Horticultural manipulation techniques to improve yield, fruit size and quality in 'Wai Chee' litchi (*Litchi chinensis* Sonn.)

by

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DECLARATION

I, Izak J Froneman, the undersigned, hereby declare that the work contained in this dissertation is my own original work and that I have not previously in its entirety or in part submitted it at any university for a degree.

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ABSTRACT

In South Africa, a shortage of litchi cultivars to expand the very short harvesting period has seriously threatened the viability of the local export orientated Litchi Industry. Only two cultivars have dominated commercial plantings for more than a century, namely 'HLH Mauritius' and 'McLean's Red'. The marketing period of litchis from South Africa coincides with large export volumes from competitor countries such as Madagascar, resulting in lower returns for local growers. To address this situation, the late season cultivar 'Wai Chee' was imported amongst others from Australia. 'Wai Chee' is harvested at a time in South Africa when there is a gap in worldwide litchi production, making the cultivar potentially very profitable. However, its potential and subsequent use in the industry is affected by small fruit size and questionable internal quality. As the South African Litchi Industry is export orientated and the qualities of 'HLH Mauritius' fruit currently dictate export requirements, solutions need to be found to improve fruit size and fruit quality in 'Wai Chee' litchi.

In this study, a number of horticultural manipulation techniques were investigated with the aim of enhancing yield, fruit size and quality in 'Wai Chee' litchi.

The use of chemical applications of foliar nutrients and plant growth regulators were found to improve certain fruit characteristics in 'Wai Chee'. Foliar nutrient applications of nitrogen, potassium and calcium during the early stages of fruit set and -development improved fruit set and subsequently yield, and also increased fruit mass, fruit size and flesh mass. Treatments with potassium nitrate (KNO_3), calcium nitrate ($CaNO_3$) and calcium metalosate proved to be the most enhancing nutrient applications.

Applications of synthetic auxins and auxin-like substances during the 2-3g stage of fruit development improved fruit size, fruit mass and flesh mass in 'Wai Chee'. The combination treatment of Tipimon® (2,4,5-TP), applied at the 2-3g stage, followed by Maxim® (3,5,6-TPA) a week later, yielded the best results in this regard.

With biological practices, pollination was found to have an influence on litchi tree- and fruit characteristics. Pollen source proved to have an influence on fruit set and fruit retention at harvest in 'Wai Chee'. Initial fruit set was lower when using cross-

pollination compared to the use of self-pollination in female flowers of ‘Wai Chee’, whereas final fruit retention was higher with the use of cross-pollinators when compared to retention of fruit with self-pollination. Although some beneficial effects with different pollen donors on fruit characteristics were observed, these effects were not significant, and would therefore necessitate further investigation. Pollen donor effect on quality parameters such as Titratable acid (TA)- and Total Soluble Solid (TSS) content of fruit was not significant.

Cultivar differences regarding fruit characteristics and maturation rate were detected with the use of cultural practices such as bunch covering materials in ‘HLH Mauritius’ and ‘Wai Chee’. Beneficial effects on fruit size were obtained with thicker covering materials with nominal mass of 70 and 80 g/m^2 respectively on ‘HLH Mauritius’, while with ‘Wai Chee’, thinner covering materials (60 and 65 g/m^2) showed enhancing effects. Maturation rate was significantly delayed only on ‘Wai Chee’ with the use of thicker covering materials (70 and 80 g/m^2). Differences in colour were detected amongst different covering materials, but these should be verified with chromameter technology. Covering of fruit bunches for better fruit size and a later harvest date would, especially for ‘Wai Chee’ as a late season cultivar, be beneficial, since better prices are realised towards the end of the season.

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CHAPTER 1

1 GENERAL INTRODUCTION

1.1 PROBLEM STATEMENT

The Litchi (*Litchi chinensis* Sonn.), which belongs to the Sapindaceae or soapberry family, originated in the subtropical areas of southern China, northern Vietnam and the Malay Peninsula (Groff, 1921; Menzel & Simpson, 1990). Trees were taken to most of the tropical and subtropical world during the last 400 years, but the crop is not widely grown because it does not flower and crop successfully over a wide range of climates (Joubert, 1986; Stern & Gazit, 1999, 2000). Production problems preventing litchis from becoming a major horticultural crop include irregular flowering, poor fruit retention, alternate bearing and small fruit size (Batten, 1986; Stern & Gazit, 2000; Menzel, 2001).

The litchi is one of the most environmentally sensitive of the fruit tree crops (Menzel, Watson & Simpson, 1988). It is adapted to the tropics and warm subtropics between 13-32° N and 6-29° S. In the Northern Hemisphere, litchis are available from March to August, while in the Southern Hemisphere fruit are harvested from October to March. The litchi requires cool to cold winters and warm to hot summers accompanied by high relative humidity in order to flower and crop successfully (Menzel, 1983; Batten & McConchie, 1995; Huang, Subhadrabandhu, Mitra, Ben- Arie & Stern, 2005). Litchis are cultivated for their popular fruit and they have a long history of cultivation in China and throughout much of South-east Asia. The demand is for large, bright red fruit with small seeds and crisp, sweet flesh (Menzel & Simpson, 1990).

In South Africa, only two cultivars have dominated commercial plantings for more than a century, viz. 'HLH Mauritius' and 'McLean's Red'. This situation has led to a very short marketing period that also coincides with large export volumes from competitors such as Madagascar, resulting in lower returns for local growers. The late season cultivar 'Wai Chee' was imported amongst others from Australia in the early 1990's to address the limited cultivar range and subsequent short harvesting season. 'Wai Chee' is harvested at a time in South Africa when there is a gap (months 2-4) in worldwide litchi production, making the cultivar potentially very profitable (Table 1).

