. Storage treatments used to achieve quality fruit**

Quality of citrus fruit involves good taste, good appearance and must retain these qualities until it reaches the consumer. The quality of fruit, or the potential for quality, is often affected or determined long before harvest, and storage may be regarded as the preservation of a predetermined quality

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(Bangerth, 1979). The main requirements for citrus in long-term storage are the conservation of water/turgidity/juiciness and maintenance of flavour and market attractiveness (Curtis, 1988). The way fruit has to be stored greatly depends on its preharvest life, e.g. accumulation of calcium, sugars, acids, and cell size and number. Growth regulators and phytohormones affect most of these processes (Bangerth, 1979). Fruit quality is influenced by many factors apart from production practices, and many complex interrelationships exist among the various factors. To optimize fruit quality with cultural practices, it is necessary to know the specific quality factors preferred by the consumer, the specific fruit quality problems existing in the orchard in question, and the effects of cultural practices causing these potential problems (Reitz and Embleton, 1986). The development and application of the latest postharvest technology is vital to maintain the quality and freshness of citrus expected by the customers. Changing consumer demands and rapidly changing technology are influencing packinghouse operations (Tugwell, 1988).

Major factors that prevent citrus fruit from being marketed fresh are blemishes due to abiotic and biotic factors. The incidence of blemishes caused by biotic factors (insect, mite and fungus) is usually much greater in humid subtropical and tropical regions than in semi-arid or arid subtropical regions (Davies and Albrigo, 1994). Mould wastage and water loss are the most important factors in determining the postharvest life of fresh citrus fruits (Tugwell, 1988). The basic physiology of citrus fruits requires that fruit is protected from deterioration due to mould wastage and dehydration. Citrus rind is very easily damaged and injuries result in the development of wastage and increased moisture loss (Tugwell, 1988). Mishandling also very easily spoils the appearance of citrus rind. Rough handling of turgid fruit, excessive dehydration and exposure to low temperatures can rupture oil cells (Tugwell, 1988). Fruit should also not be harvested when the trees are wet or within a week of a severe drought stress having been relieved by rain or irrigation. Post-harvest rinses, fungicides and waxes should be applied as non-recovery sprays in packing line handling systems (Grierson and Ben-Yehoshua, 1986). The temperatures of hot air used for drying the fruit before packing without damaging the fruit depends on the frailty of the fruit. Tender speciality fruit, such as degreened tangerines and other soft citrus types, should be heated as little as possible and not brushed at all (Grierson et al., 1986). Wet fruit can withstand much higher air temperatures than dry fruit, however temperatures above 54°C are not recommended. It is possible to dry fruits with infrared rays. This is used as a predrying as it does not dry the ends of the fruit. In South Africa and Israel, the time in the dryer is slowed down through multiple layering (Grierson et al., 1986).

For distant shipping, viability of the rind is of paramount importance. It may be strengthened by on-tree treatments with growth regulators (mainly gibberellin) or by various postharvest treatments

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including growth regulators, fungistats, fungicides and a wax coat to reduce water loss. Polyethylene wraps can be used to increase shelf and shipping life (Monselise, 1986).

### 2.5.1. Uses and advantages of growth regulators.

Growth hormones have been used to improve postharvest fresh fruit quality of citrus since the 1920's when kerosene fumes were used to enhance peel color of lemons. Used on citrus as preharvest sprays or postharvest dips it can improve rind colour and firmness, delay fruit drop, decrease incidence of decay, improve fruit size and facilitate mechanical harvesting. Each growth regulator group elicits similar responses among cultivars, although the time and rate of the application varies (Davies, 1986). One of these treatments of citrus fruit is ethylene. Ethylene is used for ripening and colouring (degreening) of citrus in storage (Sinclair, 1984). An ethylene treatment results in a drastic increase of ABA (abscisic acid) in the citrus peel, which accelerates fruit senescence. Another growth hormone, 2,4-dichlorophenoxyacetic acid (2,4-D) can be used to reduce preharvest abscission and drop (Sinclair, 1984).

Most growth regulators have cosmetic effects on external characteristics important to the fresh fruit trade (i.e. fruit appearance, colour, size, rind texture) while having little influence on internal quality (Davies, 1986). Growth regulators are able to delay rind senescence and abscission processes (Gallasch, 1981). Gibberellic acid ( $GA_3$ ) is the most important of the gibberellins used commercially.  $GA_3$  is usually a growth promoter in its biological activity. Its main roles are shoot elongation, overcoming growth-retarding substances such as abscisic acid and ethylene, activating dormant seeds and buds, including parthenocarpy, timing of flowering and fruit-setting, delaying ripening and chlorophyll loss (senescence) in fruit. In citrus, preharvest sprays are used for the reduction of rind softening, staining, spotting and decay as well as delay in colouring (Sinclair, 1984). Combinations of gibberellic acid with 2,4-D may enhance peel strength and prolong the usable shipping season for grapefruit (Reitz and Embleton, 1986). Growth hormones can have the following effects on citrus fruit (Fucik, 1981; Davies, 1986):

- $GA_3$  reduced rind puffing and creasing and 2,4-D appears to make the peel less susceptible to rind disorders by delaying maturity.
- A combination of  $GA_3$  and 2,4-D reduced mold and rind blemishes more effectively than 2,4-D alone. Applications of 2,4,5-trichlorophenoxyacetic acid (2,4,5-T) and 2,4-D 1-6 months prior to harvest of 'Valencia' oranges increased fruit size and stem diameter as well as thicker rinds and subsequent lower juice content.

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- GA<sub>3</sub> applications had a pronounced improvement on rind quality of navel orange.
  - Applications of KGA (potassium gibberellate) increased the rind puncture force (RPF) or peel puncture resistance.
  - GA<sub>3</sub> sprays also reduced creasing and increased rind strength in 'Shamouti' oranges.
  - GA<sub>3</sub> applied prior to bloom increased fruit set and delayed colour development and maturity of 'Orlando' and 'Minneola' tangelos.

Gallasch (1981) found that 'Marsh' grapefruit fruit sprayed with GA was greener, had finer texture and thinner rinds and a greater resistance to rind rupture which indicated an improved marketability, because fruit treated with GA could resist damage better during handling than control fruits.

Tugwell et al. (1996) selected two locations with a history of albedo rind breakdown (creasing) to test the effective control by GA sprays. They found rapid increases in fruit size after the first 8 weeks. However, this increase in fruit size could cause thin peels and a low calcium content of the albedo, because cell division of the albedo tissue ceased after 8-9 weeks and the subsequent cell growth was because of cell expansion (Grierson, 1986). Spraying with 20 ppm GA when fruits were 30-50mm in diameter could delay this type of development and reduce the severity of creasing (Grierson, 1986).

The 'Minneola' tangelo has a short marketing season. Within 3 weeks of becoming palatable, rind separation occurs resulting in puffy fruit, which causes difficulties in handling (Tugwell et al., 1992). Tugwell et al. (1992) reported that a field application of 10 ppm GA applied with 10ppm 2,4-D prior to colour break in late March (South Australia) reduced rind separation and deterioration and extended the harvest season by 4 weeks. Fruit harvested during July up to mid-August could be stored for 6 weeks before the fruit became unacceptable due to lack of flavour (Tugwell et al., 1992). Chitzanidis et al. (1988) reported that a field application of only GA<sub>3</sub> (10ppm) applied before colour break, delayed colour development of the fruit and increased peel puncture resistance. Fruit size and rind thickness were not affected by the application of growth regulators. GA<sub>3</sub> had a beneficial effect on fruit quality by reducing penicillium decay and rind breakdown and by delaying rind softening (Chitzanidis et al., 1988). Fucik (1981) found that preharvest applications of 2,4-D and GA<sub>3</sub> to grapefruit in Texas reduced water loss of the fruit during postharvest storage (21°C, 60% RH for 30 days).

A GA<sub>3</sub> application to the entire tree of 'Satsuma' mandarin is able to prevent late peel growth, which takes place after cessation of pulp growth and retards the loss of juice from the ripe fruit, allowing on-tree storage of the fruit for more than two months after commercial ripening. It also prevents puffiness

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and retards pigment changes (Garcia-Luis et al., 1985). GA<sub>3</sub> can also be used as a control of physiological blemishes due to the fact that it delays peel senescence of several citrus species. Leakage of electrolytes, as judged by conductivity increase, has been shown to be a simple and useful experimental method for assessing damage to cellular membranes (Nolte et al., 1990). This is a possible way of assessing damage by RB as well, but needs to be studied before it can be recommended. GA<sub>3</sub> may have a beneficial impact on membrane integrity. Since a GA<sub>3</sub> concentration of 3 μM produces preservative effects on both the solute efflux capacity from intact tissue and the survival of protoplasts, it is apparent that the site of GA<sub>3</sub> action is in the symplast, possibly related to cellular membranes (Nolte et al., 1990).

The major contribution of growth hormones can be enhancing fruit appearance, size, and shipping quality (Davies, 1986). Growth regulators can also mitigate chilling injury (CI) and fumigation injury (Grierson and Ben-Yehoshua, 1986). However the response to these growth regulators is considerably modified by climatic and other influences, e.g. plot location in the orchard (Gallasch, 1981). The future of growth regulators is in their value in extending or shortening the harvest season, expanding markets for fresh fruit, and reducing crop losses due to preharvest drop. The two major problems limiting the use of growth regulators in citrus production remain the inconsistent performance due to environmental factors and difficulty in obtaining legal clearance to use new materials (Davies, 1986).

### **2.5.2. Applications before storage for better storage quality**

The storage of citrus is limited by susceptibility to cold injury. Seal packaging allows citrus to be stored at higher temperatures avoiding chilling and any wastage that develops is contained inside the wrap (Tugwell, 1988). Advantages of high-density polyethylene (HDPE) film wrapping are that it can extend storage life, reduce the need for refrigeration, lessen water loss and reduce CI. At high temperatures, off-flavours may occur with films inadequately permeable to O<sub>2</sub> and CO<sub>2</sub> (Grierson and Ben-Yehoshua, 1986). Most citrus fruit is waxed commercially and not sealed because of financial implications (Grierson and Ben-Yehoshua, 1986).

Chun et al. (1990) found that fruit quality depended on whether the fruit was wrapped or wax coated and on the use of TBZ after treating commercially cleaned and chlorine-treated (150ppm) fruit with tiabendazole (TBZ,1000ppm), Deccossan 315 (a quaternary ammonium compound, 500ppm) and a coat of wax or wrapped individually in film. The fruit was stored for 3 weeks at 4.4°C, 90% RH and then held at 21°C, 95%RH for 2 weeks (Chun et al., 1990). Biological safe sucrose based coating is

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also becoming increasingly popular commercially, because of its flavour preserving properties in citrus (Curtis, 1988). It is a stable white powder (Semperfresh (TM)), and is mixed with water in concentrations between 0.75% and 2%, which is used as a dip, or as a spray to cover fruit and vegetables. The liquid dries as an invisible, odourless, tasteless, biodegradable, completely non-toxic coating which envelops the fruit as a differentially permeable membrane. It has a hygroscopic nature that may hold a thin film of water. There might be a problem with gas exchange as it restricts O<sub>2</sub> but not CO<sub>2</sub> (Curtis, 1988).

Ester coatings are compatible with most conventional fungicides for controlling storage rots (Curtis, 1988). There is a reduction of water loss compared to untreated fruit, but it is less than with conventional waxes. Fruit remains more turgid and juicy than untreated fruit. Ester coatings can preserve fruit flavour better than that of untreated and commercially treated (waxed) fruit. Although it does not have the high shine of wax coatings, combinations of wax and ester will probably satisfy the requirements of market attractiveness, without the loss of flavour. It can prolong storage life by weeks (Curtis, 1988). Pro-long (a permeable sucrose ester coating) can also extend the shelf life of limes at high RH and low temperatures by controlling weight loss and degreening (Motlagh and Quantick, 1988).

### **2.5.2.1 Wax applications**

Waxing of oranges and grapefruit stored at 21-31°C and at 71-96% RH minimized weight loss. A coating with Shield Brite AP 40 wax, applied singly or in combination with fungicides (0.1 % benomyl or 0.5% sodium dehydroacetate) increases the marketable life and enhances the appearance without adversely affecting the chemical quality attributes (TSS, Total acidity and pH) of the fruit (Aworh et al., 1991). Waxing also improves the lustre of oranges and grapefruit by an attractive glossy finish of the treated fruits. Untreated fruit develops a dull wrinkled appearance and hardened rinds by the end of the third week of storage at 21-31°C and at 71-96% RH. By reducing moisture loss, waxing minimizes shriveling, thus contributing to the better appearance of waxed fruits relative to controls (Aworh et al., 1991).

Waxing 'Murcott' tangerines leads to increased internal CO<sub>2</sub>, and the consequent off-flavor (Cohen et al., 1990). Commercial spray coatings and experimental formulations of wax with 15% solid matter were found to have greater transpiration and gas exchange, probably because there was less wax applied. This resulted in greater water loss but less flavor deterioration. Increases in ethanol seemed to

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adversely affect the taste, while the role of acetaldehyde was not clear. It is probable that acetaldehyde improves flavour (Cohen et al., 1990). Water-type waxes are only effective in lowering moisture loss at the end of a 4-week cold storage and in the later marketing period. Coating reduced the incidence of physiological disorders in 'Valencias' but not in Navels. The water-type waxes on 'Valencias' were more effective than solvent-type wax (Cuquerella et al., 1981).

Citrus fruit for export to distant markets needs to have an excellent shine, its weight loss reduced to a minimum, and organoleptic quality must remain unchanged during a period of more than 30 days. Fruit waxing is of great importance to achieve these aims (Namesny and Decoud, 1988). The natural wax coating of citrus is deposited in lightly attached irregular platelets, which is largely removed by washing and highly alkaline fungicidal treatments. It is therefore necessary to replace it with an artificial coating (Grierson and Ben-Yehoshua, 1986). The introduction of polyethylene into wax formulations results in greater storage life without deleterious effects. Major effects are the reduction of weight loss by up to 60%, inhibition of shrinkage and a shiny gloss of the fruit. In anything but ultra-high humidity storage, peels of unwaxed, washed citrus fruit tend to dry to a thin, hard, unsightly shell, thereby making the fruit unmarketable. For usual storage conditions (<90% RH), prior waxing is considered essential. The effects of waxing are particularly contradictory for chilling-susceptible species. Waxing decreases susceptibility to chilling in grapefruit, 'Temple' and 'Tangor', but increases susceptibility to chilling in limes (Grierson, and Ben-Yehoshua, 1986).

#### **2.5.2.2 Oil-emulsions**

Aljuburi and Huff (1985) found that immersing grapefruit in vegetable oils and vegetable oil-water emulsions prior to storage at 3°C markedly delayed and reduced symptoms of chilling injury. Oil-water emulsions of 20% oil were more effective than higher percentage oil emulsions or 100% oil. CI appears to involve loss of water at low temperatures. Steps must be taken to avoid water loss to reduce the symptoms of CI. Water-oil emulsions can reduce the amount of pitting of peels by delaying the time to which 10% of the fruit was unmarketable by 90 days. The surface oil is absorbed by the fruit and does not have an oily, unappealing surface. Vegetable oils are an effective cheaper alternative to polyethylene films and require less capital outlay (Aljuburi and Huff, 1985).

#### **2.5.2.3 Individually sealed fruit**

Weight loss due to transpiration is a serious factor limiting the post harvest life of citrus (Kawada and Kitagawa, 1988). Individual sealing of citrus fruit with heat shrinkable polyethylene (PE) film can

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beneficially affect the post-harvest quality of the fruit by reducing moisture loss and subsequent fruit deformation (Barmore et al., 1983). The individual seal packaging of fruits and vegetables in high-density polyethylene (HDPE) film can also delay deterioration markedly. The mode of action of seal packaging was investigated by Ben-Yehoshua et al. (1981) by studying several physiological parameters of citrus fruit and bell peppers. HDPE seal-packaging effects were related to a water saturated micro-atmosphere in the sealed enclosure around the fruit. This saturated atmosphere alleviated water stress, which existed in harvested fruit as a result of its severance from the water supply of the mother plant. It had quite a few effects on firmness, weight loss and water deficit. The individual sealing of lemons in HDPE film markedly delayed the softening and shrinkage of the fruit as determined by residual deformation (in mm) and percentage weight loss. Ben-Yehoshua et al. (1981) discovered the following effects on membrane integrity: Sealing markedly inhibited both the increase of conductivity and the leakage of free amino acids. Apparently, sealing delayed the senescence of live tissue. The results could be explained by the theory that the major effect of sealing is through the provision of a water-saturated ambient atmosphere

Plastic film wrapping of 'Minneola' tangelos can improve the colour in storage to a deep or more reddish-orange colour. It also reduced decay especially in the 21°C holding time (Chun et al., 1990). According to Kawada and Kitagawa (1988) and Kawada et al. (1981) the benefits of individual wrapping of unwaxed fruit are:

- minimization of weight loss which in turn decreases softening, deformation and rind injury,
- prevention of decay problems such as cross infection, so called 'soilage' (blemishing of sound fruit by mould spores) and making it easy to discard decaying fruit,
- extension of the marketing season,
- reduced transpiration rate,
- prevention of CI,
- no increase in in-fruit CO<sub>2</sub> concentration,
- reduced ethylene production,
- no increase juice volatiles content.

Factors involved in modification of in-package atmospheres in polymeric film packaging of fruits and vegetables are (Kawada and Kitagawa, 1988; Kawada et al., 1981):

- Permeability of film (type of film, thickness of film, surface area of film),
- Respiration and gas exchange system of the commodity (type of commodity, maturity of commodity, size and amount of commodity in package),



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- Other (initial free air volume in a package, initial in-package atmospheres, external conditions (Temperature, humidity, partial pressure of gas),
  - Film packaging should be considered as a supplement to proper refrigeration.

Film wrapping on loose skin citrus types are not beneficial, because it causes peel puffing and thus increases decay (Kawada et al., 1981). CI of 'Valencia' and 'Navel' oranges can be prevented by sealing and by waxing. Wrapping produce in plastic film reduces the rate of postharvest weight loss. The retention of moisture prolongs the onset of visible symptoms of senescence, such as shriveling, wilting, aging or pulp softening and aids in retaining of a fresh appearance (Miller and Risse, 1988).

Recommended storage and transit temperatures for citrus have traditionally been a compromise between temperatures low enough to reduce physiological deterioration and high enough to avoid development of rind disorders due to chilling injury. With sealing in HDPE it is possible to reduce weight loss and deterioration of quality sufficiently to allow storage at temperatures above the critical level for CI (Tugwell and Gillespie, 1981). Plastic wrapped 'Minneola' tangelos can be stored for 4 weeks at 20°C before significant loss of flavour occurs. No rind blemish develops on wrapped 'Minneola' tangelo fruit stored for up to 8 weeks at 20°C (Tugwell et al., 1992).

The rapid loss of acidity from early harvested mandarins and grapefruit wrapped in HDPE film and held at ambient temperature resulted in the fruit becoming more palatable during storage. Citrus fruit intended for long term storage in HDPE wraps should be harvested early to midseason or treated with gibberellic acid to delay maturity and maximize storage life (Tugwell and Gillespie, 1981).