Table 1 Litchi Supply calendar

Month	10	11	12	1	2	3	4	5	6	7	8	9
China							■	■	■	■		
Thailand								■	■	■		
Vietnam								■	■	■		
Taiwan									■	■	■	
Australia		■	■	■	■							
India								■	■	■	■	
Mauritius		■	■	■								
Reunion		■	■	■								
Madagascar		■	■	■	■							
South Africa			■	■	■							
Zimbabwe			■	■	■							
USA									■	■		
Israel										■	■	■

Although the cultivar proved to be precocious and high yielding, it's potential and subsequent use in the industry is affected by small fruit size and questionable internal quality with a poor TSS: acid ratio. As the South African Litchi Industry is export orientated and the qualities of 'HLH Mauritius' fruit dictate export requirements, solutions need to be found to improve fruit size and -quality in 'Wai Chee' litchi.

The South African Litchi Industry is export orientated, with exports amounting to four million tonnes of which 95% of the crop is sent to EU markets (PPECB Export Directory, 2006). It is therefore important to ensure profitable yields with high quality, marketable fruit. Since the 'Wai Chee' cultivar was released to the Industry for commercial utilization only recently, it is important to ensure the optimum production of the cultivar. Although the cultivar is precocious and capable of producing exceptional yields in high-density plantings, average fruit size remains relatively small at 16-18g when compared to the size of 'HLH Mauritius' fruit (22-24g).

1.2 POSSIBLE SOLUTIONS AS INDICATED BY LITERATURE

A literature review indicated that a number of horticultural manipulation techniques exist that may have a beneficial effect on litchi yield, fruit size and –quality. Different chemical-, biological- and cultural practices were subsequently considered as possible solutions:

Growth regulators

Considerable work has been done in recent years on the improvement of litchi fruit size by the application of different synthetic auxins at different stages of fruit development. It was shown by Stern & Gazit (1997) that it is possible for litchi trees to carry more fruit to maturity than a high fruit drop rate might imply. A high crop load did not affect future productivity, tree growth or fruit size, which is a known and negative phenomenon in many deciduous and tropical crops. Synthetic auxins have been used to increase litchi yield through higher fruit retention and also to improve size of fruit in Israel. Maxim® (3,5,6-trichloro-2-pyridyl-oxyacetic acid), a synthetic auxin known for improving fruit size and -weight in citrus and litchi, has shown satisfactory results (Agusti, Almela, Juan, Primo- Millo, Trenor & Zaragoza, 1994; Agusti, El-Otmani, Aznar, Juan & Almela, 1995; Stern, Stern, Miller, Xu & Gazit, 2001; Stern, Stern, Harpaz & Gazit, 2000). The use of two synthetic auxins namely Tipimon® (2,4,5-trichlorophenoxypropionic acid) and Maxim® is done commercially in Israeli litchi orchards and is considered as standard practice (Stern & Gazit, 2000).

Ethylene is one of several plant growth regulators that influence growth and developmental processes including ripening and senescence of fruit (Abeles, Morgan & Salveit, 1992). The use of ethylene inhibitors as a tool to control ethylene production on a large number of horticultural crops has been well documented (Serek, Sisler & Reid, 1994; Fan & Mattheis, 1999; Ku & Wills, 1999; Macnish, Joyce, Hofman & Simons, 1999 and Hofman, Jobin- Décor, Meiburg, Macnish & Joyce, 2001). Preliminary research and test results with Harvista®, a product developed by the Rohm and Haas Company in the USA, showed that harvest can be selectively delayed to allow time for additional fruit growth and colour development. Aminoethoxyvinylglycine, also known as AVG, is another product that blocks ethylene production in plants. Studies by Autio & Bramlage (1982), Bramlage, Greene, Autio & McLaughlin (1980) and Halder-Doll & Bangerth (1987) have shown that AVG, when applied as a pre-harvest spray, will suppress ethylene biosynthesis in apple fruit. This has led to the development of AVG as a commercial application (ReTain®) for harvest management and improvement of post harvest fruit quality in apples (Silverman, Petracek, Noll & Warrior, 2004).

Nutrition

Balanced nutrition is considered to be a very important factor in determining productivity and quality in litchi (Aylward, 2002; Singh & Babita, 2002). The main nutrients influencing fruit size include nitrogen, potassium, phosphorus and calcium. Nitrogen affects growth and productivity with deficiency symptoms on fruit manifesting in undersized fruit, with a reduced edible portion and poor eating quality (Thomas, Ferguson & Crane, 1995). Koen, Langenegger & Smart (1981) reported increased litchi fruit mass and sugar:acid ratio and a decrease in the proportion of malformed fruit after nitrogen application. Phosphorus is important in cell division and cell expansion and acts as an energy source (through ATP) for continued cell growth, whilst potassium aids in the movement of photosynthates from leaves into storage organs (fruit), which results in increased fruit size. Calcium is an important component of the cell membrane structure and is part of cell wall formation. Kift, Penter & Roets (2002) found that certain calcium products can improve fruit set and -size as well as yield in litchi.

Pollination

The advantages of cross-pollination to improve fruit set have been shown to exist in fruit trees (Bergh, 1968; Sedgley & Griffin, 1989; Degani, Goldring, Adato, El-Bastri & Gazit, 1990). The use of cross-pollination to improve fruit set, yield and fruit size in litchi was investigated by Stern, Gazit, El- Bastri & Degani (1993) in Israel. In trials performed in adjacent blocks of ‘Floridian’ (also known as ‘Brewster’) and ‘Mauritius’ litchi, the importance of cross-pollination in litchi was demonstrated. Almost all fruit on trees close to the second cultivar were hybrids. There was also a pronounced selective abscission in favour of hybrid fruit over self-pollinated fruit. The work also showed that there could be a positive relationship between hybrid percentage and yield in some cultivars, as a beneficial effect of cross- pollination by ‘Mauritius’ on ‘Floridian’ was observed. Furthermore, a significant correlation was found between pollen source and the mass of fruit and seed in both cultivars.

Cultural practices

The use of cultural practices such as girdling to improve flowering, fruit set, fruit retention and -size in litchi was mentioned by Menzel & Simpson (1987), Zhang, Yuan, Wang, Qiu & Li (1997) and Stern & Gazit (2003). However, as girdling has the potential to advance harvest time in litchis (Oosthuizen, 1993), which is not beneficial for a late cultivar such as ‘Wai Chee’, fruit bunch covering was rather chosen as a suitable cultural manipulation technique.

Fruit bunch covering is a method to improve litchi fruit colour, quality and size by using paper bags to cover fruit clusters, creating a favourable micro- climate inside the bag. Tyas, Hofman, Underhill & Bell (1998) in Australia, Sethpakdee (2002) in Thailand and Zhang *et al.* (1997) in China, all found that covering fruit clusters with paper bags have a beneficial effect on fruit colour and size with no negative impact on yield. Additional benefits include protection of fruit from insects, bats and birds, as well as delayed maturity of fruit.

1.3 OBJECTIVES OF THE STUDY

The objectives of the current study were as follows:

- to study the effects of some plant growth regulators, nutrient sprays and ethylene inhibitors on yield, fruit size and quality in 'Wai Chee'
- to test the significantly better performing chemicals and their combination in different programmes in the following year.
- to study the biological effect of pollen parent on litchi fruit set, fruit retention and - size in 'Wai Chee'
- to examine the effect of cultural practices such as covering of fruit clusters with paper bags on fruit size and quality in 'Wai Chee' litchi.

CHAPTER 2

2 LITERATURE REVIEW

2.1 DISTRIBUTION

The litchi (*Litchi chinensis* Sonn.) is a member of the Sapindaceae family, which includes its close relatives longan and rambutan (Joubert, 1986; Greer, 1996). It originates from the region between southern China and northern Vietnam, between 19° and 27° N, where wild trees can still be seen growing in elevated and lowland rainforests (Menzel, 1991; Menzel & Simpson, 1994; Oosthuizen, Froneman & Roe, 1995; Stern & Gazit, 2003). From these regions production has spread to many tropical and subtropical parts of the world including Taiwan, Thailand, India, Mauritius, Malagasy Republic, South Africa, Reunion, Israel, Florida, Hawaii, Mexico and Australia (Greer, 1996).

Litchis have a long history in Southeast Asia with Chinese records going back to about 2000 BC (Ochse, Soulle, Dijkman & Wehlburg, 1961; Lemmer, 2002). However, the distribution to other countries only commenced from about 1600 AD (Joubert, 1986; Menzel & Simpson 1994; Greer, 1996). According to Liu (1954) and Menzel, Watson & Simpson (1988), the litchi was introduced in countries such as Burma and India as well as to countries on the East Coast of Africa (Mauritius and Madagascar) by the end of the 17th century. It reached South Africa from the island of Mauritius around 1876 (Oosthuizen, 1993). From the 18th century distribution took place from India to glasshouses in England and France (De Villiers, 2002). From there it reached Florida and Hawaii in the late 19th century (Singh & Singh, 1954; Menzel *et al.*, 1988). It is thought to have reached Australia around 1854, brought by migrant Chinese mineworkers. From South Africa the litchi was introduced to Israel around 1930 to 1940 (Galan Sauco & Menini, 1989). Today, the litchi is also being cultivated in many other countries in Central and South America as well as in Africa, Asia, many Pacific islands and Spain (Fivaz, 1995). They are cultivated for their very popular fruit and the demand is for large, bright red fruit with small seeds and crisp, sweet flesh.

Commercial litchi production only takes place from 15°-35° Northern and Southern latitudes, and is usually found at low elevations in the subtropics and between 300 and 600 m in the tropical locations. The main commercial industries are centred in China, Taiwan, Vietnam, Thailand, India, Madagascar and South Africa (Wittmann, 2002). Smaller industries constitute Australia, Israel, Mauritius, Indonesia and the USA (Menzel, 2001). While litchi production does occur in the Northern Hemisphere, the litchi export trade is mainly from the Southern Hemisphere, with Madagascar and South Africa being the main exporters (Greer, 1996).

Production problems preventing litchis from becoming a major horticultural crop include irregular flowering, poor fruit retention, alternate bearing and small fruit size (Batten, 1986; Stern & Gazit, 2000; Menzel, 2001).

2.2 CLIMATIC REQUIREMENTS

The litchi is native to the subtropical parts of south-east Asia where it flowers and crops successfully only in a small range of climates. Because of its phenology, the litchi is one of the least adaptable of the tropical and subtropical crops (Menzel & Simpson, 1988). The range in climates in the main production areas of the world is very narrow for litchis compared with other tropical fruit such as citrus and banana that are grown across several climatic zones (Menzel, 2001). Commercial plantings can be found in frost-free areas between 15° and 35° latitude. Litchis are usually cultivated at low elevations in the subtropics and from 300-600 m in tropical locations, with cool or cold winters and warm to hot summers (Menzel, 1983; Menzel & Simpson, 1994). Fruit production is highest in warm, subtropical areas, while trees tend to grow vegetatively without producing flowers in moist equatorial zones with higher winter temperatures (Menzel & Simpson, 1988, 1994). Young trees are severely damaged or even killed at temperatures of between -2 and -3°C, with mature trees being more tolerant to low temperatures and only severely damaged at a temperature of -4 to -5°C (Storey, 1973). Litchi flowers are killed by frost and temperatures below 0°C (Joubert, 1970). Generally litchis are grown in areas with mean winter minimum temperatures below 15°C, mean summer maximum temperatures between 27 and 33°C, low rainfall during the winter (less than 25 mm) and higher rainfall during summer (more than 300 mm). The water requirement of the litchi is between 1270 and 1524 mm per annum and irrigation is necessary when the rainfall is less than this (Groff, 1944; Storey, 1973). Rainfall is generally highest in summer and least in winter or spring.

Shoot growth begins at 10°C, but both shoot and panicle growth is still slow at 15°C. Optimal shoot growth occurs between 24 and 30°C, with high rainfall and humidity also affecting growth positively. Vegetative flushes last for shorter periods and follow one another more rapidly at 25-30°C than at 15-20°C (Kift, 2002a).