### **2.5.3. Storage temperatures used as postharvest treatment**

Storage temperatures have a number of different influences on the storability of citrus fruit. Extensive research has been done to determine the optimum storage temperatures for each different citrus cultivar. It is difficult to make specific recommendations on postharvest handling and storage conditions to all citrus producing areas. This is because storage conditions for one area may be undesirable for other areas. The demands of the fruit also change during the season. Early and mid-season fruit require different storage and handling to late-season fruit. Citrus can usually be stored for 2 to 3 months after attaining an acceptable eating maturity, without losing quality. Any recommendation is at best an approximation (Grierson and Ben-Yehoshua, 1986). The response of citrus fruit to temperature is much less dramatic than that of climacteric fruit such as apples or

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bananas. The rate of postharvest respiration of citrus fruit is low and is directly proportional to temperatures in the range of 10-35°C. Too high temperatures can cause fungal decay, rapid water loss, increased softening and an enhanced decrease in ascorbic acid (Vit.C) content (Grierson and Ben-Yehoshua, 1986). Any treatment such as precooling, waxing or film wrapping that eliminates the need for expensive refrigeration is of obvious economic value. The factors that must be taken into account for optimal storage and shipping temperatures are susceptibility to CI, storage period required, development of decay at higher temperatures, effect of waxing or seal-packaging, preharvest treatments, time of harvest, prestorage cooling and cold sterilization for fruit fly eradication (Grierson and Ben-Yehoshua, 1986).

Low temperatures during storage and transit depress fruit respiration, water loss and growth of decay organisms, but can also cause CI (Davies and Albrigo, 1994; Chitzanidis, 1986; Grierson and Ben-Yehoshua, 1986). Storage temperatures below 5°C can also induce off-flavours (Grierson and Ben-Yehoshua, 1986). Grapefruit stored at temperatures below 13°C becomes pitted and discoloured (Aljuburi and Huff, 1985).

Sweet oranges and mandarins can be stored for 2 months or more at 0-4°C with very little loss in fruit quality. Limes, lemons and grapefruit cannot be stored at temperatures less than 10°C because they develop CI, which is manifested as pitted necrotic regions in the peel. The susceptibility to CI varies seasonally, and may be related to abscisic acid (ABA), proline, reducing sugar content, squalene content in the peel and water loss from the peel. Holding fruit for 1 week at moderately cool temperature (10-16°C) reduces the subsequent injury at temperatures as low as 1°C. Relative humidity should be maintained as high as possible, usually between 85 and 95% to retard water loss due to vapour pressure gradients between the fruit and the air. High humidity also promotes wound healing and growth of some decay organisms such as *Diplodia* stem-end rot (Davies and Albrigo, 1994). The optimum storage conditions for 'Sunburst' fruit based on rind breakdown or CI and the extent of decay after storage, are 4°C for 4 weeks (Hatton et al., 1986). This temperature conforms to the storage temperature of most mandarin-type citrus fruits. Storage characteristics were not influenced by 4 different rootstocks, namely sour orange, rough lemon, 'Cleopatra' mandarin and 'Carizzo' citrange (Hatton et al., 1986).

El-Zeftawi et al. (1989) found that when storing 'Valencia' oranges at relatively warm temperatures (15°C) for 12 weeks, they exhibited minimal disorders, particularly pitting, but pitting increased when stored at 5°C for 18 weeks. Also, varying the temperature could maximize the length of storage

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without increasing fruit wastage. More storage disorders in Newton Late Valencias seemed to be related to the 'Symons' sweet orange rootstock than to 'Rangpur Lime' or 'Emperor' mandarin rootstocks. They also found that all the dip treatments used in the experiment (benomyl at 750 ppm and GA<sub>3</sub> at 500 ppm, benomyl alone or in combination with 2,4-D at 500 ppm), reduced the incidence of disorders. Pitting increased with time depending on storage temperature. Fruit warming is an effective way to reduce CI at 5°C for 9 weeks and 9 weeks at 15°C. (El-Zeftawi et al., 1989)

Martinez-Javega et al., (1992) found that storing 'Fortune' mandarins at 2°C or 5°C caused CI and increased both ethanol and acetaldehyde levels. The CI can assume various forms - pitting, scald or oil gland darkening. CI can be avoided in citrus fruit if they are returned to a warming temperature before degenerative changes occur. Fruit stored at temperatures lower than 10°C presents a high incidence of CI. Over-mature fruit (stored at 56 days cold storage and 2 days at 20°C) shows less susceptibility to CI, but is more inclined to develop stem-end rind breakdown. Fruit quality can be maintained during storage for 8 weeks at 10°C if individual film wrap is applied as it reduces moisture loss and delays changes due to senescence. The same results can be obtained with individual film wrapped mandarins at 5°C with intermittent warming of 16 hours/week at 20°C (Martinez-Javega et al., 1992).

Salcines (1981) found that the optimum storage temperature for degreened grapefruit is 14°C for 12 weeks, and 12°C for non-degreened fruit. The total losses were the highest for fruit picked towards the end of the harvest season. When Schirra and Chessa (1988) stored 'Tarocco' oranges the respiration rate increased at shelf-life temperature after chilling-temperature storage. An abnormal accumulation of juice volatiles (ethanol and acetaldehyde) was observed in fruit stored below 6°C. CI symptoms appeared after 1 month, progressively increasing to become the most accentuated after eight weeks of storage. The most severe alterations were at 4°C, then at 2°C, but at 6 and 8°C it was much less, and could be considered negligible at 12°C (Schirra and Chessa, 1988).

#### **2.5.4. Curing at high or low temperatures during storage period**

In South Africa application of wax on citrus fruit is with a hot dip treatment (Van Wyk, 1996). When the water temperature is higher than 40°C, it may lead to increased rind breakdown in 'Minneola' fruit (personal communication: Van Wyk, 1996). Cohen et al. (1994) reported that heat treatments, such as curing at 34-36°C for 48-72 hours or hot dips, presented a promising non-fungicide means to control postharvest decay of citrus fruits and to reduce their sensitivity to suboptimal storage temperatures. Hot water caused a few fruit surface changes. A SEM examination of grapefruit peel revealed the

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presence of cuticular micro-cracks on the surface of non-treated fruit or of the fruit dipped in water at ambient temperature (18°C) (Cohen et al., 1994). Fruit dipped in hot water (52°C) demonstrated morphological changes of the fruit surface. It had no cuticular cracks and looked smoother than that of the fruit not treated with heat. These changes might be related to the effect of heat on the epicuticular wax layer on the fruit surface. Phytotoxicity problems were mostly observed with a continuous flow installation. It could possibly be explained by an increased sensitivity of the heated fruit to mechanical impact; damage to the natural fruit surface coating caused by rolling or brushing of the hot fruit or by sharp temperature contrast when the heated fruit is sprayed with ambient-temperature water or solution; and variability of heat-durations when part of the fruit is over-heated and another part is under-heated. The repair of cuticular cracks could also be related to alleviation of citrus chilling injury (CI) by hot water dips, since CI development was shown to be accompanied by cracking of the fruit surface (Cohen et al., 1994). These changes in the fruit surface might explain the hot water effects on citrus fruit, being the reduction of weight loss, inhibition of softening and alleviation of chilling injury (Rodov et al., 1996; Del Rio et al., 1992).

Schirra and D'Hallewin (1996) also reported that a hot water treatment (dip) of 52°C for 3 minutes reduced CI significantly. The adding of Semperfresh FLO (SPF) (a sucrose polyester edible coating) slightly increased the incidence of CI. SEM observations revealed that hot water dipping caused an apparent fusion and a redistribution of the epicuticular wax layer with consequent elimination of the discontinuity zones (cracks) normally present in ripe fruit. This action might have prevented pathogens from penetrating and might be involved in resistance to chilling injury. Hot dipping produced a surface action, not interfering substantially with fruit physiological behaviour or influencing internal quality attributes. SPF treatment caused a slight increase in respiration rate during cold storage followed by a significant decrease at the end of a simulated shelf life. Titratable acidity and TSS of the juice were not influenced by the prestorage dip treatments (Del Rio et al., 1992; Schirra and D'Hallewin, 1996). This 3 minute hot water dip treatment at 52°C allowed the storage of 'Fortune' mandarins in good condition for 6 weeks at 5 to 6°C plus three days at 20°C, with a good level of acidity, a balanced taste and minimal losses due to pathogens or CI. The effect of edible coatings appeared not to be more beneficial than the use of water alone (Schirra and D'Hallewin, 1996).

Curing of sealed lemons of normal and decay-prone types and of sealed 'Goliath' pummelo inhibited postharvest decay without deleterious effects on fruit quality and prevented the development of *Penicillium digitatum* on inoculated fruit (Ben-Yehoshua et al., 1987). Curing of non-sealed fruit was

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less effective in reducing decay than curing of sealed fruit and caused prohibitive weight loss, shrinkage and softening. Curing of sealed and waxed 'Shamouti' and 'Valencia' oranges, in comparison to sealed fruit, resulted in some CO<sub>2</sub> injury of the peel and off-flavours. Temperatures used for curing were 25 to 42°C for 1-3 days. After curing, fruit were stored at 17°C or at the optimum temperature of 13°C for lemons, 11°C for pummelos, and 6°C for 'Shamouti' oranges (Ben-Yehoshua et al., 1987).

Seal packaging and curing double and even treble the life of pummelo fruit (*Citrus grandis*). Curing for 48-72 hours at 34-35°C, later than 48 hours after harvest, is effective in reducing decay without causing damage. Fruit held at 11°C developed less decay than fruit held at 17°C, but the differences between the storage temperatures decreased after effective curing. Sealing in plastic film is essential for the curing, in order to provide a water-saturated atmosphere, to prevent weight loss and shrinkage and to protect from damage incurred by high temperatures. During the 4-6 months of storage, weight loss of cured and sealed fruit was practically prevented and the quality and flavour of fruit was maintained. Decay even after 9 months of storage at 11°C was only 9.5%. It seems therefore that seal packaging and curing make it possible for pummelo fruit to be stored without any chemical treatment. Success of seal packaging becomes more meaningful when considering the sensitivity of pummelo to damage by waxing (Ben-Yehoshua et al., 1988).

Wright et al. (1996) used fruit harvested and handled normally by the packinghouse and delivered the next day. The fruit were treated with saturated vapour heat at 48°C for 30, 60 and 120 minutes and 38°C for 24 and 48 hours. Subsequently the fruit were stored at 0°C for 28 days. Assessments were made for incidence of CI, changes in TSS acidity and percentage juice. Following a further 14 days at room temperature (22°C) the same assessments were made. CI was visually rated as the number of water soaked lesions per fruit (Wright et al., 1996). The vapour heat treatment of 48°C after 0°C storage, reduced CI significantly. Acid percentages declined over time - the longer the treatment time the lower the acid percentage. This could be the result of metabolism of organic acids during the prestorage time. Heat treatments at 48°C and 38°C had significant effects on percentage juice. TSS and percentage acid declined slightly in heat-treated fruit, but in some instances where levels were marginally high, a decrease in acidity might be beneficial. Higher temperatures (48°C) for shorter periods (2 to 3 hours) were more effective than lower temperatures (38°C) for longer time periods (up to 24 hours) (Wright et al., 1996).

High temperatures (heat stress) induce an adaptive response in fruits to chilling injury. Chilling temperatures (below 5°C) degrade membrane lipids and cause a change or transition in the molecular

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ordering of membrane lipids. This decreases flexibility, creating a brittle, inflexible membrane (Wright et al., 1996). Chun et al. (1988) used individually plastic-film-wrapped or waxed 'Marsh' grapefruit conditioned (34°C, 3 days) at low (30% ± 5% RH) or high (95% ± 5% RH) humidities before cold storage (10°± 1°C; 90% ± 5% RH). Fruit quality tended to be adversely affected by conditioning (first two weeks in storage), but this effect became less obvious with storage at 21°C. Fruit coated with plastic film had a better general appearance than waxed grapefruit. Conditioning at either high or low humidity reduced the incidence of penicillium rot (Chun et al., 1988). Tugwell et al. (1992) found that 'Minneola' tangelo fruit could be successfully cold treated for 16 days at 1°C followed by storage at 12°C for 8 weeks with no significant development of cold induced rind blemish. Curing is advantageous for chilling-susceptible species such as grapefruit (Grierson and Ben-Yehoshua, 1986) because it has no deleterious effects on the quality indicators of grapefruit (Del Rio et al., 1992).

Kawada and Kitagawa (1992) found that prestorage curing (3-5 days, ambient temperature) and conditioning (gradual decreasing of storage temperature) were necessary to control injury and prolong storage life. Wild and Hood (1989) were able to reduce CI in 'Valencia' oranges when stored for 15 weeks at 1°C after immersion in hot dips of water or water suspensions thiabendazole (TBZ) (1000mg/l). Pretreatment rind injury inflicted to the fruit before cold storage, only slightly increased CI incidence. The best results were obtained when the water was at 53°C, and the fruit stored at 1°C, because the water treatment had a curing effect on the fruit (Wild and Hood, 1989).

A low temperature storage of 0°C for citrus fruit is a disinfestation requirement for several export markets. According to Wright et al. (1996) all citrus fruit will develop CI when stored at temperatures below 10°C. This is contrary to the findings of Van Rensburg et al. (1995) when 'Clementines' and 'Minneolas' could successfully be stored at -0.5°C without CI. They reported that this could be because the fruit is 'preserved' at -0.5°C and most metabolic processes were effectively suppressed, or even ceased. This low temperature treatment is also used for in-transit sterilization of fruit destined for Japan. Fruit is kept at -0.5 °C for 12 days during shipping for sterilization for Mediterranean fruit fly (Anonymous, 1995). A cold treatment of 16 days at 1°C is required for disinfestation of citrus against Queensland and Mediterranean fruit flies. Tugwell et al. (1992) found that a treatment of 1°C for sixteen days caused a small non-significant rise in rind blemishes on 'Washington Navel' oranges and 'Lisbon' lemons, but the same treatment for 32 days however significantly increased rind blemish. This only became evident 4 weeks after cold treatment. Cold sterilization is used as an alternative to Ethylene dibromide (EDB) treatments (Tugwell, 1992).

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### 2.5.5. Controlled atmosphere in storage chambers

Unlike other fruit such as apples, persimmons and tomatoes, citrus fruit do not respond much to controlled or modified atmospheres (Kawada et al., 1981) as high CO<sub>2</sub> concentration decreases respiration only in climacteric fruits (Kubo et al., 1990). Rind breakdown (ageing) is increased under controlled atmosphere (CA) conditions, but cannot be controlled by the addition of either 2.5 or 5% CO<sub>2</sub> (Chase, 1969). Grierson and Ben-Yehoshua (1986) reported that CA could possibly be used to control the CI syndrome. Applications of CO<sub>2</sub> either as pretreatment at a high dose or lower concentrations throughout the storage have reduced CI of grapefruit. High concentrations of CO<sub>2</sub> during CA storage reduce incidence of pitting in lemons. With CA storage, removal of ethylene is critical for lemons and oranges (Grierson and Ben-Yehoshua, 1986).

Treatments with low O<sub>2</sub> and high CO<sub>2</sub> on 'Valencia' fruit cause an increase in ethanol and acetaldehyde content. This is correlated with a decrease in flavour. Ethanol and acetaldehyde are the products of anaerobic respiration (Ke and Kader, 1990). 'Valencias' stored at 6°C for 5 months store better in ethylene free conditions. A catalytic scrubber or another air purification system can be used to destroy ethylene and other volatiles in the air. Ethylene concentrations should be kept at 0.1 - 1.0 ppm to reduce off-flavours typical of fruit stored for a long time (Testoni et al., 1992; Grierson and Ben-Yehoshua, 1986; Pesis and Avissar, 1988).

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### 3. Influence of storage temperatures on rind breakdown and quality parameters in 'Minneola' tangelos

#### 3.1. Introduction

Citrus in Southern Africa is produced in four production areas determined by climatological factors (Anonymous, 1995):

- WARM areas in Mozambique, Zimbabwe, Most of the Northern Province and low lying areas of Mpumalanga, Swaziland and Kwazulu/Natal;
- INTERMEDIATE areas in the higher areas of Northern Province and Mpumalanga with the lower areas of “central Transvaal”;
- COOL areas in the North West and higher areas of “central Transvaal” and Kwazulu/Natal;
- COLD areas in the Eastern and Western Cape.

The 'Minneola' tangelo is classified as a soft citrus type (Anonymous, 1995) and although soft citrus is supposed to be produced mainly in the cool and cold areas of the country, it is grown in all four production areas.

At present it is not certain exactly what causes rind breakdown in 'Minneolas' or if it only occurs in fruit from a specific area. The ideal situation would be to use fruit from all the production areas in an experiment, but due to the number of fruit that would have to be analyzed it was more practical to use fruit from two different production areas representing two climatic regions in Southern Africa.

Although moisture and temperature are not the only causes for development of rind breakdown, a study that highlights some of these factors would be a positive contribution to finding the cause of rind breakdown. According to Tugwell et al. (1992) 'Minneola' fruit should be stored at temperatures not less than 12°C to avoid chilling induced rind blemish. They found rind blemish to be the most severe when fruit was stored for more than 4 weeks at 10°C. Van Rensburg (1997, personal communication) found a higher occurrence of RB when shipping temperatures were higher and storage temperatures at 4.5°C or 11°C, studying a temperature range of – 0.5 to 11°C for 3 weeks for shipping of the fruit. 'Star Ruby' grapefruit developed chilling injury (CI) (pitting of the rind) if stored continuously at 4.5°C for longer than 10 weeks (Du Toit Pelser and Lesar, 1994). For marketing reasons, fruit need to be stored for 16-18 weeks. No CI occurred when fruit was stored for 1 week at 20°C, 4 weeks at 4.5°C (shipping), 12 weeks at 8°C (overseas storage), and 1 week at 20°C (selling) (Du Toit Pelser and

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Lesar, 1994). No CI also developed when the fruit was stored in the following ranges: a) 1 week at 20°C, 8 weeks at 4.5°C (shipping), 8 weeks at 8°C (overseas storage), 1 week at 20°C (selling), or b) 1 week at 20°C, 4 weeks at 11°C (shipping temperature when colour is poor), 8 weeks at 4.5°C and 4 weeks at 11°C (overseas storage) and 1 week at 20°C (selling) (Du Toit Pelsler and Lesar, 1994). A major factor in the postharvest deterioration of citrus fruit is weight loss due to transpiration causing desiccation, shriveling, accelerated softening, loss of the attractive appearance of the fruit and accelerating senescence (Grierson and Ben-Yehoshua, 1986). Loss of water vapour is due to the vapour pressure gradient between the peel and the air surrounding the fruit. Control of RH and temperature is critical to reduce weight loss. The RH in storage should be as high as possible to prevent commercially unacceptable weight loss without causing damage to the containers (Grierson and Ben-Yehoshua, 1986).

'Minneola' fruit produced in Southern African production areas is stored after sorting, grading and packing, in cold storage at 11°C for a cooling period for approximately 4 weeks. During this period the fruit is shipped to the overseas storage rooms. On arrival, the fruit is stored for  $\pm$  4 weeks at 4.5°C and then  $\pm$  2 weeks at 8°C. The relative humidity is kept between 80 to 85% with a minimum of 75% and a maximum of 95%. The atmosphere is monitored for CO<sub>2</sub> increase and a maximum of 0.5% is permitted. These storing parameters are used for most soft citrus types (Van Wyk, 1996). According to Netteville (1995) the total time protocol for 'Minneolas' from packing to marketing at retail level is 7 weeks. The maturity requirements of citrus are based on juice content, soluble solids content, titratable acidity, and the ratio of the two (Kader and Arpaia, 1992). The total soluble solid (TSS) content normally increases as the period of storage is prolonged, while acidity and ascorbic acid content decreases (Lal, 1985).

This study was conducted to evaluate the effect of various storage temperatures on moisture content and physiological properties of the fruit after harvest, and the eventual effect on the incidence of rind breakdown. Furthermore, rind breakdown was correlated with the physiological parameters to determine, if any of the physiological properties of the fruit could be used as a possible warning system for the development of rind breakdown later in storage.