Controlled temperature experiments done in Australia by Batten & McConchie (1995) showed that flower induction takes place when buds start growing under cooler conditions and not when buds are dormant. However, a period of dormancy is needed in autumn (April/May in South Africa) before floral induction occurs in young, growing buds during inductive winter conditions (June, July, August in South Africa). If trees are flushing vegetatively in April-May before winter so that the flush is still hardening off during winter, no flowering occurs. Terminal-flowering subtropical and tropical tree crops grow by recurrent flushing. When these trees flush, they must have a period when the flush hardens off before they can begin growing again. The period from the beginning of a flush to the next one can be seven weeks in summer in South Africa, while the period can be more than four months in winter. Consequently, if a flush starts in May it can be September before terminal buds can grow again and therefore no flowering will occur because temperatures are too high for floral induction.

Batten & McConchie (1995) suggested manipulating techniques such as pruning, irrigation and fertilisation in order to get buds to start growing under inductive winter conditions, stretching from June to August in Southern Hemisphere conditions. Menzel (1983) mentioned girdling, root pruning, fertilizer management and use of growth regulators as different means of influencing flower initiation. Applications of plant growth inhibitors like Ethephon-R that causes new flush abscission are also commercially used in South Africa to control undesirable autumn flushing of trees (Kift & Roets, 2002).

Glasshouse experiments with controlled temperatures further showed that ‘Tai So’ (or ‘HLH Mauritius’ in South Africa) flowers well at day/ night temperatures of 15/10°C, while only 50% of terminal branches flower at 20/15°C. In contrast, it was found that ‘Wai Chee’ flowers well at both 15/10°C and at 20/15°C day/ night temperatures, indicating that ‘Wai Chee’ is less dependant on lower temperatures for floral induction. According to Menzel & Simpson (1986), less vigorous cultivars are generally more precocious than vigorous cultivars at 20/15° C. The vigorous ‘Tai So’ cultivar forms leafy panicles at these temperatures, while ‘Wai Chee’ mostly forms leafless panicles. Leafless panicles with pure flowers are usually more fruitful than those with leaves (Kift, 2002b).

Under field conditions, good flowering of 'Tai So' occurs in areas where the mean winter minimum temperature drops below 15°C for three months, with temperatures below 12°C being more conducive to flowering. Day temperatures also influence floral induction during winter months. More consistent flowering is found in areas where day temperatures in winter fall below 20°C for a few weeks (Batten & Mc Conchie, 1995). Menzel, Rasmussen & Simpson (1989) found that flowering is not induced by low night temperatures of 10°C when accompanied by day temperatures of 30°C. Floral bud development is influenced by temperatures after flower induction (Kift, 2002a). Higher temperatures of 25-30°C reduce the number of female flowers in 'Tai So' after panicle emergence and some floral buds revert to vegetative growth (Menzel & Simpson, 1991). Areas with warm winter temperatures (> 20°C) are not suited for litchi production.

Litchi flowers open at approximately 10°C, with the optimum temperatures for pollen development, pollination and fertilization being between 19 and 26°C. Extended periods of temperatures above 30°C during anthesis can reduce fruit set. Continuous low temperatures, as well as cloudy, rainy and humid weather also negatively affect flower opening and pollination. Rain and overcast weather over several days at anthesis causes crop failure because both male and female flowers are damaged and pollinator activity is reduced (Wittmann, 2002). Extremely hot and dry weather conditions at this stage are also detrimental to fruit set, with normal fruit development ceasing when temperatures fall below 15°C (Kift, 2002a). Water stress at this stage needs to be avoided. As soon as flower panicles appear, trees should be watered to prevent drying and dropping of flowers, and to enhance fruit set. It was shown by Menzel and McConchie (1996) that water deficits at flowering and early fruit set can reduce initial fruit set by a third. In addition, it was also found that final fruit number may be reduced by two thirds compared to irrigated treatments, due to increased fruit cracking. Cell division in the pericarp takes place within 60 days after pollination and if too few cells are present due to stress, later cracking occurs during fruit growth, when the expansion capacity of the pericarp is exceeded. High temperatures accompanied by low humidity (less than 50%) can cause skin browning and fruit cracking during fruit development. In India, as much as 50% of the crop may be lost due to fruit cracking. These losses occur when the temperature rises above 38°C and the relative humidity is below 60% (Sharma & Ray, 1987). Optimum conditions for fruit development will be in a high rainfall area with high relative humidity and temperatures between 25 and 30°C. Such conditions will ensure optimal fruit growth with minimal damage through sunburn or cracking.

2.3 CULTIVARS

The litchi is indigenous to southern China where it has been cultivated for thousands of years and has undergone intensive selection (Menzel & Simpson, 1990). The litchi (*Litchi chinensis,* Sonn.) which belongs to the *Sapindaceae* or soapberry family, originated in the area between southern China, northern Vietnam and the Malay Peninsula. Litchi trees grow wild in abundance in elevated and low lying rainforests, especially in Guangdong and Hainan Island near northern Vietnam where litchi is one of the main species (Groff, 1921; Menzel & Simpson, 1990, 1991, 1994). Litchi is a dominant species in some of these forests and wild types may account for up to 50% of the virgin forest composition. Wild specimens resemble commercial varieties in appearance, but the aril of these specimens is relatively thin and acid (Menzel, Huang & Liu, 2005).

The first official scientific recording on litchi cultivation in China appeared in the second century BC, while unofficial records date back to 1766 BC (Menzel & Simpson, 1990). According to a 'litchi register' there were 16 cultivars in Guangdong by 1034 and 30 in Fujian province by 1059 (Anon, 1978). These figures rose to a hundred by 1076 in Guangdong and a similar number in Fujian in 1597. According to these antique scriptures Chinese growers could distinguish the best types for cultivation on the plains, hills or riverbanks by the second century BC. Propagation of the crop by these early pioneers was done exclusively by seed, until clonal propagation became available (air-layering in the 4th century and grafting in the 14th century AD) (Menzel & Simpson, 1991). Today there are more than 100 litchi cultivars in China, probably resulting from this early sexual reproduction by seed and the consequent selection of the best material by the early growers (Groff, 1921).