### **3.2. Materials and methods**

Fruit from the cold (Western Cape) and warm areas (Swaziland) were analysed in 1996, and in 1997 fruit only from the cold areas (Western and the Eastern Cape) were analysed. Production areas were

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changed between the two years because incidences of rind breakdown developed in the storage trial in 1996 were not significant and rind breakdown on marketed fruit in 1996 was reported mainly from the producers from the Eastern Cape (personal communication: Van Wyk, 1996). In 1996 fruit was obtained from two growers of the Western Cape (Franschoek and Goedehoop) and one grower in Swaziland (Ngonini). Normally treated and packed fruit by packinghouses were used for storage temperature trials. The fruit had been commercially washed and waxed before storage (Wright et al., 1996; Albrigo et al., 1981). In 1996 each grower provided 6 cartons of fruit, which were divided in 2 treatments with 3 replications (one carton being a replication). For each storage regime there were 3 replications (3 cartons) and for each sampling period (every second week) 5 fruit from each replication was picked randomly (5 fruit/replicate/sampling time). In 1997 the grower from the Western Cape supplied 12 cartons of fruit, which were divided in 4 treatments with 3 replications and the growers from the Eastern Cape supplied 6 cartons of fruit, which was divided in 2 treatments with 3 replications. For each storage regime there were 3 replications (3 cartons) and for each sampling period (every second week) 3 fruit from each replication was picked randomly (3 fruit/replicate/sampling time). The sampling size was lowered in 1997 to allow time to do the sampling and analysis on one day. In both years the fruit were first subjected to 4 weeks simulated shipping conditions followed by 6 weeks of storage and a period of 2 weeks at room temperature simulating the marketing period (Wenda and Kelly, 1987). The shipping / storage time accounted for 12 weeks in 1996 and 14 weeks in 1997. The treatments applied are listed in Table 3.2.1.

Table 3.2.1. Temperature treatments applied in 1996 and 1997

1996	Treatment	Shipping	Storage	Marketing period
	A	4 weeks at 11°C	4 weeks at 4.5°C, 2 weeks at 8°C	2 weeks at ambient temperature
	B	4 weeks at 11°C	6 weeks at 8°C	2 weeks at ambient temperature
1997	A	4 weeks at 4.5°C (E-Cape @ 11°C)	6 weeks at 11°C	4 weeks at ambient temperature
	B	4 weeks at 11°C	6 weeks at 4.5°C	4 weeks at ambient temperature
	C	4 weeks at 4.5°C	6 weeks at 4.5°C	4 weeks at ambient temperature
	D	4 weeks at 11°C	6 weeks at 11°C	4 weeks at ambient temperature

In 1997 fruit was obtained from one grower from the Western Cape (Goedehoop) and from two growers from the Eastern Cape (Gamtoosrivier and Sondagsrivier). The treatments in 1997 were also altered, and concentrated more on the difference between shipping and storage temperatures. The fruit

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from the Eastern Cape (Gamtoosrivier and Sondagsrivier) was received after the treatments started, and was stored at 11°C for the first 4 weeks at the packinghouse, influencing only treatment A. As analysis of this fruit only started at week 6 in storage, data for this period cannot be shown. There were also just enough fruit for 2 treatments (A and B), so only the fruit from the Western Cape was stored at treatment C and D.

Ideal RH conditions of 85-90 % could not be maintained (1996 and 1997) due to technical reasons in the 11 and 8°C storage rooms. However, a RH of 70% could be achieved by placing bowls of water in the storage room. At 4.5°C, the RH was maintained at ±90%. To eliminate the effect of fruit size, the cartons were mixed first and fruit size was measured at sampling.

Determinations of percentage juice, TSS, acid concentrations and maturity scaling were conducted according to procedures used by OUTSPAN INTERNATIONAL LTD. for determining internal quality for export fruit (Van Wyk, 1996). Fruit infested with pathogens was removed from the cartons to prevent further infestation. The storage time of each producer started at a different week to enable determinations of each production area on the same day. Samples were taken every second week and fruit quality was determined according to the methods described by Gorini & Testoni (1988), Tugwell et al. (1992), Van Wyk (1996). After visual evaluations, the fruit was weighed. For fruit size determinations, radial and axial measurements were taken with a caliper. 'Minneola' fruit can have a prominent 'neck' that influences fruit shape and size. After cutting the fruit radially, the rind thickness was measured on three randomly chosen places. The mean rind thickness was calculated to give a more representable value for rind thickness. The percentage juice was determined after determining the juice mass. This was done by cutting the fruit in half and extracting the juice with a rotational juice extractor. The juice was then forced through a piece of cheesecloth. The remaining pulp was weighed with the empty peels giving a value for the total solid material. The percentage juice can be calculated as follows:  $(\text{Total fruit weight} - \text{Total solid material}) / \text{Total fruit weight} \times 100$ . The total soluble solids in the juice were determined with an automatic light refractometer (Euromax, Health Care Apparatus, Holland). The percentage acid in the fruit juice was determined by titration with 0.1562N NaOH using a titrator (DL 25, Mettler Toledo, Switzerland). The ratio between the TSS and % acid in the fruit was calculated since it indicates the amount of sugars present in the juice, while the TSS measurements represent the amount of carbohydrates, including both sugars and acids (Davies and Albrigo, 1994).

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The amount of rind breakdown marks on the fruit was counted and scaled in categories of 0 to 3 marks (category 1), more than 3 (category 2) and more than 10 marks per fruit (unmarketable) (Wright et al., 1996). The categories used were in conjunction with the blemishes allowed for overseas and local marketing. Fruit from category 1 was still suitable for overseas distribution but not as first grade fruit. Fruit from category 2 was suitable for local distribution only. Maturity ratings were done because soft citrus fruit types tend to develop signs of over-maturity when kept in cold storage for long periods. It was determined by using the Colour prints for Blemish and Appearance Standards (Set No. 46, OUTSPAN International, South Africa). The percentage weight loss was determined every second week by placing 5 fruit in a net bag and keeping the bags in separate cartons in the treatments and replications (El-Nabawy et al., 1981). Decayed fruit was removed from the net bags to prevent further infestation. The mass of the decayed fruit was subtracted from the total of the bag and the mean weight per fruit was determined for the amount of fruit left in the bag. Peel moisture was determined in 1997 by calculating the percentage moisture of the peel after drying the peel for three days in an oven at 100°C using the following equation:  $(\text{Dry peel weight} / \text{Total solid material}) \times 100$ .

Data were analysed by the Dept. of Agrimetrics (ARC, Pretoria, South Africa) using the statistical program GENSTAT 5 (GENSTAT 5 Committee, 1998). Both trials were analysed as completely randomised designs to test for differences between treatments. The data were normally distributed with homogenous treatment variances. Treatment means were separated using Fishers' protected t-test least significant difference (LSD) at the 5 % level of significance (Snedecor and Cochran, 1980). A correlation matrix between physiological parameters and the occurrence of RB was drawn up for both years and all the parameters measured. The correlation matrix was done with Genstat 5 at  $df=16$ .

### **3.3. Results and discussion**

As there was no difference between fruit weight and size in response to the different treatments, the results will not be discussed. These parameters were only used in the correlation matrix (See 3.3.5). Fruit weight and size were strongly dependent upon the producer. The distinction made between different producers implicates different cultivation methods, differences in soil, climate and age of trees.

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### 3.3.1. Effect of the various storage regimes on fruit quality

#### 3.3.1.1 . Change in acid, total soluble solids concentrations and TSS: acid ratio

##### *Change in acid concentrations*

1996

The rate of decrease in titratable acid (TA) is one of the main factors controlling the maturity of citrus fruit (Davies and Albrigo, 1994). Differences in total acidity in 1996 caused by the two storage regimes were non-significant (Fig.3.3.1.). Focusing on the trend of decrease of total acidity over time, a more or less constant difference between the beginning of storage and the end could be observed. Except for the fruit from Franschoek (Western Cape), the acid concentrations decreased over the 12-week storage period in fruit from the different producers following more or less the same pattern. These trends were expected, since organic acids are partly used for energy production, alcoholic fermentation and building blocks for the synthesis of sugars (Echeverria and Valich, 1989). At the end of the storage period (Fig.3.3.2.), the only significant difference was between the total acidity in fruit from the different production areas and producers. The effect of the storage treatments was not significant.

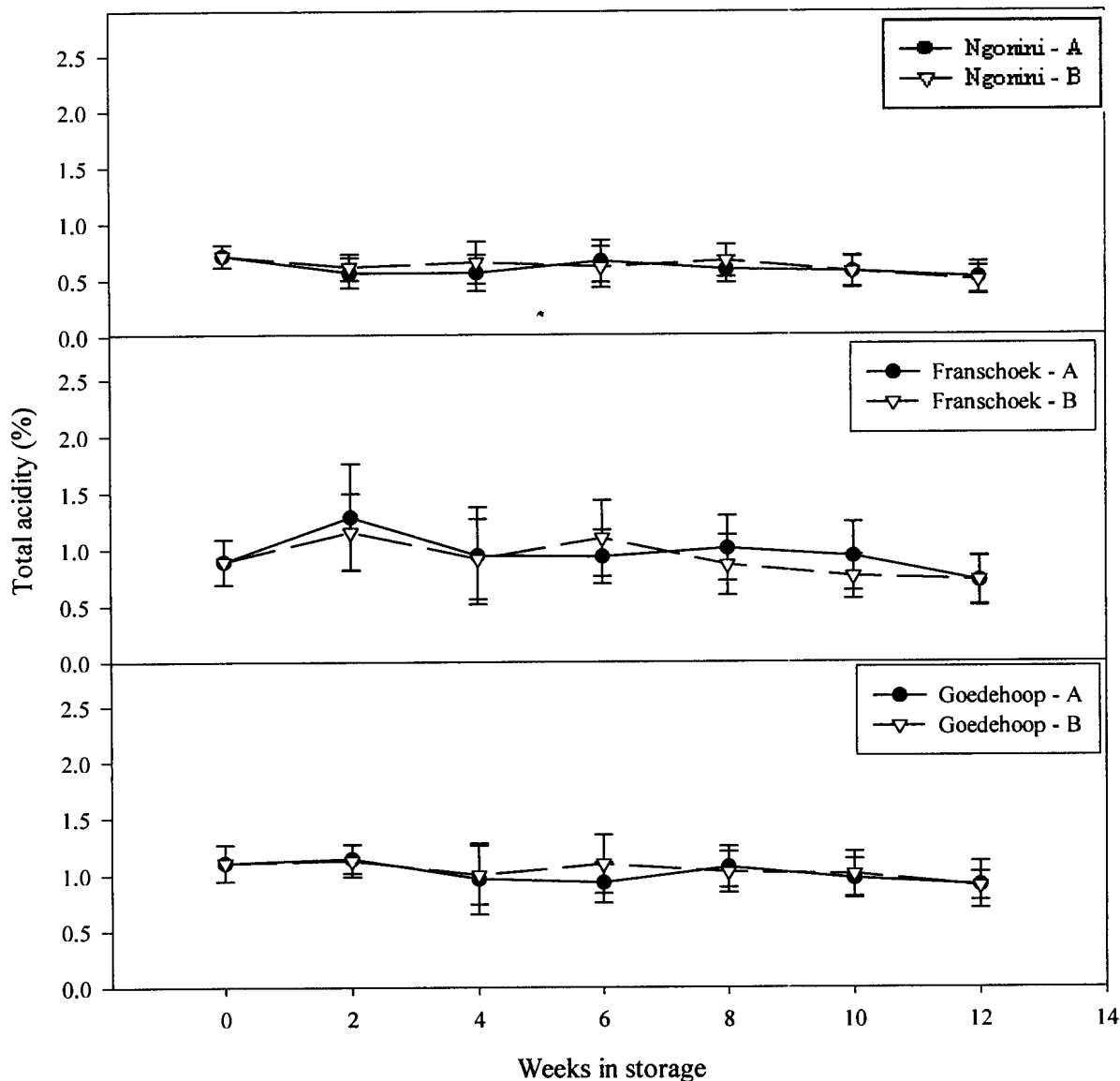


Fig. 3.3.1. Change in total acidity during the 12-week storage period in 1996 for two different storage regimes (A – 4 weeks at 11°C, 4 weeks at 4.5°C, 2 weeks at 8°C, 2 weeks at room temperature; B - 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), from 2 production areas and 3 producers (Swaziland - Ngonini; Western Cape - Franschoek and Goedeheop). Data values represent means  $\pm$ SD of 3 replicates of 5 fruit samples.

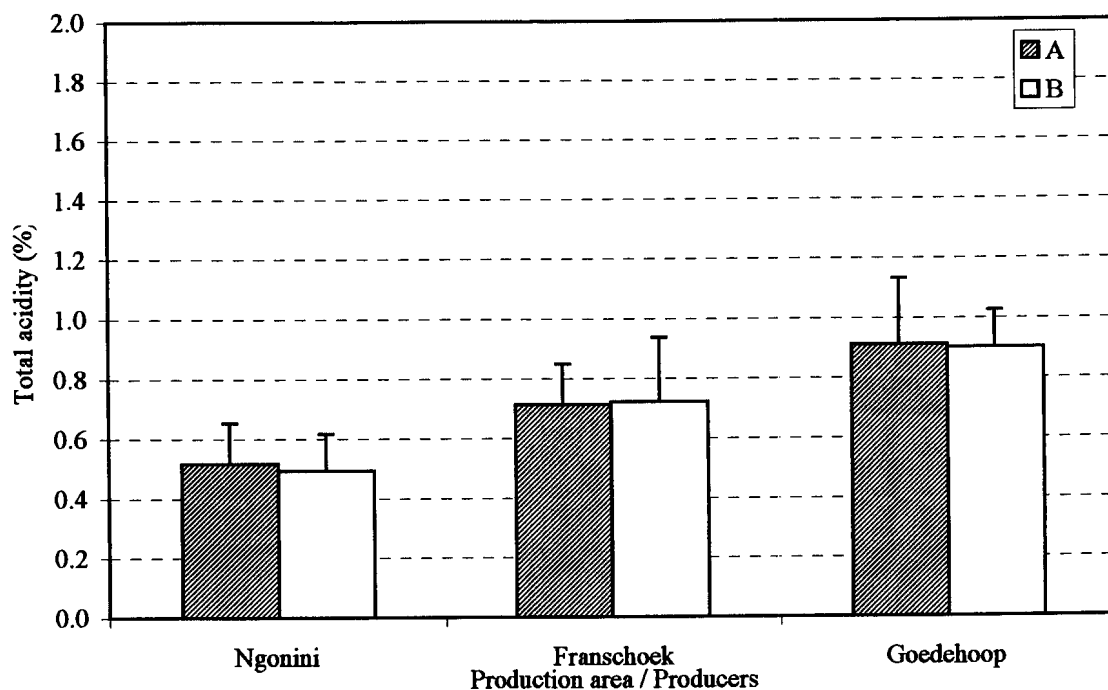


Fig.3.3.2. Total acidity (%) at the end of the 12 week storage period in 1996 for two different storage regimes (A – 4 weeks at 11°C, 4 weeks at 4.5°C, 2 weeks at 8°C, 2 weeks at room temperature; B - 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), from 2 production areas and 3 producers (Swaziland - Ngonini; Western Cape - Franschoek and Goedehoop). Data values represent means  $\pm$ SD of 3 replicates of 5 fruit samples.

### 1997

In 1997, a significant difference (at 5%) in acid concentration between all three producers was obtained from the sixth week and onwards in storage. Although the differences between producers were significant, this is not clear in Fig. 3.3.3. The differences between total acidity in the fruit across the four treatments from Goedehoop (Western Cape) were not significant, while a slight increase in total acidity was observed at all four temperature treatments (Fig.3.3.4.). None of the changes due to storage or shipping temperatures were significant (5%) (Fig. 3.3.3. and Fig.3.3.4.)

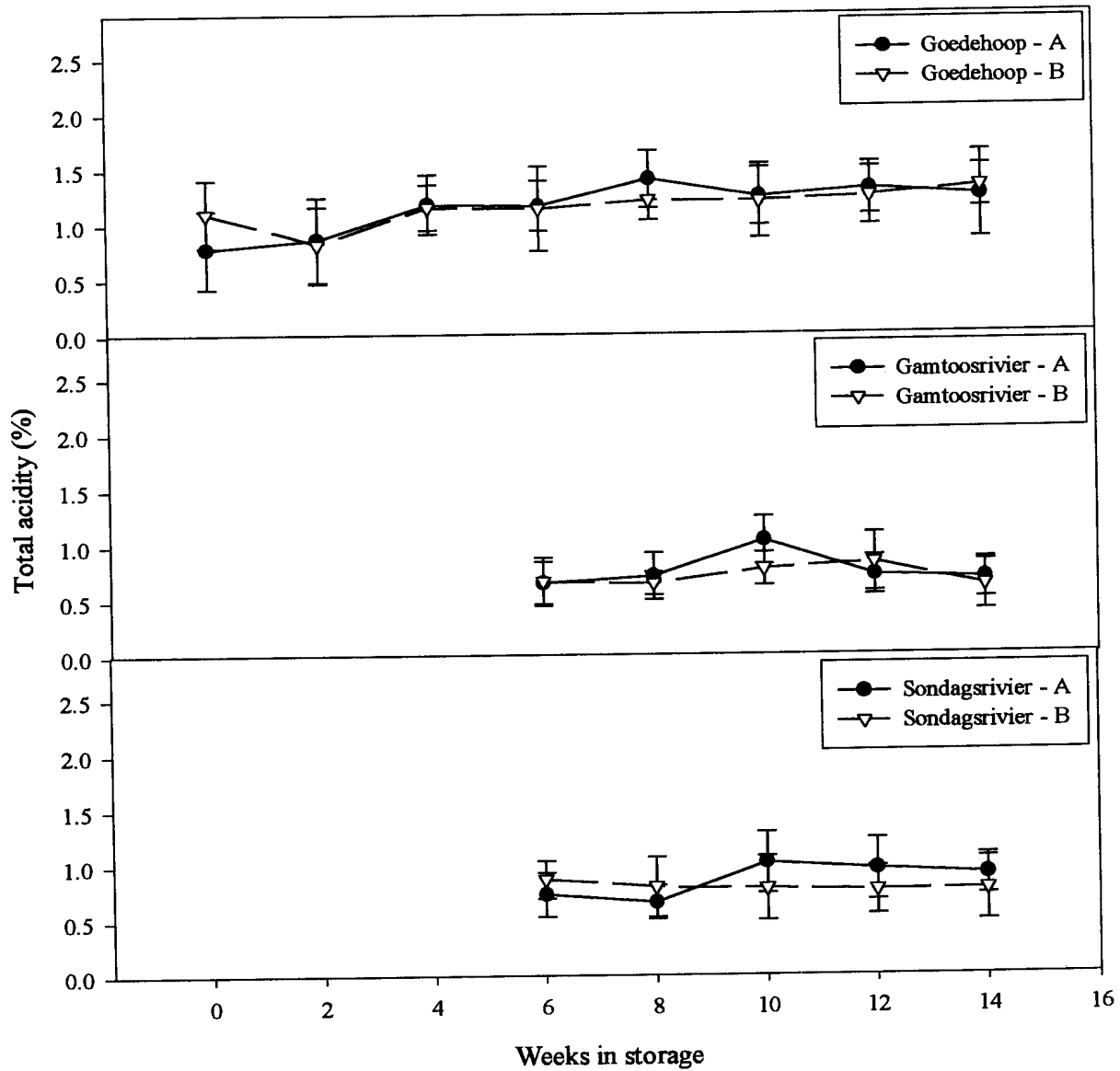


Fig. 3.3.3. Change in total acidity (%) during the 14-week storage period in 1997 for different storage regimes (A – 4 weeks at 4.5°C, 6 weeks at 11°C, 2 weeks at room temperature; B – 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), from 2 production areas and 3 producers (Western Cape - Goedeheoop; Eastern Cape - Gamtoosrivier and Sondagsrivier). Data values represent means  $\pm$ SD of 3 replicates of 3 fruit samples.



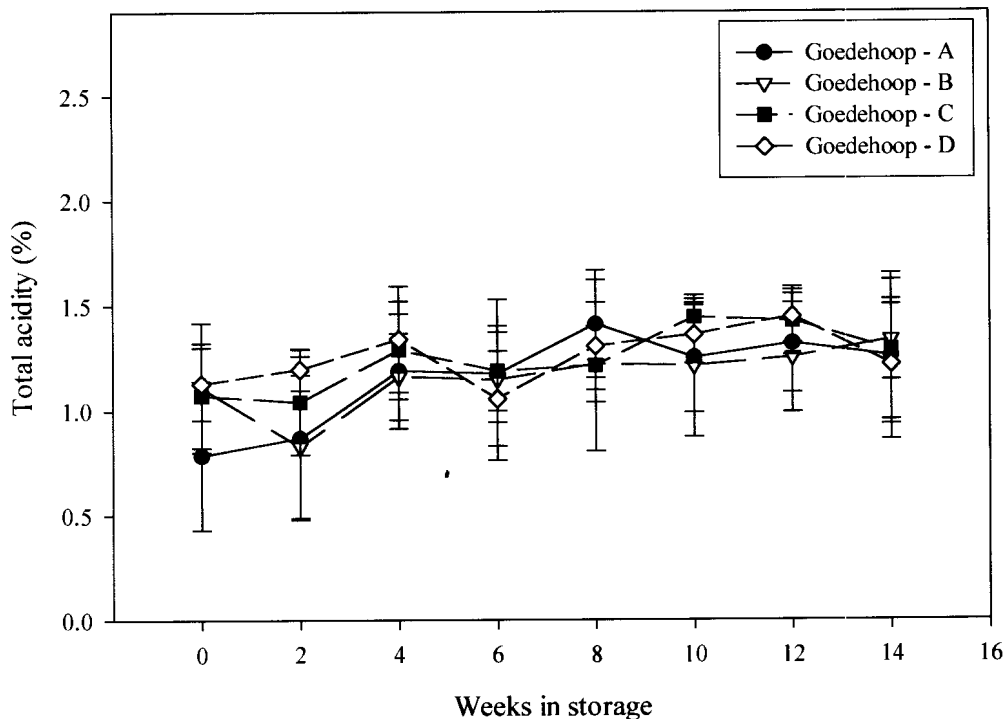


Fig. 3.3.4. Change in total acidity (%) during the 14-week storage period in 1997 for different storage regimes (A – 4 weeks at 4.5°C, 6 weeks at 11°C, 2 weeks at room temperature; B – 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature; C – 4 weeks at 4.5°C, 6 weeks at 4.5°C, 2 weeks at room temperature; D - 4 weeks at 11°C, 6 weeks at 11°C, 2 weeks at room temperature), from 1 production area (Western Cape) and 1 producer (Goedeheop). Data values represent means  $\pm$ SD of 3 replicates of 3 fruit samples.

The total acidity at the end of the storage period of 14 weeks was lower with a shipping temperature of 11°C in fruit from the Eastern Cape (Gamtoosrivier and Sondagsrivier), than fruit with a shipping temperature of 4.5°C (Fig. 3.3.5). Although this occurred in fruit from both the producers from the Eastern Cape, the changes in total acidity in fruit from the Western Cape was not as clear. The fruit from Goedeheop (Western Cape) stored at a shipping temperature of 11°C, before storing at 4.5°C had a higher total acidity at the end of the storage period than any of the other treatments, while the opposite was true in fruit from Gamtoosrivier and Sondagsrivier (Eastern Cape) (Fig.3.3.5). Usually a lower temperature has a preserving effect on citrus fruit, which would cause the conversion of sugars and acids to be slower (Davies and Albrigo, 1994; Grierson and Ben-Yehoshua, 1986). Higher temperatures during storage also have a declining effect on titratable acid (Davies and Albrigo, 1994; Monselise, 1986). The change in percentage acid could be more due the origin of the fruit than the different storage regimes, because the only significant difference (5%) in total acidity was between the production areas.

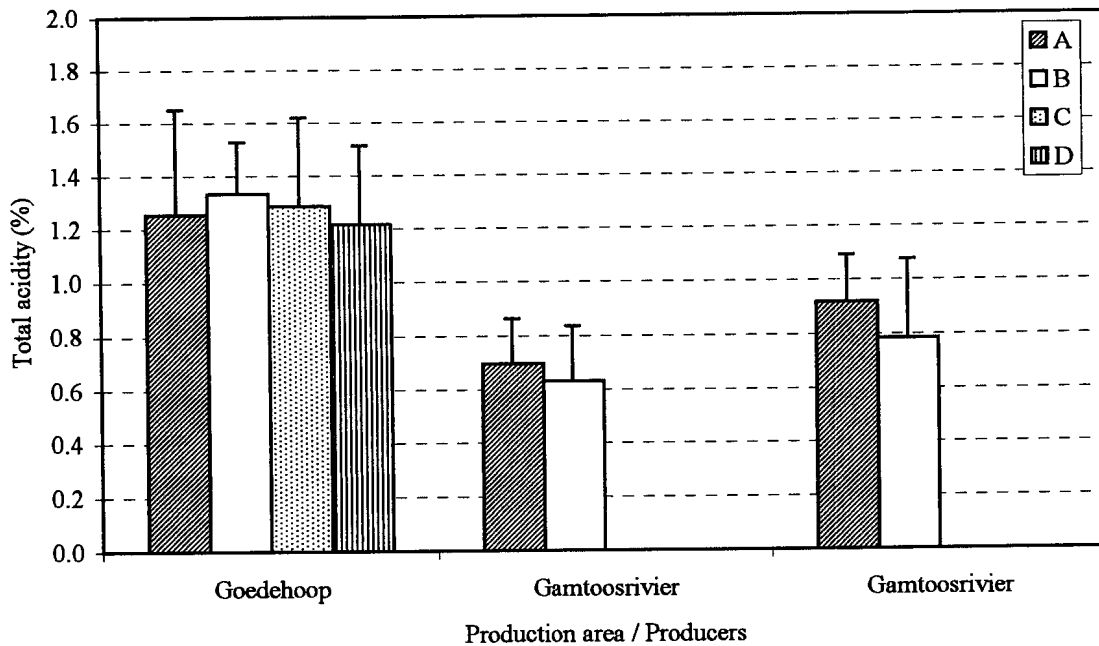


Fig. 3.3.5. Total acidity at the end of the 14 week storage period in 1997 for different storage regimes (A – 4 weeks at 4.5°C, 6 weeks at 11°C, 2 weeks at room temperature; B – 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), from 2 production areas and 3 producers (Western Cape - Goedeheoop; Eastern Cape - Gamtoosrivier and Sondagsrivier). Data values represent means  $\pm$ SD of 3 replicates of 3 fruit samples.

Difference in total acidity between the fruit from different production areas was significant (5%) (Fig. 3.3.5.). Fruit from Goedeheoop in the Western Cape had a much higher total acidity than the fruit from the Eastern Cape. The fruit from the Eastern Cape (Sondagsrivier and Gamtoosrivier) was received later than the fruit from the Western Cape but was more or less the same age. In fruit from the Eastern Cape the two different temperature regimes had a slight, but non-significant influence on the total acidity. Davies and Albrigo (1994) found that the rate of decrease of titratable acid is positively correlated with the average temperatures during the season. For example acid levels decrease much more rapidly in low tropical than in subtropical growing regions due to higher mean temperatures. Higher temperatures increase respiration rates causing less storage of acids in the vacuoles and their more rapid utilization in metabolism. This was observed in 1996 (Fig.3.3.2) where the total acidity from fruit from a warmer climate (Ngonini, Swaziland) was lower than that of fruit from the cooler climate (Franschoek and Goedeheoop, Western Cape) and in 1997 where the fruit from the Eastern Cape (Gamtoosrivier and Sondagsrivier) had a lower total acidity than the fruit from the Western Cape (Goedeheoop). Although both these regions are classified as cooler production areas, the Eastern Cape is known to be hotter than the Western Cape.

The differences in the changes of Total Soluble Solids (TSS) between the treatments and producers (Fig.3.3.6. and Fig. 3.3.7.) are not as clear as the difference between total acidity (Fig. 3.3.1 – Fig.3.3.5). In 1996 the differences in TSS between the producers were not significant. The temperature treatments also did not have a significant effect (Fig.3.3.6 and Fig. 3.3.7.).

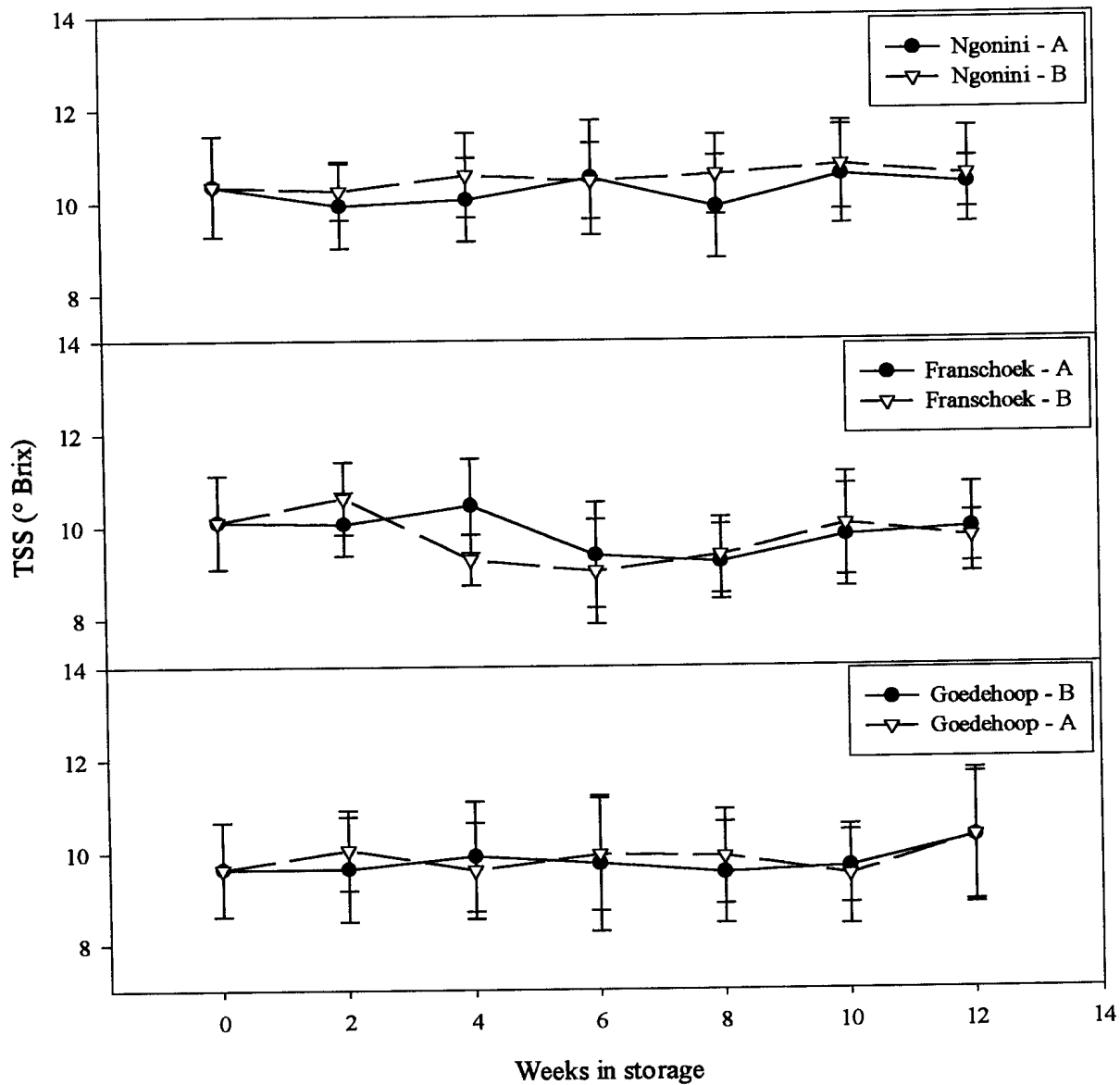


Fig. 3.3.6. Change in TSS (°Brix) during the 12-week storage period in 1996 for different storage regimes (A – 4 weeks at 11°C, 4 weeks at 4.5°C, 2 weeks at 8°C, 2 weeks at room temperature; B - 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), from 2 production areas and 3 producers (Swaziland - Ngonini; Western Cape - Franschoek and Goedeheoop). Data values represent means  $\pm$ SD of 3 replicates of 5 fruit samples.

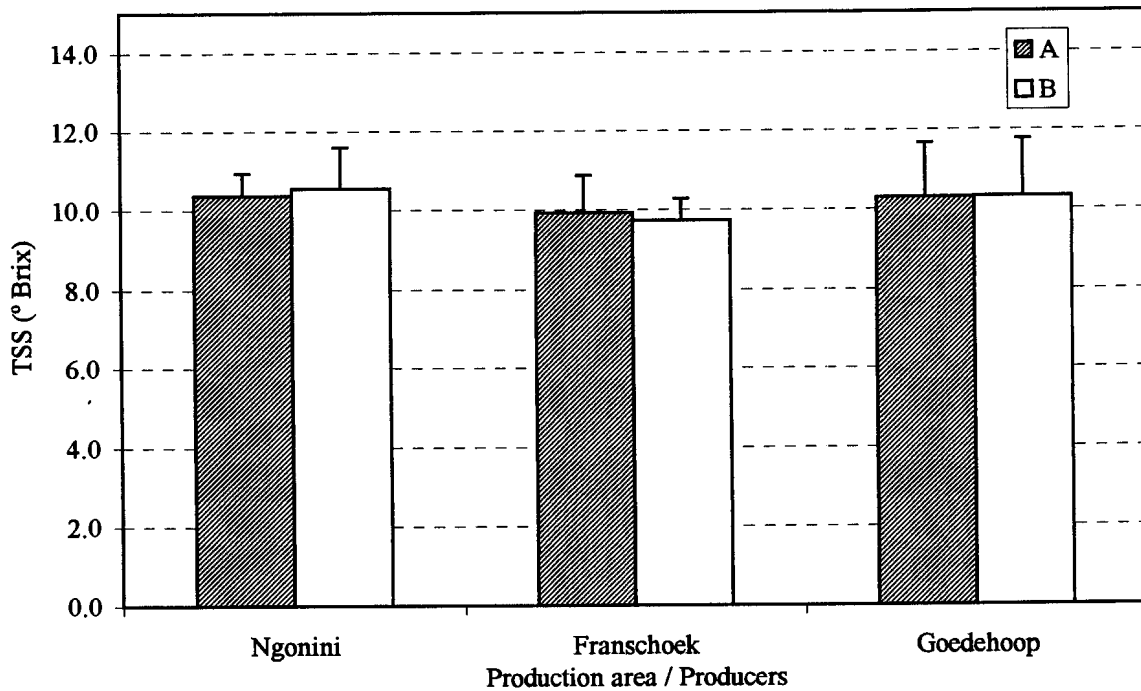


Fig.3.3.7. TSS (°Brix) at the end of the 12 week storage period in 1996 for different storage regimes (A – 4 weeks at 11°C, 4 weeks at 4.5°C, 2 weeks at 8°C, 2 weeks at room temperature; B - 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), from 2 production areas and 3 producers (Swaziland - Ngonini; Western Cape - Franschoek and Goedehoop). Data values represent means  $\pm$ SD of 3 replicates of 5 fruit samples.

The only clear indication observed (Fig. 3.3.6.) is that a lower shipping temperature (4.5°C) delayed the decrease in TSS by 2 weeks in the first 4 weeks of storage only in fruit from Franschoek. A slight rise in TSS over time can also be observed in all the treatments. This might be due to the oxidation of sugars taking place inside the fruit while in storage (Echeverria and Valich, 1989). Echeverria and Ismail (1987) reported that the TSS (°Brix) in sweet oranges, tangerines and limes also continue to increase for some time during postharvest storage.

Although the difference between producers in 1997 was significant (at 5%) from the sixth week of storage, this is not clear in Fig. 3.3.8. The TSS of the fruit obtained from the Western Cape (Goedehoop) was higher than that of the fruit from both producers from the Eastern Cape (Gamtoosrivier and Sondagsrivier). The difference between fruit from the Eastern Cape was not that clear (Fig.3.3.8.). There also was no clear trend regarding the changes in TSS in these fruit. The fruit from the Western Cape (Goedehoop) showed a definite rise in TSS (Fig.3.3.8. and Fig.3.3.9), especially in the first part of the storage period. This rise followed more or less the same trend in all the treatments (Fig.3.3.9).

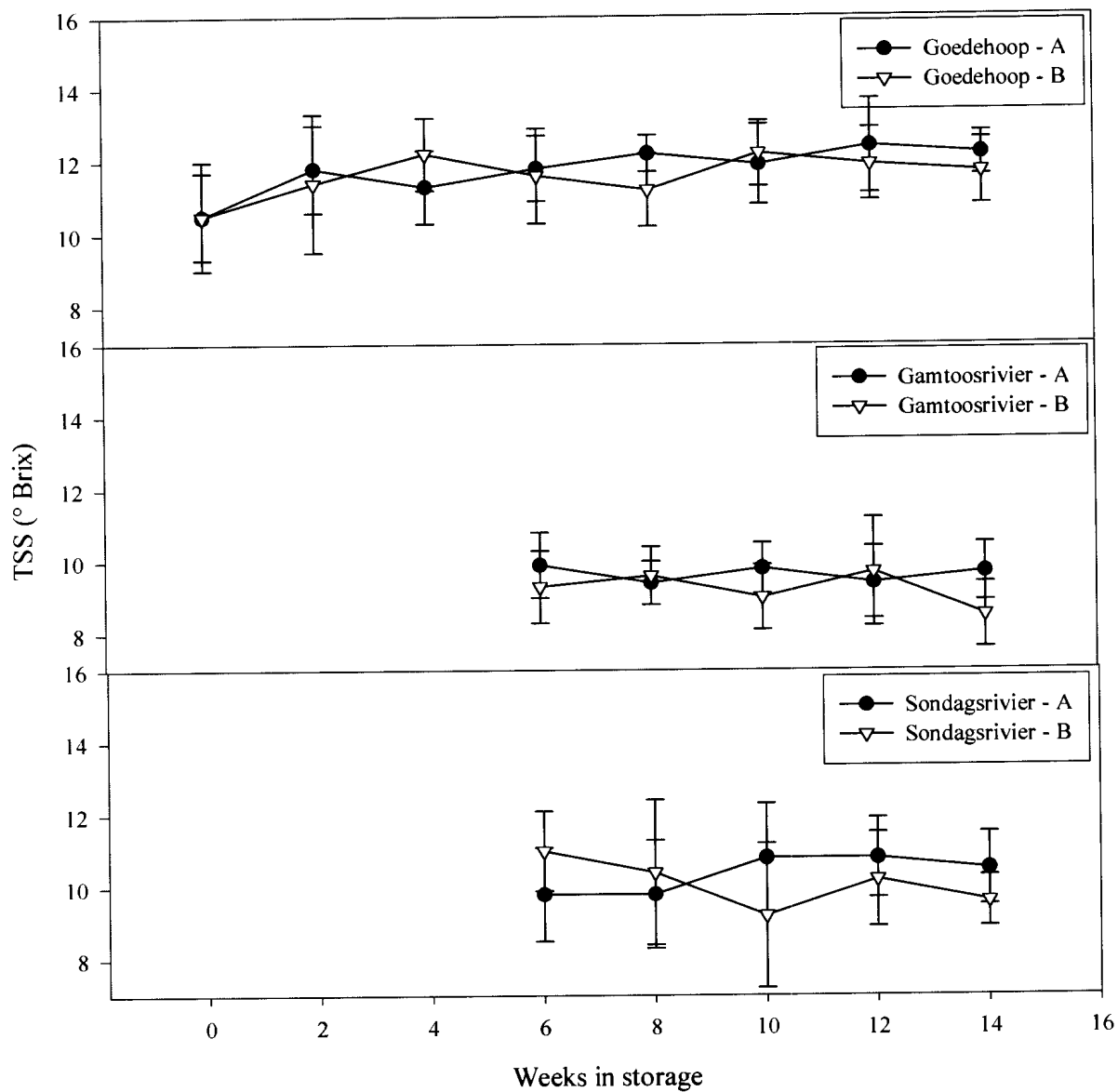


Fig. 3.3.8. Change in TSS (°Brix) during the 14-week storage period in 1997 for different storage regimes (A – 4 weeks at 4.5°C, 6 weeks at 11°C, 2 weeks at room temperature; B – 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), from 2 production areas and 3 producers (Western Cape - Goedeheoop; Eastern Cape - Gamtoosrivier and Sondagsrivier). Data values represent means  $\pm$ SD of 3 replicates of 3 fruit samples.