From China, the litchi became distributed to most of the tropical and subtropical world from about 1600 AD and is very popular in China and Southeast Asia, but is less known in Africa, America and the Middle East. Southeast Asia accounts for more than 95% of total world production at 2 000 000 tonnes (Menzel *et al.*, 2005). The major commercial industries are found in China, Taiwan, Vietnam, Thailand, India, Madagascar and South Africa. There are small commercial industries in Israel, Australia, Spain, the USA, Mexico, Mauritius and Reunion (Menzel, 2001). Despite the large number of litchi cultivars available in China, only about 15 are cultivated commercially in the litchi growing areas (Zhang *et al.*). In countries such as South Africa, Madagascar and Mauritius, commercial production is based on one or two main cultivars, leading to a very short production period.

In China, where the litchi originated, it is reported that the litchi has more cultivars than any of the other fruits. The most important cultivars in the litchi growing areas of Guangdong, Fujian and Guangxi are ‘Sum Yee Hong’ (‘Third Month Red’), ‘Tai So’ (‘HLH Mauritius’), ‘Chen Zi’ (‘Brewster’), ‘Souey Tung’, ‘Haak Yip’, ‘Fay Zee Siu’, ‘Kwai May’, ‘Wai Chee’ and ‘No Mai Chee’. More than 50% of plantings in Guangdong consist of the ‘Wai Chee’ cultivar. Because of late flowering, low temperatures of spring are avoided, leading to consistent yielding of this cultivar (Figure 1). Cultivars for export include ‘Sum Yee Hong’, ‘Fay Zee Siu’, ‘Haak Yip’, ‘Kwai May’, ‘Wai Chee’ and ‘No Mai Chee’ (Menzel, 2001).

Figure 1 Yield potential of the 'Wai Chee' litchi cultivar.

Commercial litchi industries in other countries are mostly based on only a few cultivars; almost all of Chinese origin. Commercial cultivars include ‘Tai So’ and ‘Wai Chee’ in Thailand, ‘Haak Yip’ in Taiwan, ‘Tai So’ or ‘HLH Mauritius’ in South Africa, Mauritius, Madagascar and Reunion, ‘Brewster’ and ‘Tai So’ in Florida and Hawaii and ‘Tai So’, ‘Bengal’, ’Kwai May Pink’ and ‘Wai Chee’ in Australia (Menzel & Simpson, 1990, 1991). In India and in southern Thailand, local seedling selections of Chinese cultivars are exploited. Seedling cultivars developed in the last 50 years and becoming increasingly important include ‘Kaimana’ (Hawaii), ‘Sah Keng’ (Taiwan) and ‘Salathiel’ (Australia).

Litchi cultivars can be described according to general appearance and tree shape, together with the shape, size and colour of the leaves, harvesting period and various fruit characteristics (Menzel *et al.*, 2005). The descriptors are discussed by the authors as follows:

The **harvesting time** usually lasts 5-10 weeks for a range of cultivars growing in any one location, with cultivars broadly classified as early-, mid- or late maturing. The order of harvest can vary from year to year depending on seasonal conditions, with some variation between different regions, presumably due to differences in environment or cultural practises.

Cultivars can also be identified by **tree size and shape** as well as **length and spread of branches**, but this may vary with weather and culture. For example, ‘Tai So’ is vigorous, with a spreading habit and sharp, weak crotch angles, while ‘Wai Chee’ is slow- growing, compact and dome-shaped.

Leaves vary in shape, size and colour. ‘Tai So’, for example, has glossy, large, dark green leaflets, curling upwards from the midrib. Leaflets of ‘Wai Chee’ are small, oval and curve upwards from the midrib and down towards the tip (Figure 2). New growth flushes are red with ‘Wai Chee’ and ‘Kwai May Pink’ and green-bronze in ‘Tai So’.

Fruit shape in litchi is very distinctive. ‘Kwai May Pink’ has round fruit, which is different from the heart shape of ‘Haak Yip’ or the egg shape of ‘Tai So’. The shoulders of fruit can be flat or smooth as in ‘Wai Chee’ and ‘Kwai May Pink’ or uneven as with ‘Souey Tung’ and ‘Bengal’. The tip of the fruit can be pointed as with ‘Bengal’, round like ‘Wai Chee’ (Figure 2) and ‘Kwai May Pink’, or blunt as in ‘Brewster’ and ‘Souey Tung’.

Figure 2 'Wai Chee' fruit and leaf characteristics.

Fruit skin or peel can be thin as in ‘Souey Tung’ and ‘Haak Yip’, or thick like with ‘Wai Chee’, ‘Kwai May Pink’ and ‘Bengal’. Skin colour may be bright red with ‘Bengal’, dull red in ‘Wai Chee’, purple-red in ‘Haak Yip’, orange-red in ‘Kwai May Pink’ or pink-red with ‘Brewster’. With skin segments at full maturity, variations can occur from smooth (‘Haak Yip’) to swelling with ‘Wai Chee’ or sharp-pointed as in ‘Kwai May Red’. The protuberances on each segment can be sharp as with ‘Tai So’, smooth as in ‘Haak Yip’ or sharp-pointed as in ‘Bengal’.

The **aroma, taste, juiciness and texture** can also be used to help with description. For example ‘Kwai May Pink’ is spicy, ‘Wai Chee’ is watery, ‘Kwai May Red’ is firm and ‘Bengal’ is very sweet.

Seed size can also be a cultivar indicator, although the trait can vary with season and orchard. ‘Salathiel’ fruit have small seeds, while ‘Haak Yip’, ‘Souey Tung’, ‘Wai Chee’ and ‘Bengal’ have few small seeds. The characteristic varies in ‘Tai So’ and ‘Kwai May Pink’.

2.4 CULTIVAR HISTORY IN SOUTH AFRICA

Litchi cultivation in South Africa stretches back to 1876 when the first plants arrived from the island of Mauritius. From the province of KwaZulu-Natal where the first litchis were introduced, trees spread to the eastern subtropical parts of Mpumalanga and Limpopo (Oosthuizen, 1993). Despite a cultivation history of more than a century, only two cultivars have dominated the South African litchi industry, namely 'HLH Mauritius' ('Tai So') and 'McLean's Red' (similar to 'Bengal'). The 'HLH Mauritius' cultivar is the only cultivar grown in all the production areas. 'McLean's Red', a late season variety, is restricted to the high altitude, cool areas of the Tzaneen area of the Limpopo Province. This situation has led to a very short marketing period of only 6-8 weeks in South Africa, including the Christmas holiday period. The South African crop is also produced at a time when competitors like Madagascar are exporting large volumes, resulting in lower returns for local growers. In the 1950's the South African litchi industry imported cultivars from countries such as India, Florida (USA) and Taiwan, but early observations indicated poor adaptability to South African conditions. Much confusion and many mistakes were also encountered in the identities of imported cultivars. It appeared that names for the same cultivars often varied across different areas and regions in their countries of origin. It later transpired that many inferior or wrongly named cultivars were imported. Only in the early 1990's were renewed efforts made to obtain cultivars with superior qualities from Australia. Cultivars such as 'Wai Chee', 'Haak Yip', 'Souey Tung', 'Kwai May Pink', 'Gee Kee' and 'Salathiel' were imported and established in evaluation blocks in different areas to determine their suitability for South African conditions. In 1999, 'Fay Zee Siu' (previously imported) and 'Wai Chee' were released to the South African litchi industry for commercial utilisation (Froneman, 1999). 'Fay Zee Siu', early maturing and with good fruit size and quality, was recommended for early production areas. 'Wai Chee', a late maturing variety with heavy crops, was recommended for late (cooler) areas to preferably stretch the production season to February/March. Although the cultivar is precocious and capable of producing exceptional yields in high density plantings, average fruit size remains relatively small (Froneman, Husselman, Rheeder, Human & Sippel, 2003).

2.5 FLOWERING, POLLINATION AND FERTILIZATION

The litchi inflorescence bears three different flower types (Figure 3) on the same tree (Joubert, 1985; Goren, Degani & Gazit, 1998; Kift, 2002). The first flowers that open are male flowers (M1) with 4-12 well-developed stamens, an aborted ovary and a degenerated pistil. When pollen of these flowers ripens, the anthers turn yellow and split open to release the pollen grains.

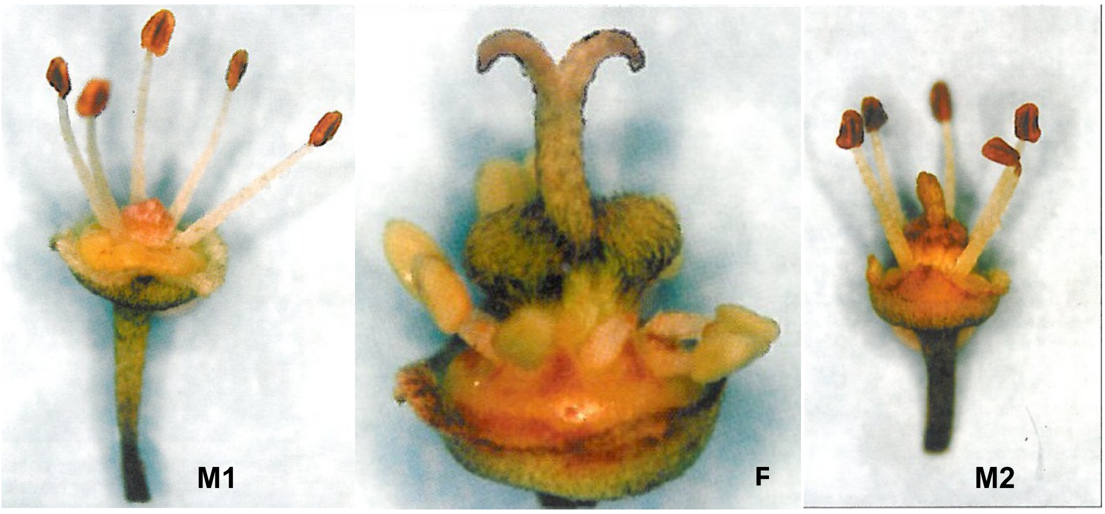


Figure 3 Different flower types on a litchi panicle. M1= First male flowers, F= Female flowers and M2= Second male flowers.

The second wave of flowers to open is the functionally female flowers. Female flowers have a well-developed stigma, style, ovary and stamens with very short filaments. Pollen sacs of these flowers never split open to release pollen and female flowers are only pollinated by pollen from the first or second wave of male flowers. The stigma of female flowers becomes receptive immediately after they have split pinnately and remain receptive for 3-4 days. The third wave of flowers to open is functionally male (M2) with well-developed anthers and rudimentary non-functional female parts (Figure 4). According to Stern & Gazit (2003) there is little overlap of the different flower types on the same inflorescence. The synchronization is also not perfect among inflorescences on the same tree and even less so between inflorescences on different trees of the same cultivar. It was further mentioned that in a single cultivar planting, each flowering wave usually lasts for 7-12 days, with two periods of overlap, continuing for 1-3 days each between the female flowers and each of the two male flowering waves. Litchi flowers are functionally unisexual on the same panicle and pollination can either occur by self- or cross- pollination. During the female flower stage, self- pollination is effected by pollen from the two partially overlapping male (M1 and M2) bloom waves on the same tree and cross pollination by pollen from a different cultivar. By planting two cultivars with female flowers coinciding with one of the male flowering stages of the other cultivar, pollination can be improved by ensuring the availability of pollen throughout the female flowering stage (Stern & Gazit, 2003). Successful pollination and fertilization are prerequisites for the set of a normal litchi fruit. Stern (1992) found non-pollinated flowers of two cultivars all abscised within four weeks, whereas about 10 per cent of the pollinated flowers had survived at the end of this period.

Figure 4 'Wai Chee' litchi with male 2 flowers.