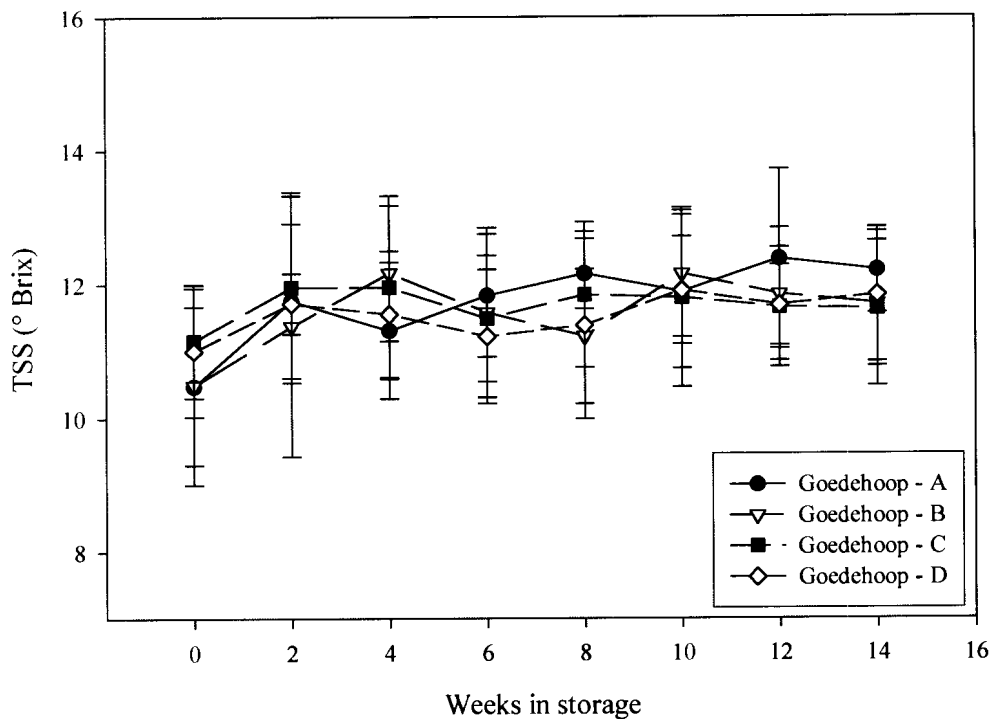


Fig.3.3.9. Change in TSS (°Brix) during the 14-week storage period in 1997 for different storage regimes (A – 4 weeks at 4.5°C, 6 weeks at 11°C, 2 weeks at room temperature; B – 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), from 1 production area (Western Cape) and 1 producer (Goedehoop). Data values represent means  $\pm$ SD of 3 replicates of 3 fruit samples.

In 1997 the effects of the different treatments were visible, although not significant, across all the producers and production areas (Fig.3.3.10). The fruit treated with a low shipping temperature and a higher storage temperature (treatment A) had a higher TSS at the end of the 14 week storage period than the fruit treated with a high shipping temperature and a lower storage temperature (treatment B). This could probably be due to a more preserving effect of the low shipping temperature, by slowing respiration more effectively with the low temperatures at a later stage of storage (Van Rensburg et al., 1995). The same trend with respect to lower temperature storage after shipping was observed with the fruit from Goedehoop (Western Cape) that was stored at one temperature for the whole period (treatments C, 4.5°C and D, 11°C). However, the low storage temperature for the whole period (treatment C) led to a lower TSS of fruit at the end of the storage period in comparison to the storage regime where only the shipping temperature was low (treatment A). This could be an indication of the importance of the first two weeks in the storage period of citrus fruit with regard to temperature regimes during shipping.

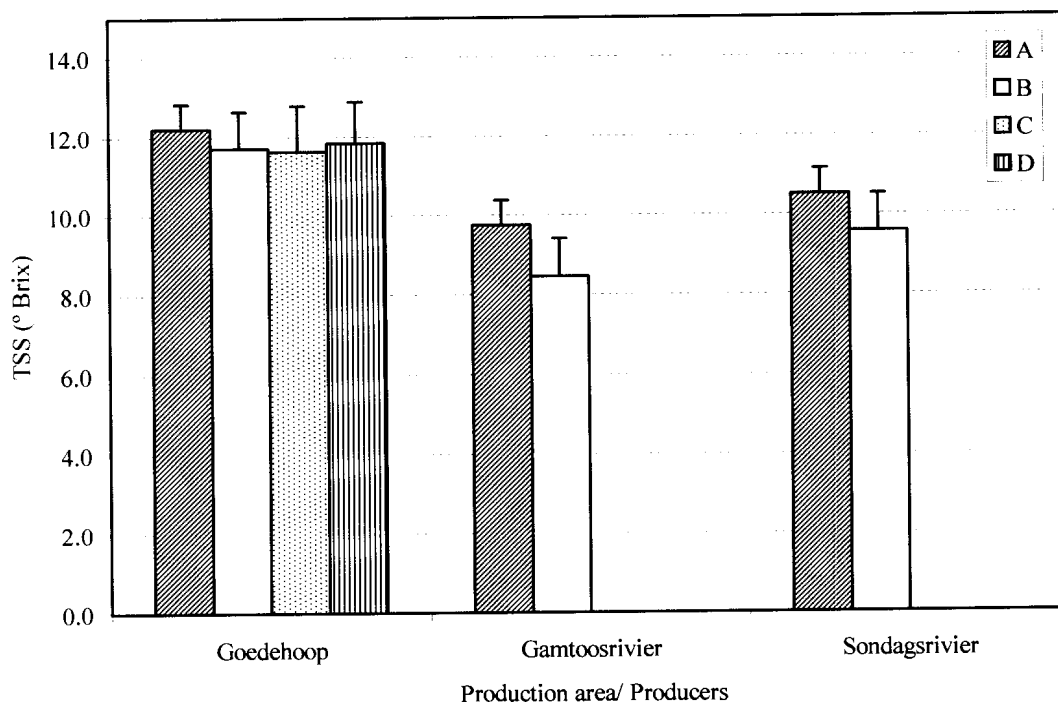


Fig. 3.3.10. TSS (°Brix) at the end of the 14-week storage period in 1997 for different storage regimes (A – 4 weeks at 4.5°C, 6 weeks at 11°C, 2 weeks at room temperature; B – 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), from 2 production areas and 3 producers (Western Cape - Goedeheoop; Eastern Cape - Gamtoosrivier and Sondagsrivier). Data values represent means  $\pm$ SD of 3 replicates of 3 fruit samples.

### *TSS:Acid ratio*

#### 1996

Citrus fruit are non-climacteric and need to be mature before harvesting so that a suitable juice percentage and TSS concentration can be attained on the tree (Grierson and Ben-Yehoshua, 1986). The TSS:Acid ratio is considered the most reliable index of maturity in citrus fruit (Grierson and Ben-Yehoshua, 1986). The TSS:Acid ratio changes before harvesting are due to a simultaneous increase in TSS and decrease in titratable acid (TA) (Grierson and Ben-Yehoshua, 1986; Aworh et al., 1991). A rise in the TSS value due to respiration has the effect that the TSS:Acid ratio also increases with storage (Echeverria and Ismail, 1987).

In both 1996 and 1997 the differences in TSS:Acid ratio of fruit from different producers were significant at 5%, while no significant difference was observed between any of the temperature treatments (Fig. 3.3.11, Fig. 3.3.12 and Fig. 3.3.12.). Since no trend between fruit treated with different temperatures was observed at the end of the storage period, the area where fruit were

produced appeared to have the biggest influence on the internal quality of the fruit as suggested by Monselise (1981).

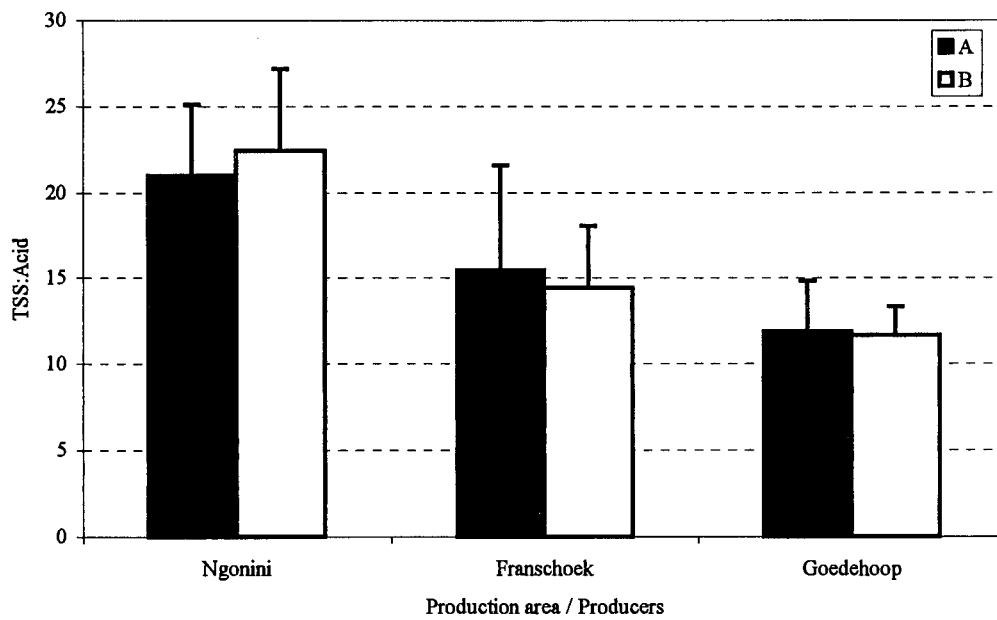


Fig.3.3.11. TSS:Acid ratio at the end of the 12-week storage period in 1996 for different storage regimes (A – 4 weeks at 11°C, 4 weeks at 4.5°C, 2 weeks at 8°C, 2 weeks at room temperature; B - 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), from 2 production areas and 3 producers (Swaziland - Ngonini; Western Cape - Franschoek and Goedeheop). Data values represent means  $\pm$ SD of 3 replicates of 5 fruit samples.

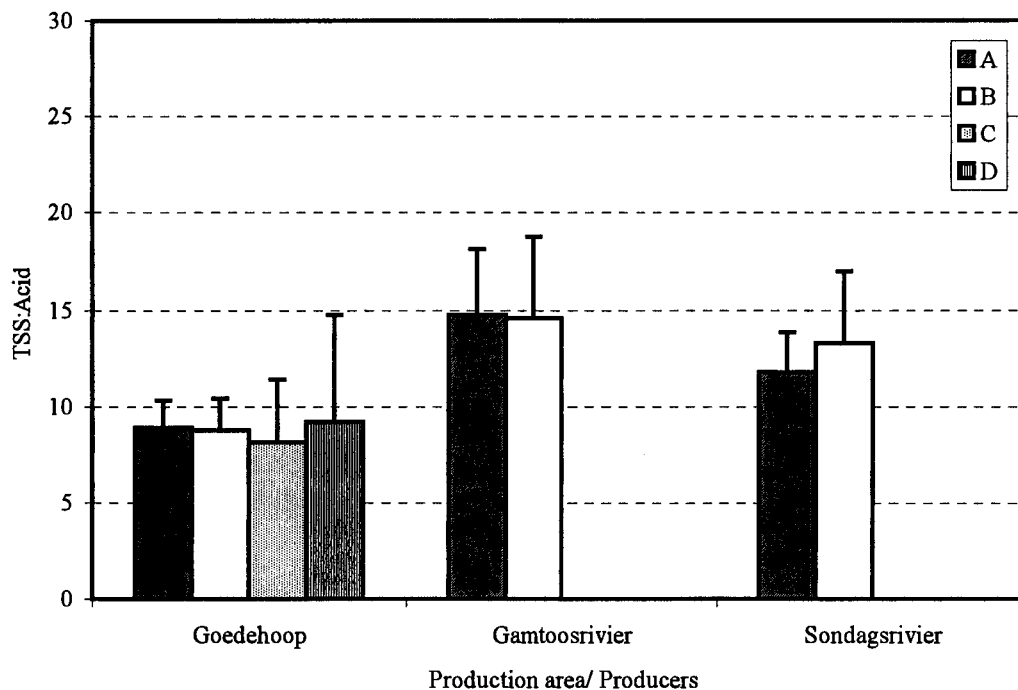


Fig.3.3.12. Change in TSS:Acid ratio during the 14-week storage period in 1997 for different storage regimes (A – 4 weeks at 4.5°C, 6 weeks at 11°C, 2 weeks at room temperature; B – 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room



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temperature), from 2 production areas and 3 producers (Western Cape - Goedehoop; Eastern Cape - Gamtoosrivier and Sondagsrivier). Data values represent means  $\pm$ SD of 3 replicates of 3 fruit samples.

### **3.3.1.2 . Change in percentage juice, peel thickness and percentage peel moisture**

#### 1996

In 1996 the percentage juice content of the fruit only began to differ significantly (5%) after eight weeks in storage at the two different storage regimes. This difference seemed to be associated mainly with the different producers and not the storage temperature (Fig.3.3.13). At the end of the experiment the percentage juice was lower in all treatments than in the beginning of the storage period (Fig.3.3.13 and Fig.3.3.15). These results were contrary to those reported by Bertolini et al. (1986). They recorded an increase in juice yield during cold storage.

The change in percentage juice from fruit from Goedehoop (Western Cape) showed a clear trend in both treatments (Fig.3.3.13.). In the first 6 weeks of the storage period the percentage juice decreased and then it increased until the end of the storage period. In both treatments, the percentage juice at the end of storage was higher than at the beginning. The percentage juice from the Ngonini (Swaziland) and Franschoek (Western Cape) increased at a later stage (10 weeks), but declined over the whole storage period (Table 3.3.1). This might be related to changes in peel thickness (Fig. 3.3.14) as suggested by Fucik (1981). Changes in peel thickness were generally larger in fruit originating from the Western Cape (Goedehoop and Franschoek) compared to Swaziland (Ngonini) (Fig.3.3.14). Treatment B (11°C shipping, 4.5°C storage) caused larger changes in peel thickness in fruit from the Western Cape (Goedehoop and Franschoek) in contrast to fruit from Swaziland (Ngonini) although this change was not significant. The fruit from Ngonini had thicker, stronger peels and the fruit were generally larger than the fruit from the Western Cape. The percentage peel moisture was not measured in 1996.

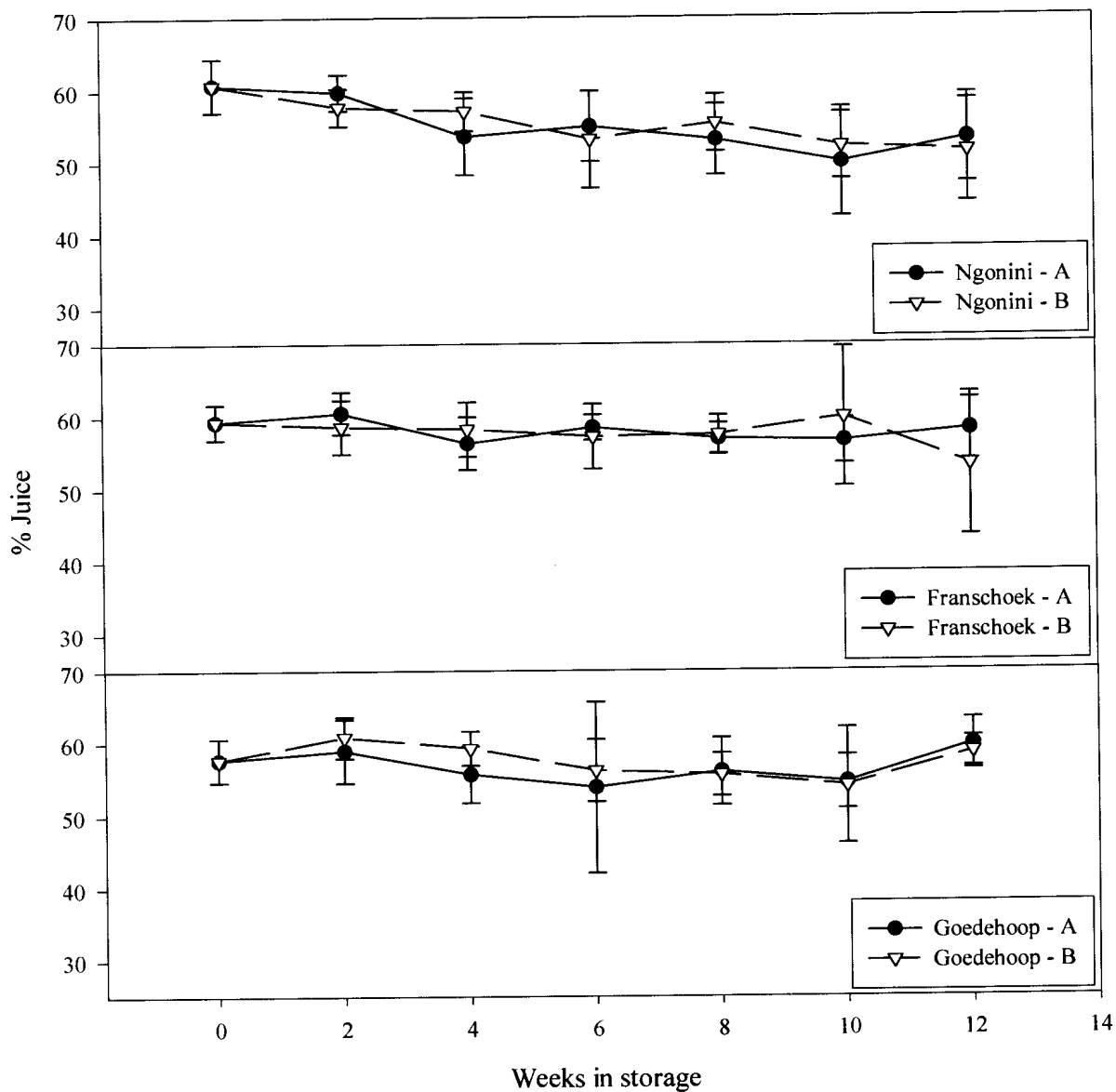


Fig. 3.3.13. Change in % juice during the 12-week storage period in 1996 for different storage regimes (A – 4 weeks at 11°C, 4 weeks at 4.5°C, 2 weeks at 8°C, 2 weeks at room temperature; B - 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), from 2 production areas and 3 producers (Swaziland - Ngonini; Western Cape - Franschoek and Goedeboom). Data values represent means  $\pm$ SD of 3 replicates of 5 fruit samples.

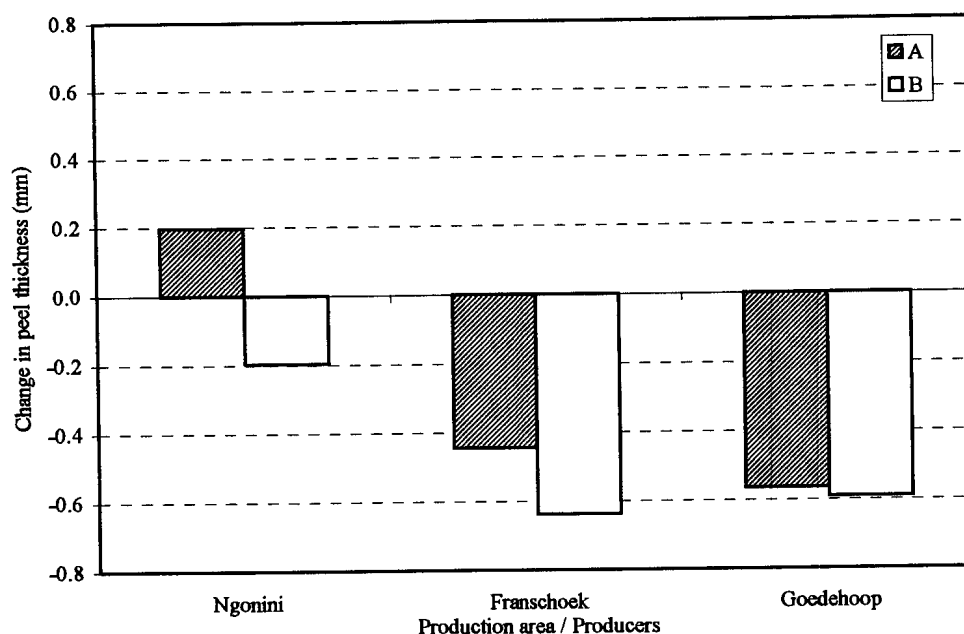


Fig.3.3.14. Changes in peel thickness (mm) at the end of the 12-week storage period in 1996 for different storage regimes (A – 4 weeks at 11°C, 4 weeks at 4.5°C, 2 weeks at 8°C, 2 weeks at room temperature; B - 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), from 2 production areas and 3 producers (Swaziland - Ngonini; Western Cape - Franschoek and Goedeheop). Data values represent the difference in means of 3 replicates of 5 fruit samples from the beginning and the end of the storage period.

### 1997

In 1997, the rise in percentage juice at later stages of storage was not as clear as in 1996, but only occurred to a lesser extent after 10 to 12 weeks of storage. There was no significant difference in the influence of different producers or temperature treatments on the change of percentage juice except for the last measurement exhibiting a significant difference between producers (Fig. 3.3.15 and Fig.3.3.16). The lower percentage juice towards the end of the storage period could not be related to the changes in peel thickness (Fig.3.3.17) as in the previous year. A possible reason for this could be that the fruit sizes were similar from the different producers, while in 1996 there was a big difference in fruit size between the fruit from different producers. The percentage juice in fruit from Goedeheop (Western Cape) increased in storage in 1996, but decreased in 1997 probably because of the difference

in storage temperatures (Table 3.3.1).

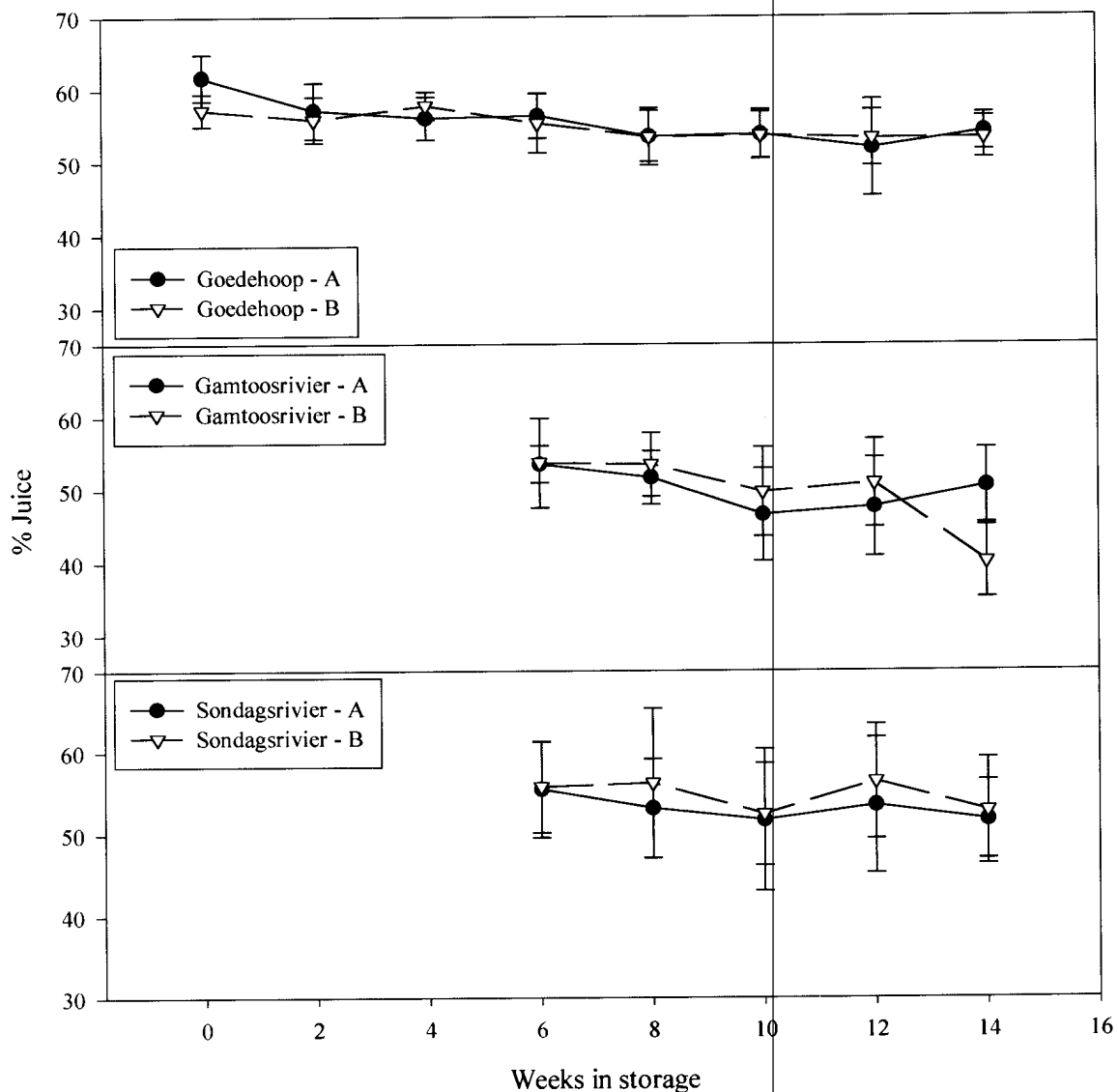


Fig.3.3.15. Change in % juice during the 14-week storage period in 1997 for different storage regimes (A – 4 weeks at 4.5°C, 6 weeks at 11°C, 2 weeks at room temperature; B – 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), from 2 production areas and 3 producers (Western Cape - Goedeheop; Eastern Cape - Gamtoosrivier and Sondagsrivier). Data values represent means  $\pm$ SD of 3 replicates of 3 fruit samples.

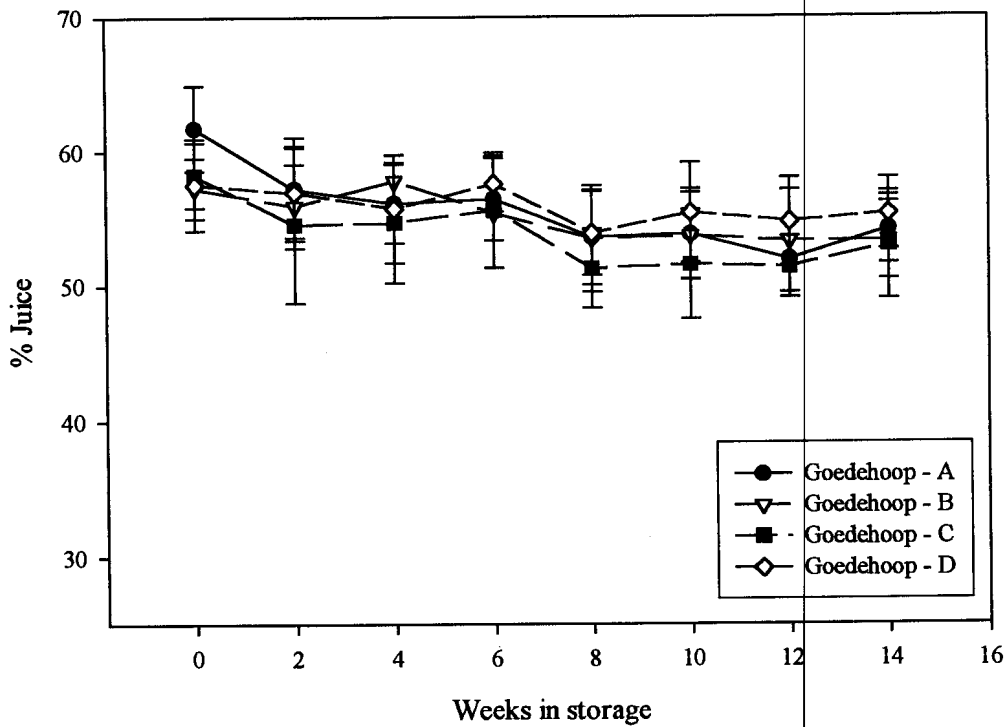


Fig. 3.3.16. Change in % juice during the 14-week storage period in 1997 for different storage regimes (A – 4 weeks at 4.5°C, 6 weeks at 11°C, 2 weeks at room temperature; B – 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), from 1 production area (Western Cape) and 1 producer (Goedehoop). Data values represent means  $\pm$ SD of 3 replicates of 3 fruit samples.

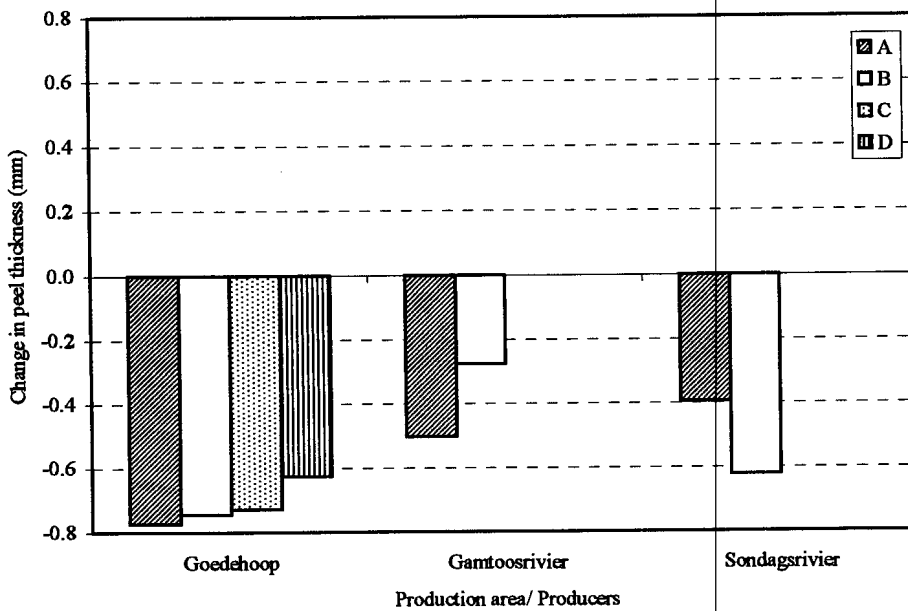


Fig.3.3.17. Change in peel thickness at the end of the 14-week storage period in 1997 for different storage regimes (A – 4 weeks at 4.5°C, 6 weeks at 11°C, 2 weeks at room temperature; B – 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), from 2 production areas and 3 producers (Western Cape - Goedehoop; Eastern Cape - Gamtoosrivier and Sondagsrivier). Data values represent difference of means of 3 replicates of 3 fruit samples between beginning and end of

storage.

Table 3.3.1. Difference in moisture content (%) in juice and peels of ‘Minneola’ tangelos between the beginning and at the end of the storage period after different temperature treatments in 1996 (storage regimes described in Fig.3.3.1), and 1997 (as described in Fig.3.3.4)

Producer	1996		1997							
	A	B	A		B		C		D	
	Juice %	Juice %	Peel %	Juice %	Peel %	Juice %	Peel %	Juice %	Peel %	Juice %
Ngonini	7.35	9.26								
Franschoek	1.20	6.00								
Goedehoop	-2.13	-1.04	-4.5	7.54	-1.8	3.89	-4.3	5.22	-2.0	2.18
Gamtoosrivier			-0.6	3.00	-4.9	13.62				
Sondagsrivier			-0.9	3.69	-0.7	2.91				

The differences in the peel moisture of fruit measured in 1997 were only significant at 5% between producers but not significant between the treatments (Fig.3.3.18). However, as the percentage juice decreased over the storage period, the moisture content of the peel increased (Table 3.3.1).

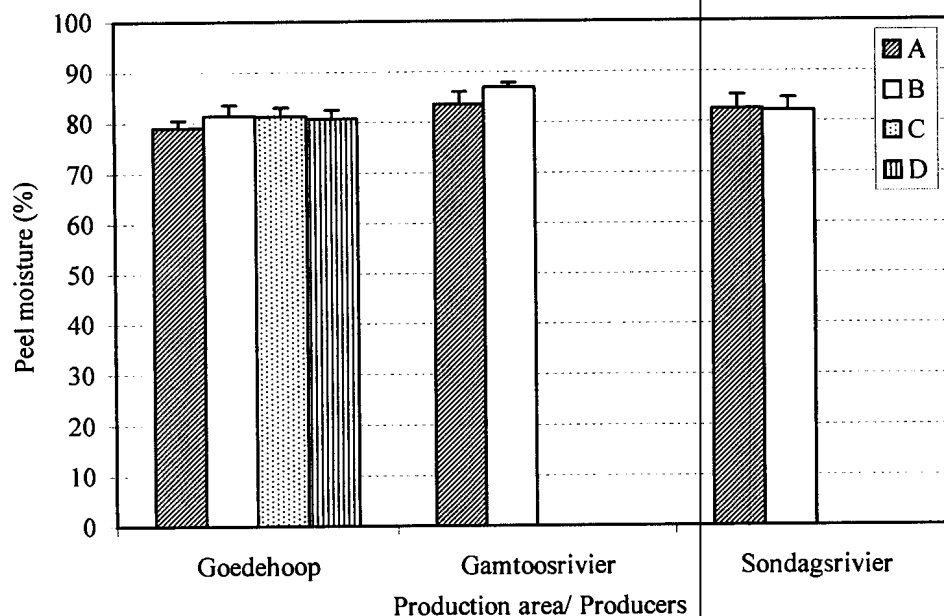


Fig. 3.3.18. Peel moisture (%) after the 14-week storage period in 1997 for different storage regimes (A – 4 weeks at 4.5°C, 6 weeks at 11°C, 2 weeks at room temperature; B – 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), from 2 production areas and 3 producers (Western Cape - Goedehoop; Eastern Cape - Gamtoosrivier and Sondagsrivier). Data values represent means ±SD of 3 replicates of 3 fruit samples.

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Monselise (1986) and Grierson and Ben –Yehoshua (1986) found that after 2 months storage at 20°C and 50-75% RH, the peel of Valencia oranges lost 9.5% of its weight while the pulp lost only 2.1%. Since the peel was used by the fruit for translocation of moisture to the atmosphere, long-term storage of ‘Minneola’ tangelos caused the fruit to become dried and brittle as reported by Grierson and Ben-Yehoshua (1986). Fucik (1981) also found that the peel of the fruit became thinner and leather-like with age. The observed increase in peel moisture cannot be explained, since one would expect the moisture in the peel to lessen when becoming thinner and dry causing percentage juice to increase.

### 3.3.1.3 . Water loss in storage

Grierson and Ben-Yehoshua (1986) found that a weight loss as low as 5% renders oranges unsaleable, while shrinkage is already visible at half this value. This means that 50% of commercially tolerable weight loss is invisible. The weight loss mainly involves the peel and not the pulp of the fruit.

#### 1996

In 1996 the weight loss differed significantly at 5% between production areas / producers. This difference is not clear in Fig.3.3.19. Weight loss caused by treatment B (shipping 11°C, storage 8°C) was higher in fruit from the Western Cape (Goedehoop and Franschoek), in comparison to fruit from Swaziland (Ngonini) (Table 3.3.2). In fruit from Ngonini the different storage temperatures only showed significant differences (5%) after the eighth week in storage. This difference disappeared after 12 weeks in storage when the fruit was kept at ambient temperatures.

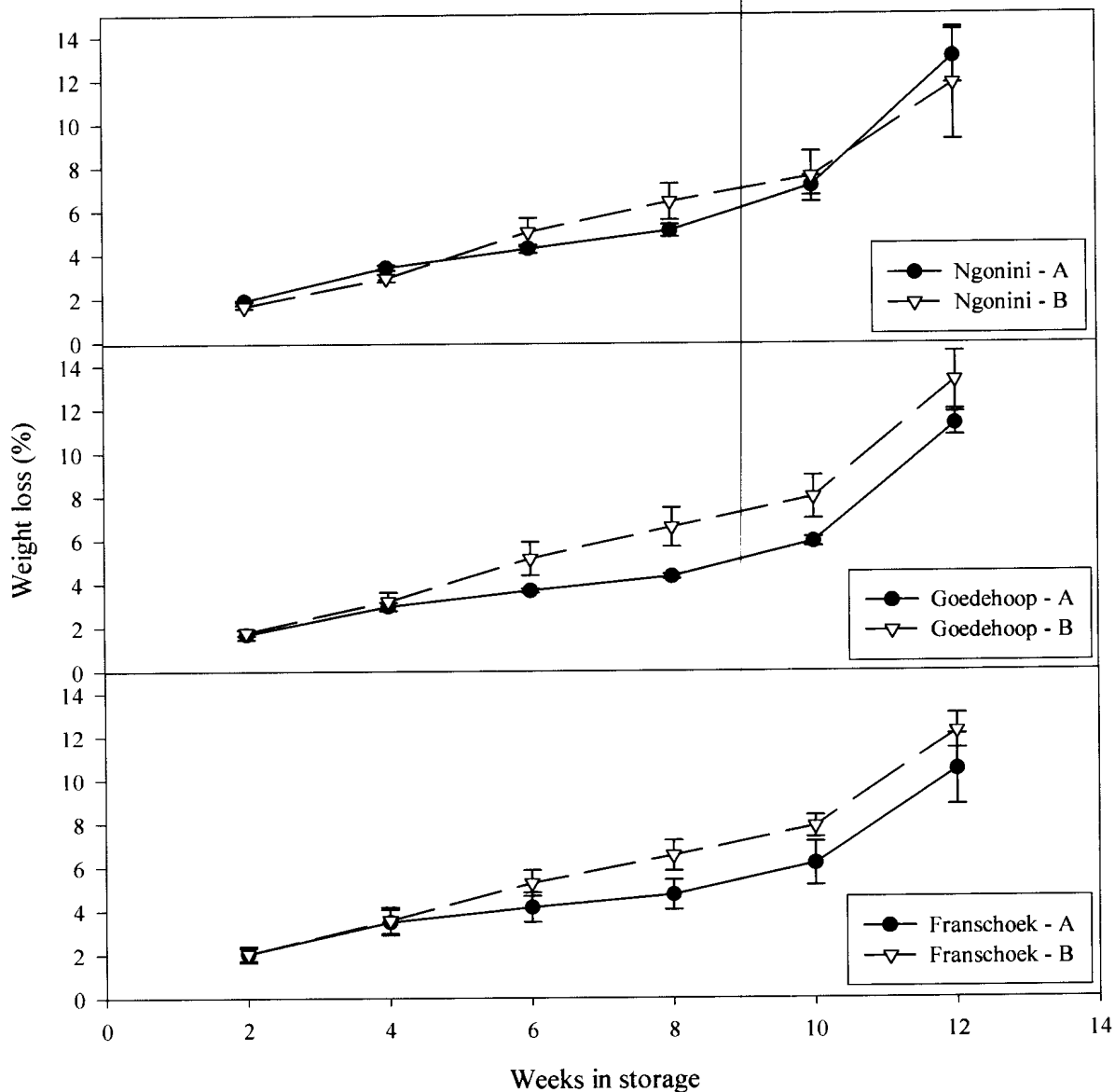


Fig.3.3.19. Weight loss during the 12-week storage period in 1996 for different storage regimes (A – 4 weeks at 11°C, 4 weeks at 4.5°C, 2 weeks at 8°C, 2 weeks at room temperature; B - 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature) from 2 production areas and 3 producers (Swaziland - Ngonini; Western Cape - Franschoek and Goedehoop). Data values represent means  $\pm$ SD of 3 replicates of 5 fruit samples.