Pollination with fertile pollen is essential for fruit set. Pollination rate is reported to be very slow at the start of the 'Mauritius' female bloom in Israel (Stern & Gazit, 1996). In Queensland, Australia, male flowers produced no pollen in the early 'Tai So' bloom in some seasons (Batten & McConchie, 1992). It was found by Stern & Gazit (1996) that M1 pollen is less fertile than M2 pollen. 'Mauritius' and 'Floridian' Male1 and Male 2 pollen was used to self-and cross-pollinate female flowers of both cultivars. Pollination with M2 pollen resulted in a significantly higher fruit set for both cultivars. It was found that the greater fertility of M2 pollen, coupled with the greater attractiveness of M2 flowers to the honeybee, make the M2 bloom more effective for pollination than the M1 bloom. Therefore, when a pollen donor is needed to increase the productivity of a certain litchi cultivar, such a pollen donor should preferably be selected where the M2 bloom overlaps with the female bloom of the other cultivar.

Studies on the effect of self- and cross- pollination showed that the litchi flowering pattern tends to promote cross- pollination (Stern & Gazit, 2003). However, the partial overlap between the female flowering stage and the two male (M1 and M2) stages enables pollination among flowers on the same tree, or among trees of the same cultivar, thereby providing an opportunity for self-fertilization (Joubert, 1986; Stern & Gazit, 1996). Litchi is considered to be self-fertile, since single cultivar litchi orchards are capable of producing good yields, as in the case of 'Brewster' in Florida, 'HLH Mauritius' in South Africa, 'Wai Chee' in China and 'Haak Yip' in Taiwan (Campbell & Malo, 1968; Joubert, 1986; McConchie & Batten, 1989; Batten & McConchie, 1992). Hand-pollination studies in South Africa (Fivaz & Robbertse, 1995), Israel (Stern & Gazit, 1998), and Australia (Batten & McConchie, 1992) confirmed the self-compatibility of several litchi cultivars. The use of self- or foreign pollen had in most cases no significant effect on fruit set. However, the study in Australia showed that some pollen donors can increase fruit set in certain cultivars.

Determination of the pollen parent's identity by isozyme analysis again confirmed the self-fertility of 'Floridian' and 'Mauritius' in commercial orchards in Israel (Stern, Gazit, El-Bastri & Degani 1993; Degani, Stern, El-Bastri & Gazit, 1995). However, these studies also demonstrated the potential of cross-pollination in litchi. Almost all fruit on trees close to a second cultivar were hybrids. In addition, there was a pronounced selective abscission in favour of hybrid fruit over self-pollinated ones. About a month after fruit set, the hybrid rate in a 'Mauritius' plot was 30 per cent, increasing to 76 per cent at harvest (Stern & Gazit, 2003). The authors stated that the survival percentage of the hybrid fruitlet may derive from inbreeding depression. Sedgley & Griffin (1989) found that self-pollinated progeny often have less vigorous embryos than cross-pollinated progeny in fruit crops.

'Mauritius' yield was not correlated with proximity or hybrid percentage to 'Floridian' trees in Israel, indicating that cross-pollination by 'Floridian' has no advantage over self-pollination. In contrast, 'Floridian' yield was higher close to 'Mauritius' and a significant positive relationship was observed between the yield and hybrid percentage or proximity to 'Mauritius', indicating a beneficial effect of cross-pollination by 'Mauritius' (Degani *et al.*, 1995). Therefore, the benefit of cross-pollination in litchi seems to vary with cultivar.

2.6 FRUIT SET AND DEVELOPMENT

In litchi, a double fertilization process occurs 2-3 days after pollination (Zhang *et al.*, 1997). Female litchi flowers possess a bifurcate ovary with two ovules. When one of the two ovules is fertilized, either a normal or a degenerated seed will develop (Stern, Eisenstein, Voet & Gazit, 1997). According to Menzel (1984) and Galan Sauco & Menini (1989), fruit set in litchi ranges from less than 1-50% depending on the year, tree, cultivar and environmental conditions.

Kift (2002b) and Joubert (1985) mention three stages of fruit development. The first stage mainly consists of embryo, fruit skin and seed coat development. The leathery peel of the fruit (or the pericarp) consists of three layers: the exocarp, the mesocarp and the endocarp. A longer duration of cell division during this development period leads to a larger pericarp and a larger fruit (Wang, Wei, Gao & Huang, 2000). Differences in fruit size amongst cultivars were related to variations in the number of cells rather than to their final size (Li, Huang, Huang & Zhou, 2002). This stage occurs in the initial fifty days after pollination, resulting in a heart-shaped embryo and when the stage is completed, fruit mass is about 2g. No further cell division takes place in the fruit skin at the completion of this period.

Seed and aril growth increases rapidly from day 50-65 and forms the second stage of fruit development. During this stage the aril develops around the seed. The initial appearance is a ring of white tissue around the seed base. In the beginning growth is slow, but accelerates simultaneously with cotyledon development and continues to fruit maturity. At the end of this stage, the embryo fills the seed cavity, the seed coat has hardened and the aril has started its rapid growth. Fruit mass is about 7g divided equally amongst peel, seed, and aril (Stern & Gazit, 2003).

During the final stage of fruit development aril and fruit mass increases take place. Total sugars increase from a low of 3% to between 16 and 20% during the final growth period. Depending on the cultivar and climatic conditions, the entire period of fruit development lasts 70-90 days. At maturity, the four commercial cultivars in South Africa, namely ‘Fay Zee Siu’, ‘HLH Mauritius’, ‘Mc Lean’s Red’ and ‘Wai Chee’, can reach average fruit mass of 25-27g, 21-22g, 19-20g and 15-18g respectively.

Successful pollination and fertilization are prerequisites for normal litchi fruit set (retention). The main factors affecting initial fruit set include the availability of fertile female flowers, adequate pollination with viable pollen, favourable environmental conditions, the fertilization process and fruit development (Stern & Gazit, 2003). Zhang *et al.* (1997) state that in most cases poor fruit set and low yields can be attributed to flowering without setting fruit and not lack of flowers. In order to become sufficiently strong sinks for carbohydrates, minerals and plant growth regulators, fertilization is essential.