### 1997

In 1997 the weight loss was also highly significant at 5% for the different producers / production areas. When comparing storage regimes A and B for all the producers, the storage treatments only influenced the weight loss significantly in week 6 and week 10 (Fig.3.3.20 and Table 3.3.2). Although the effects of the storage treatments were not always significant, the interactions between the producers and the storage regimes were highly significant (1%) during the whole time of storage.



According to Clarke (1980), an interaction in a factorial experiment provides information on the way the two factors interact, and when the interaction is significant the two factors cannot be examined apart. When looking at the % weight loss caused by temperature, it is important to look at the different producers separately. The differences in weight loss in fruit from Gamtoosrivier (Eastern Cape) were significant (5%) in all the weeks of storage. In fruit from Gamtoosrivier and Sondagsrivier, treatment B gave a higher weight loss, but in fruit from Goedeheop, treatment A gave a higher weight loss. This could probably be attributed to the difference in fruit size and physiological age.

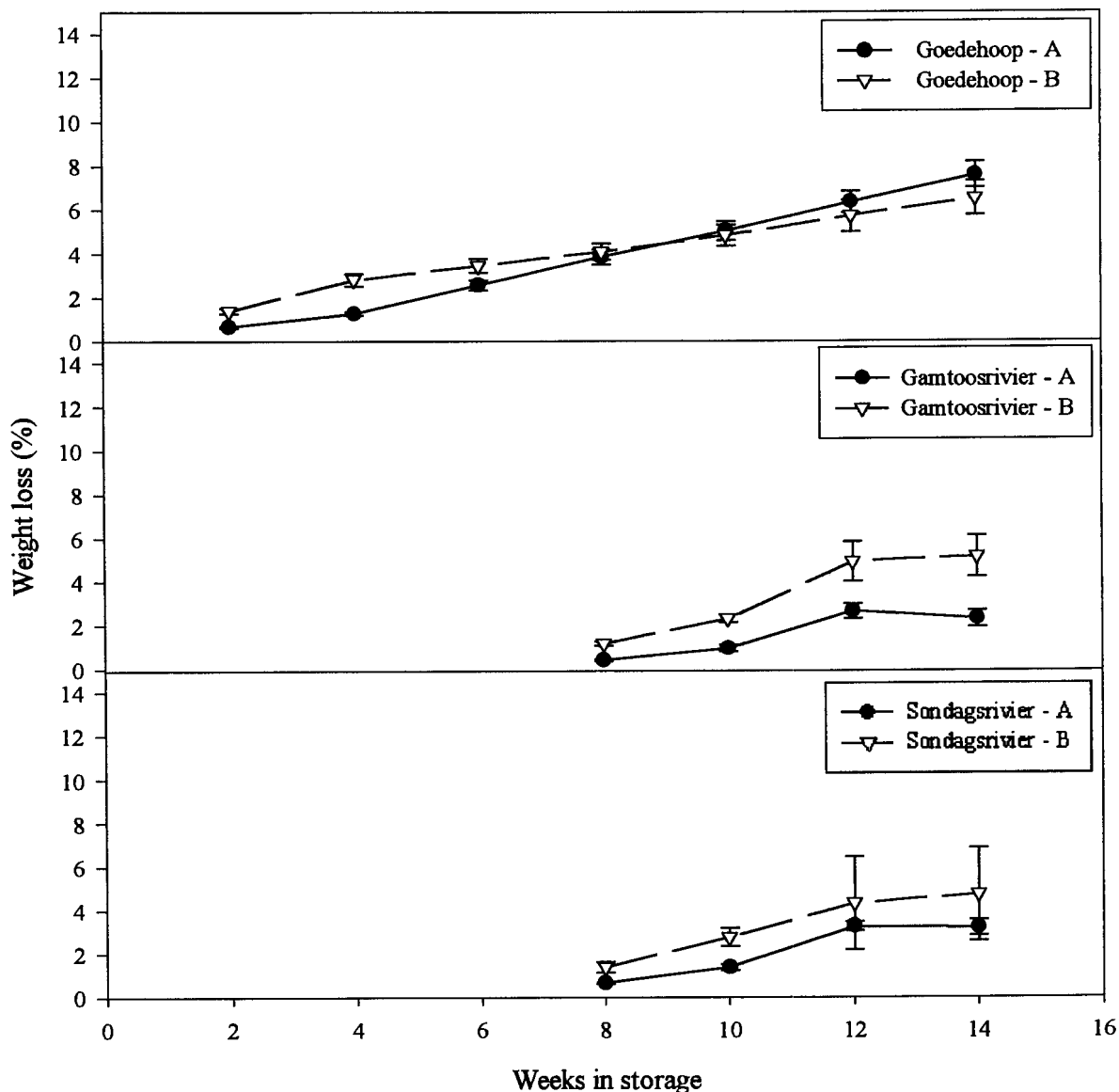


Fig. 3.3.20. Weight loss (%) during the 14-week storage period in 1997 for different storage regimes (A – 4 weeks at 4.5°C, 6 weeks at 11°C, 2 weeks at room temperature; B – 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), from 2 production areas and 3 producers (Western Cape - Goedeheop; Eastern Cape - Gamtoosrivier and Sondagsrivier). Data values represent means  $\pm$  SD of 3 replicates of 3 fruit samples.

The effect of temperature on percentage weight loss was observed in fruit from the producer in Goedehoop. In treatment A and B the fruit was moved to a different temperature after 4 weeks. The percentage weight loss in fruit from A increased as the storage temperature was higher than the shipping temperature and the percentage weight loss in B was slowed down, as the storage temperature was lower than the shipping temperature (Fig.3.3.21).

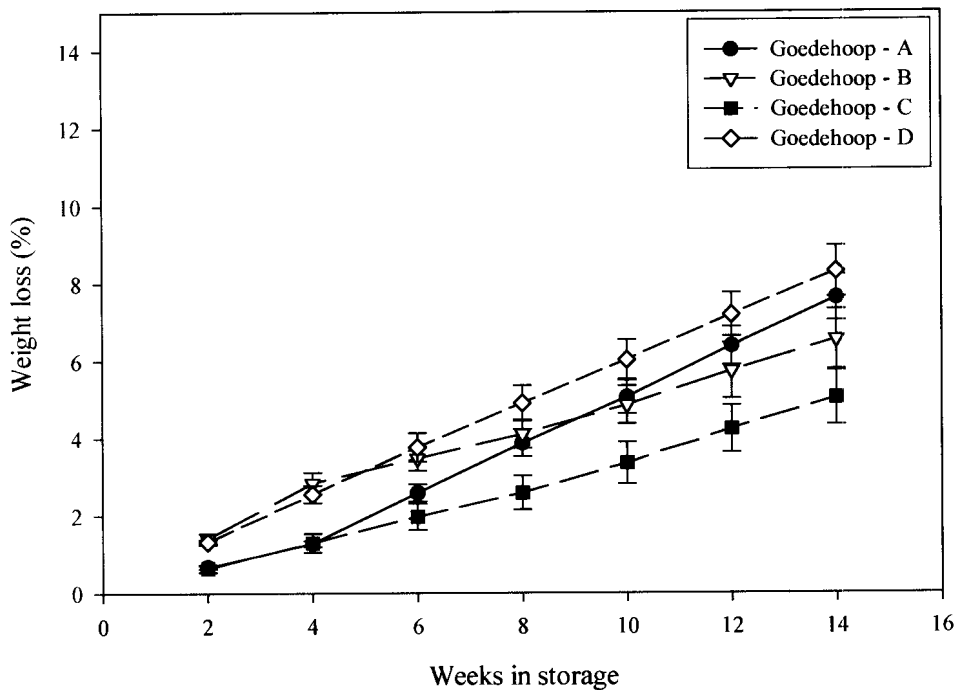


Fig.3.3.21. Weight loss (%) during the 14-week storage period in 1997 for different storage regimes (A – 4 weeks at 4.5°C, 6 weeks at 11°C, 2 weeks at room temperature; B – 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), from 2 production areas and 3 producers (Western Cape - Goedehoop; Eastern Cape - Gamtoosrivier and Sondagsrivier). Data values represent means  $\pm$ SD of 3 replicates of 3 fruit samples.

It is also evident that the lower shipping temperature did not have the same effect as seen with the TSS where it influenced the change more than the storage temperature (Table 3.3.2). Since the storage period was longer than the shipping period, the weight loss during this time was higher because of the higher temperature the fruits were exposed to. The differences between the storage regimes were highly significant (1%) until the eighth week in storage. The treatment with a shipping and storage temperature of 4.5°C (C) had the lowest weight loss, while treatment D, which had the highest (11°C) shipping and storage temperatures, had the highest weight loss (Table 3.3.2). The relative humidity in the storage chambers could also have influenced these weight losses.

The differences in weight losses were large between the two years (Table 3.3.2). In contrast to 1997 the weight loss in 1996 was almost double and might be related to problems with relative humidity

(RH) encountered in the higher temperature storage rooms. As low RH in storage causes an increase in weight loss, it might have caused the difference between the two years (Davis and Albrigo, 1994). Because all the fruit was stored at 11°C for 4 weeks, simulating the shipping period, the fruit might already have been affected by the lower RH in the 11°C storage rooms. The higher RH at the lower temperature (4.5°C) could not reverse this influence on the total percentage weight loss. Grierson and Ben-Yehoshua (1986) found that the rate of water loss during the first few days after harvest was much higher than the rest of the storage period because the peel was fully turgid before the fruit reached storage temperature. Even small changes in RH significantly (5%) affected the rate of weight loss. When grapefruit was stored for 20 weeks at 12°C with a RH of 90%, the weight loss was 6.8% but it increased to 12.4% when the RH was 80% (Grierson and Ben-Yehoshua, 1986). In 1997, the RH problems were handled by raising the RH from  $\pm 60\%$  to  $\pm 70\%$ . The RH of  $\pm 70\%$  was still not ideal, but affected strongly the total percentage weight loss in 1997 (Fig.3.3.20, Fig.3.3.21, Table 3.3.2).

Table 3.3.2. Comparison between the total percentage weight loss of all the treatments in both 1996 (A – 4 weeks at 11°C, 4 weeks at 4.5°C, 2 weeks at 8°C, 2 weeks at room temperature; B - 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature) and 1997 (A – 4 weeks at 4.5°C, 6 weeks at 11°C, 2 weeks at room temperature; B – 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature)(Statistics are not available)

Year	Origin	Treatment	Shipping temperature (°C)	Storage temperature (°C)	Weight loss %
1996	Ngonini	A	11	4.5, 8	13.06
		B	11	8	11.33
	Goedeheop	A	11	4.5, 8	11.00
		B	11	8	13.26
	Franschoek	A	11	4.5, 8	14.55
		B	11	8	13.40
1997	Gamtoosrivier	A	4.5	11	2.32
		B	11	4.5	5.04
	Sondagsrivier	A	4.5	11	3.24
		B	11	4.5	6.12
	Goedeheop	A	4.5	11	6.43
		B	11	4.5	5.76
		C	4.5	4.5	4.22
		D	11	11	7.26

### 3.3.1.4 . Maturity

Soft citrus are graded for over-maturity signs like loose skin and loose fruit segments. This normally gives an indication of the storage capacity of the fruit, because fruit with signs of over-maturity is softer than required and easily gets damaged in storage. An over-maturity grade of less than 3 is needed for the export marketing of South African soft citrus types (Van Wyk, 1996). A grade of more than 3 for over-maturity only developed in the later stages of storage and was not very temperature dependant (Fig.3.3.22).

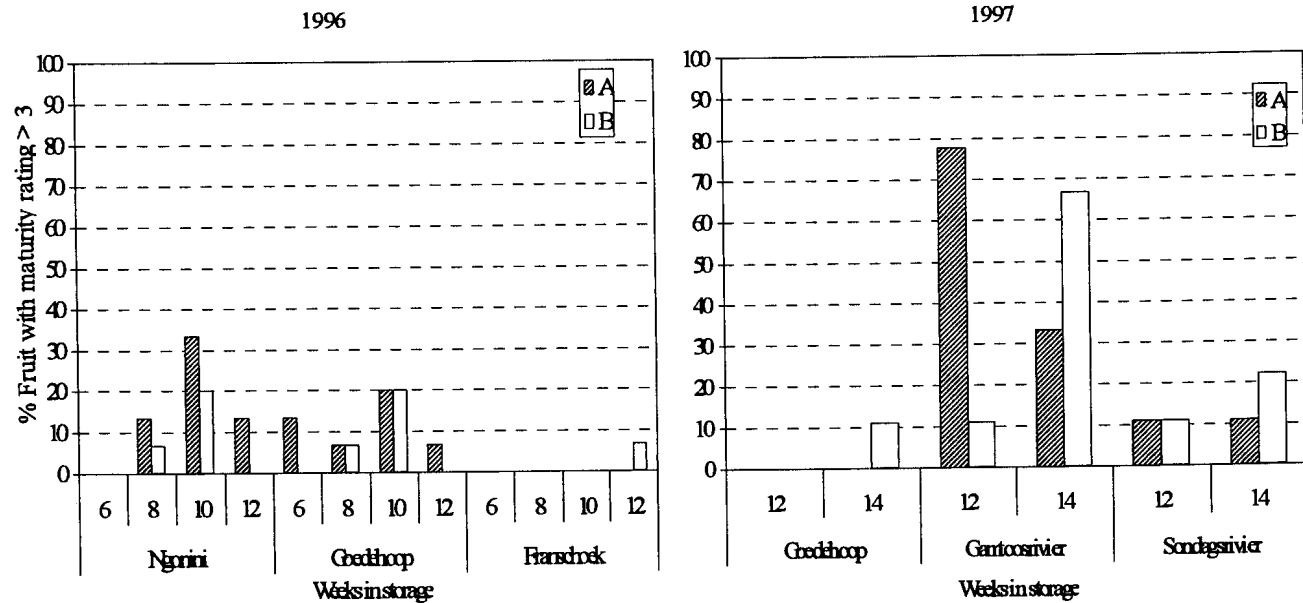


Fig. 3.3.22. % Fruit in maturity grades of >3 from 2 production areas and 3 producers (Swaziland - Ngonini; Western Cape - Franschoek and Goedehoop) after the 12 week storage period in 1996 at two different storage regimes (as described in Fig.3.3.1, A: 11°C, 4.5°C, 8°C, RT; B: 11°C, 8°C, RT) and from 2 production areas and 3 producers (Western Cape - Goedehoop; Eastern Cape - Gamtoosrivier and Sondagsrivier) in 1997 after the 14 week storage period in 1997 at two different storage regimes (as described in Fig.3.3.4, A: 4.5°C, 11°C, RT; B: 11°C, 4.5°C, RT).

There were significant differences between the influences caused by the different production areas / producers in both years. In 1996, fruit from Franschoek (Western Cape) stored at treatment B only showed signs of over-maturity after 12 weeks in storage, while fruit from the other production area / producers developed more fruit with over-maturity signs in both treatments from the sixth week and onwards. In 1997, fruit from Goedehoop (Western Cape) only developed 12 % fruit with over-maturity signs after 14 weeks in storage, while fruit from the other production area developed more signs of over-maturity after 12 weeks in both storage treatments. Although from the same production area (Eastern Cape), fruit from Gamtoosrivier developed a much higher percentage fruit with signs of over-maturity than the fruit from Sondagsrivier. This could be attributed to the fact that the fruit from the different producers was probably of different physiological ages. In 1996, 10-20% of the fruit had

over-maturity than the fruit from Sondagsrivier. This could be attributed to the fact that the fruit from the different producers was probably of different physiological ages. In 1996, 10-20% of the fruit had over maturity signs from 6-8 weeks and in 1997, 10% of the fruit had over maturity signs from 14-16 weeks. This comparison could not be explained, as the exact physiological ages of the fruit were not known. Harvesting dates and dates of fruit set are necessary to explain the difference.

### 3.3.2. Occurrence of rind breakdown in response to the different storage conditions

The severity of rind breakdown as physiological disorder at the end of a 14 week storage period can clearly be seen in Fig. 3.3.23.

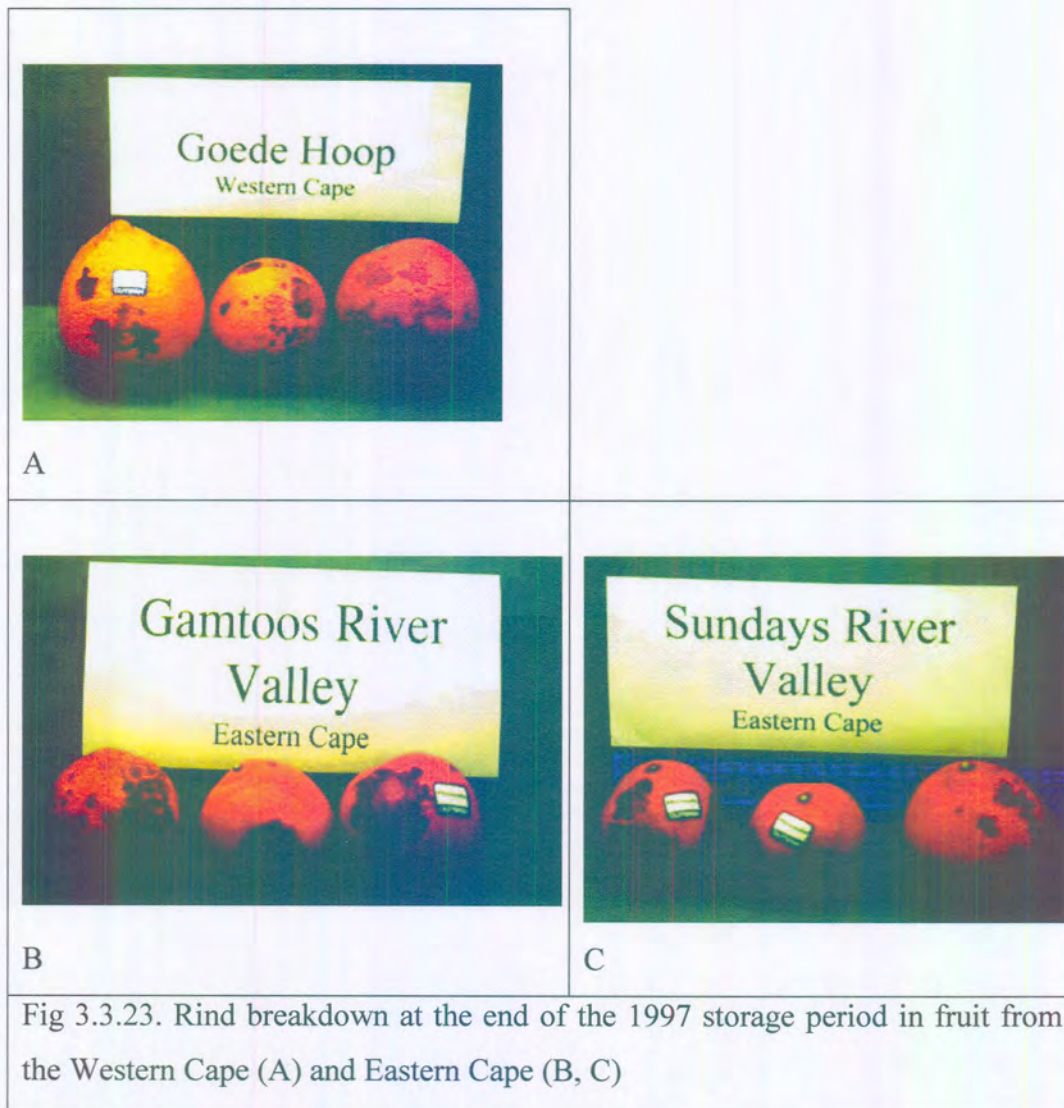


Fig 3.3.23. Rind breakdown at the end of the 1997 storage period in fruit from the Western Cape (A) and Eastern Cape (B, C)

In 1996, the occurrence of RB was very little compared to 1997 and only fruit from Franschoek developed unmarketable fruit (more than 10 marks per fruit)(Fig.3.3.24). Rind breakdown seemed to

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develop during the later stages of storage, as it only started developing after 6 to 8 weeks in 1996 and 1997. The only earlier development of RB was in fruit from Goedeheop (Western Cape) in 1997 when RB started developing from week 2 (Fig.3.3.24). There was no significant difference between treatments in 1996 and 1997. The only significant difference at 5% was observed between the producers in 1996 at week 6.

The development of RB in category 1 (which is less than three marks per fruit) was the most across all treatments in fruit from all the producers in both years. In 1996, treatment B resulted in a higher percentage fruit with RB in category 1 in all three producers. The RB development in category 2 was higher in treatment A in fruit from Franschoek (Western Cape), a little higher in fruit from Goedeheop (Western Cape) and lower in fruit from Ngonini (Swaziland) compared to storage regime B (Fig.3.3.24). After the initial treatment of 11°C, treatment A had a lower temperature treatment of 4.5°C, while fruit was stored at 8°C in treatment B. The fruit was kept at 11°C for the first 4 weeks in both treatments. During this time no RB developed. In 1996 the development of unmarketable fruit was more due to the production area / producer, as only fruit from Franschoek had RB to this extent (70 – 80%) in treatment A and B (Fig.3.3.24). The occurrence of RB in 1996, clearly showed that the producer and production area had the biggest influence on the RB development and the occurrence of RB was the highest in fruit from the Western Cape (Franschoek and Goedeheop) compared to the other production area (Swaziland).

In 1997, the development of category 1 and 2 RB were the highest in fruit from Sondagsrivier (Eastern Cape) (Fig.3.3.24) during treatment B. The amount of unmarketable fruit was also higher in fruit from Sondagsrivier than fruit from Gamtoosrivier (Eastern Cape). These fruit also had a higher weight loss during the storage period than the fruit in treatment A (Table 3.3.2). When focusing on the fruit from the Eastern Cape producers, treatment A seemed to have a more preserving effect. Treatment A had a lower shipping temperature (4.5°C) and a higher storage temperature (11°C). The same trend was observed in fruit from Goedeheop (Western Cape) but not as clear in the fruit from the Eastern Cape. Although it seemed that treatment A also had a preserving effect on the fruit from the Western Cape, this effect was much smaller when compared to the occurrence of RB in fruit from the Eastern Cape. The development of RB in category 1 was the highest in all four treatments (A,B,C,D) in fruit from Goedeheop (Fig.3.3.25). Although in 1997 there seemed to be a greater influence of the temperature of the storage treatments it was still not significant (Fig.3.3.24).

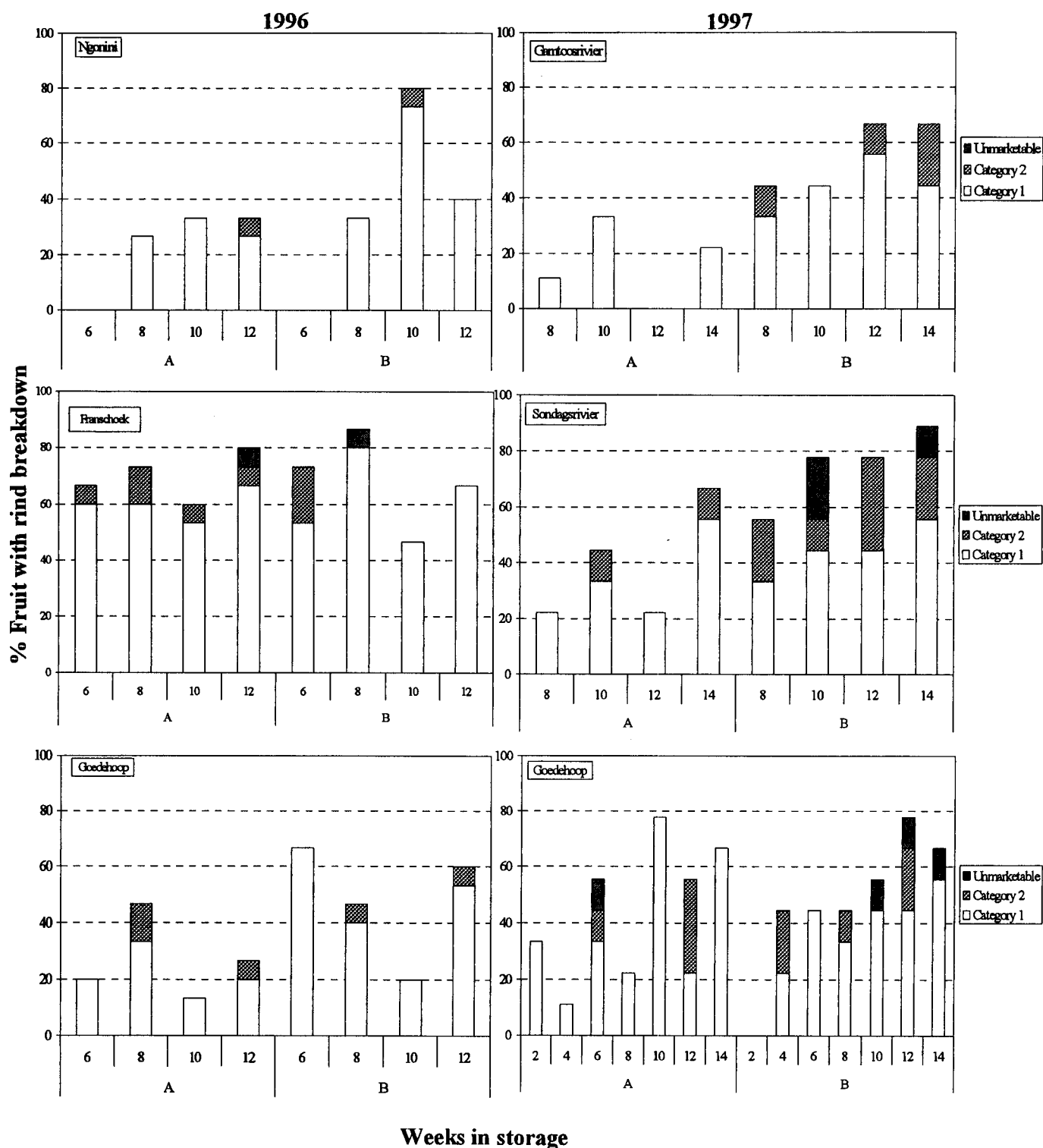


Fig. 3.3.26. Rind breakdown development in fruit from 2 production areas and 3 producers (Swaziland - Ngonini; Western Cape - Franschoek and Goedehoop) after the 12 week storage period in 1996 for two different storage regimes (A – 4 weeks at 11°C, 4 weeks at 4.5°C, 2 weeks at 8°C, 2 weeks at room temperature; B - 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature) and from 2 production areas and 3 producers (Western Cape - Goedehoop; Eastern Cape - Gamtoosrivier and Sondagsrivier) in 1997 after the 14 week storage period in 1997 for two different storage regimes (A – 4 weeks at 4.5°C, 6 weeks

at 11°C, 2 weeks at room temperature; B – 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), in 3 RB categories (Category 1 represents marks/fruit >0 and <3, suitable for export; Category 2 represents marks/fruit >3, suitable for local distribution and unmarketable fruit). Data values represent % fruit in each category.

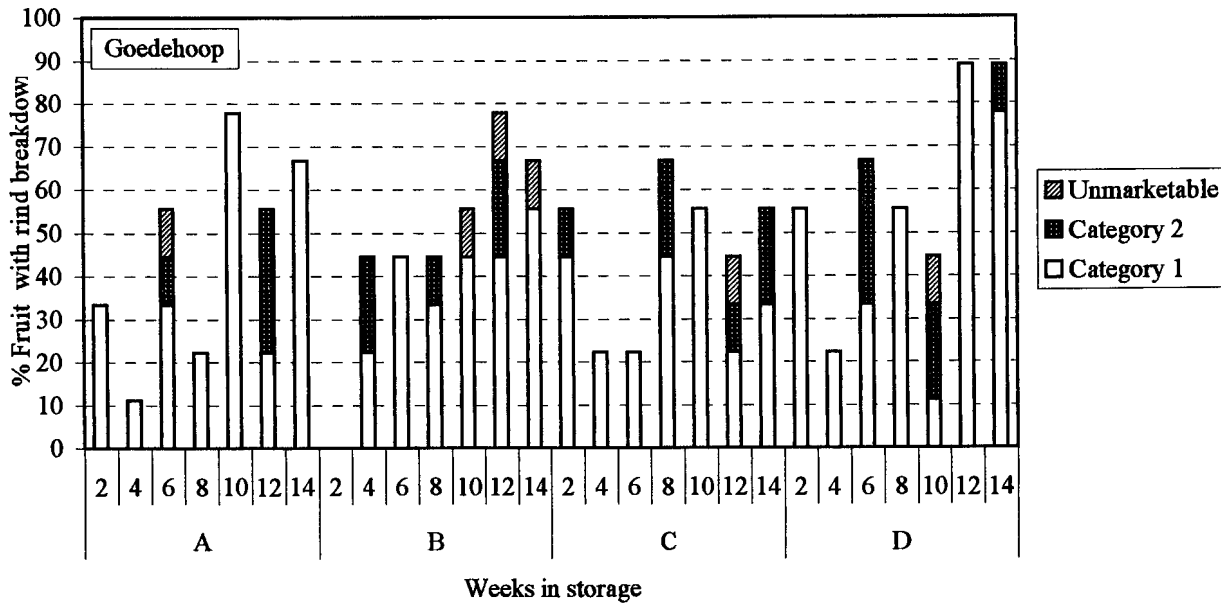


Fig.3.3.25. Rind breakdown development in fruit from Goedehoop (Western Cape) during the 14-week storage period in 1997 for different storage regimes (A – 4 weeks at 4.5°C, 6 weeks at 11°C, 2 weeks at room temperature; B – 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), in 2 RB categories. (Category 1 represents marks/fruit >0 and <3, suitable for export; Category 2 represents marks/fruit >3, suitable for local distribution). Data values represent % fruit in each category.

In 1997 the fruit with RB was counted at the end of the storage period and the percentage RB was determined from all the fruit not used in the other parameter determinations. As described by Chun et al. (1988) the temperature effect disappeared when fruit was kept at ambient temperatures. In fruit from Goedehoop (Western Cape) more or less the same incidence of RB was observed after 14 weeks in storage at both treatments (Fig.3.3.26). There was more RB fruit from a combination of category 1 and 2, than unmarketable fruit. The percentage unmarketable fruit was also higher in fruit treated with a higher shipping temperature at the end of the storage period. Du Toit Pelsler and Lesar (1994) reported the opposite when storing 'Satsumas' from Elands Rivier Valley, near Nelspruit. They found that the RB occurrence was the least when the fruit were "shipped" at 11°C (3 weeks) and then stored for 3 weeks at 2°C. The total percentage weight loss was also higher in the treatment with the higher shipping temperature (B) in fruit from the Eastern Cape (Gamtoosrivier and Sondagsrivier) but not in



fruit from the Western Cape (Table 3.3.2). As the differences were not significant, conclusive determinations cannot be made. Even though the percentage loss due to RB was relatively low, a 10% loss of all the fruit on the overseas market might result in a major economic loss. The problem with development of RB later in storage would be that the fruit would already have been shipped to the overseas markets.

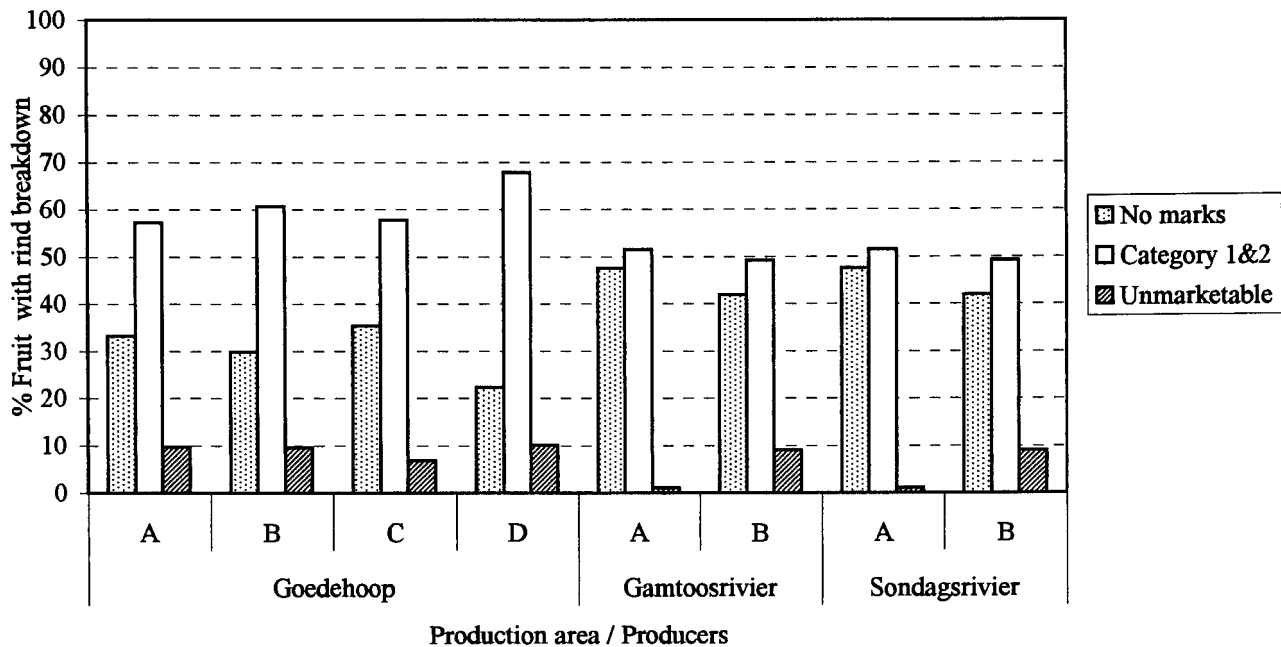


Fig.3.3.26. RB at the end of the 14 week storage period for fruit from all three producers in 1997 for different storage regimes (A – 4 weeks at 4.5°C, 6 weeks at 11°C, 2 weeks at room temperature; B – 4 weeks at 11°C, 6 weeks at 4.5°C, 2 weeks at room temperature), in 3 RB categories. (Category 1 represents marks/fruit >0 and <3, suitable for export; Category 2 represents marks/fruit >3, suitable for local distribution and unmarketable fruit). Data values represent % fruit in each category.

One of the problems with RB is that it is not predictable (Van Rensburg et al., 1995). This means that there is no way of predicting if fruit will develop RB in storage after shipping to the overseas markets. If there were physiological parameters that are correlated to the incidence of RB, it would be helpful to stop the fruit at risk from being exported. Unfortunately, in both years, the development of RB on the fruit was not correlated at  $P=0.05$  with any of the other parameters over all the weeks of storage. The correlations that existed did not follow a clear pattern.

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## 4. Conclusions

In 1996 and 1997 the main variance in most of the parameters ( $F < 0.001$ ) between the producers was highly significant (1%). Storage temperatures had very little influence in 1996 and 1997. Peel thickness and percentage juice appeared to be mostly determined by the fruit size the different producers supplied. Significant differences (5%) across all the parameters were observed between fruit from the different production areas, while fruit shape, percentage juice, and occurrence of rind breakdown did not differ significantly. In 1996, the weight loss developed in storage only showed a significant difference (5%) after 8 weeks in storage. In 1997, there was no clear pattern, but the interaction between producers and storage temperatures differed significantly. Focusing on the producer from the Western Cape, there was a significant difference between storage temperatures, confirming that citrus fruits stored at a higher temperature (11°C) will lose moisture faster than those stored at a lower temperature (4.5°C). In the fruit from the Eastern Cape (Gamtoosrivier and Sondagsrivier) the percentage weight loss seemed to be linked to the development in RB, but this was not in fruit from the Western Cape (Goedehoop). A conclusion on the interaction between RB and weight loss in storage would only be possible after a repetition of the experiment with fruit from the same producers stored at the same temperatures as in 1997. Occurrence of RB seemed to be slightly increased when the "shipping" temperature was 11°C and the storage temperature 4.5°C. Even with the higher moisture loss, there were no significant differences in RB between the fruit stored at the different temperatures, apart from the difference of the producers, indicating that changes in quality of 'Minneola' fruit might be mainly determined by the production areas / producers in addition to storage temperatures of 4.5°C and 11°C. With the rate of over-maturity developing in storage, it is quite possible that the fruit might have been stored too long. Therefore, the marketing period for soft citrus types is apparently too long and problems with physiological ageing can be abundant at this storage length.

When doing a detailed study to determine if production area is the prime reason for rind breakdown, data concerning the weather (daily minimum and maximum temperatures and rainfall), irrigation, and other cultivation practices (for instance hormonal application and fertilization) should be obtained from the producers. To make a recommendation to the growers at this stage would not be possible, as more information is needed on the climatic conditions, cultivation practices and tree ages to be able to explain why there are so many differences between the producers. The physiological age of the fruit is also important. More information is thus needed on the growing conditions of the trees, the time of full

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bloom, the time of fruit set and the harvest date. The humidity during storage, mineral nutrition, soil moisture, tree vigor and handling after harvest may also affect the occurrence of rind breakdown as it influences other physiological disorders. Another factor that could be important is the ethanol content of the air in the storage rooms as a build-up in ethanol increases the susceptibility to chilling injury.

An early warning system is still important to the exporters. As none of the measurements taken could be correlated with the incidence of rind breakdown, other measurements like ethanol and acetaldehyde content of the peel and fruit juice should be measured in a storage trial with the same temperature regimes. The search for such a measurement should still be continued, to be able to predict the incidence of RB. Also the incidence of rind breakdown was affected by the production year and area. Rind breakdown also seemed to be influenced by the length of the storage period, as the longer the storage period, the higher the incidence of rind breakdown.

An anatomical study of the rind should be done to show if there are morphological differences between the two physiological disorders, chilling injury and rind breakdown. Such a study would also help in explaining the way rind breakdown develops. Since moisture has an effect on the occurrence of rind breakdown, a future trial with fruit from moisture stressed trees would also be helpful in explaining if moisture stress has an increasing effect on rind breakdown or not.

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