In other tropical fruit trees with different types of flowers, as in mango, much attention was given to total number of flowers, sex ratio and their influence on fruit set (Galan Sauco, Fernandez Galvan & Calvo, 1984). However, little attention has been given to these aspects in litchi, although a definite correlation between sex ratio and fruit set has been mentioned (Mustard, 1960; Chadha & Rajpoot, 1969). Large differences in the sex ratio of mature inflorescences were noted among cultivars. The percentage of the three flower stages (female, male 1, male 2) was reported in Israel in one study as 32, 34 and 34% (Stern *et al.*, 1993) and in another as 33, 10 and 57% (Nadler, 1995). In Australia, the percentage female flowers were calculated for five cultivars at four locations. The fluctuations in the four locations were from 43-16% (Menzel & Simpson, 1992). In China, ‘Fei Zi Xiao’ has long inflorescences with a high percentage of male flowers, while short inflorescences tend to have a higher percentage of female flowers (Wang & Qiu, 1997).

High fruit drop in litchi often occurs after profuse flowering, leading to low productivity. Under normal conditions, litchi inflorescences have 100-250 flowers, but only a small percentage develops into mature fruit due to flower and fruitlet abscission (Chadha & Rajpoot, 1969; Singh & Lal, 1980; Yuan & Huang, 1988; Stern *et al.*, 1995, 1997; Stern & Gazit, 1997, 1999). According to Stern & Gazit (1999) two distinct fruit drop periods in litchi can be distinguished. During the first month after pollination most of the flowers and fruitlets abscise, which may be caused by inadequate pollination, fertilization or lack of an embryo (Menzel, 1984; Joubert, 1967, 1986; Stern *et al.*, 1995, 1996, 1997). When this first abscission period is over, only 5-15% of the female flowers survive to develop into small fruitlets (Stern, Kigel, Tomer & Gazit, 1995; Stern & Gazit, 2000). One to two weeks later, in week six to eight after full female bloom, a second wave of abscission occurs. This period lasts for one to two weeks, abscising 50% of the remaining fruitlets (Stern *et al.*, 1995). According to Zhang *et al*. (1997), this second fruit drop has the greatest influence on yield. Stern *et al*. (1995) mentioned that only 3 to 5 per cent of the initial fruits finally develop to mature fruits. It was also found that all abscised fruitlets during the second wave of abscission weighed between 2-6 g, containing a well-developed seed coat with either a normal or a defective embryo. Menzel (1984) stated that internal nutritional imbalances due to assimilate competition with growth flushes and other fruits, as well as hormonal imbalances, are further reasons for the second abscission period. In China, Zhang *et al.* (1997) mentioned that a cultivar such as 'Nuomici' may have up to five fruit drop climaxes, with the first, second, third and fourth peaks occurring 12, 25, 40 and 50-55 days after the opening of female flowers, respectively, and the fifth occurring before harvest. Unwanted summer flushes were mentioned as causing nutritional disorders that prevent normal development of fruit, resulting in fruit drop at a late stage of fruit development.

Wittmann (2002) stated that embryo development and abortion plays an important part in litchi fruitlet abscission. It is claimed by Xiang, Ou, Qiu, Yuan and Chen (2001) that differences in embryo development are controlled by genotype. Degani *et al.* (1995) found that self- pollinated fruit often abscise early as seen with the cultivars 'Floridian' and 'Mauritius'. The status of embryo development may therefore not be represented by the proportion of seedless fruit at maturity. It has also been shown in avocado that fruit abscission is selective and is influenced by the pollen parent (Degani, Goldring & Gazit, 1989). Xiang *et al*. (2001) showed in controlled pollination experiments with two cultivars that the effect of selective fruitlet abscission disappeared with cross-pollination. Embryo development and the percentage of fruit with shrivelled seeds were influenced by the pollen parent. Cross- pollination caused a high percentage of aborted seeds in fruit that did not drop, but developed to full maturity. However, Lu (2001) stated that embryo development was not affected by self- or cross- pollination, but that characteristic differences in embryo development were inheritable and not controlled by a single pair of alleles in the genome. Above-mentioned author also showed the importance of plant growth substances (mainly auxins) in embryo development and abortion.

2.6.1 Plant growth regulators and fruit development

2.6.1.1 Auxins

The plant growth substance or plant hormone group known as auxins have been found to be important in embryo and fruit retention and development (Weaver, 1972). The use of synthetic auxins to reduce or prevent fruit abscission in crops such as citrus, apples, peaches and pears, has been known for more than 50 years (Leopold, 1958; Weaver, 1972; Lurie, 2000). Auxins are produced in developing seeds, in shoot tips and in young leaves and play an important role in cell elongation, root growth, embryo growth, fruit growth and the abscission of organs (Kift, 2002). Recently in Israel, 'Mauritius' fruitlet drop was reduced and its yield significantly increased by spraying with 100 ppm of the synthetic auxin 2,4,5- trichlorophenoxypropionic acid (2,4,5-TP) and 25 or 50 ppm of 3,5,6 trichloro-2-pyridyl-oxyacetic acid (3,5,6-TPA). Stern *et al* (1995) and Stern & Gazit (1997) found that optimum application time was at the 2g-fruitlet stage (about 6 weeks after full female bloom) for the cultivar 'Mauritius' (Figure 5).

At the 2 g stage the litchi embryo is in the heart stage and begins its rapid growth, followed by the second wave of fruitlet drop. This second wave of fruitlet drop was significantly reduced by the synthetic auxin applications. Not only did TPA reduce fruit drop, but also increased fruit size and mass relative to the control and TP-treated trees. The two synthetic auxins combined showed an even more substantial increase in yield than either auxin alone (Stern *et al.*, 2000). It was also found by Stern, Stern, Miller, Xu & Gazit (2001) that 2,4,5-TP and 3,4,5-TPA increased yield as well as fruit mass in 'Fei Xi Xiao' and 'Hei Ye' in China.

Figure 5 Heart-shaped embryo six weeks after pollination (±2g).

Fruit size as a quality parameter in litchi is as important as yield in the determination of profitability (Wittmann, 2002). Increased fruit size and limited percentage rejected fruit due to unacceptable size may increase fruit pack-out percentages, and consequently the returns to the grower (Agusti *et al.*, 1994). Weaver (1972) & Agusti *et al.* (1995) mentioned that auxins can stimulate cell division, enhance cell enlargement, induce fruit set and development, affect fruit drop and improve fruit size. Wittmann (2002) stated that fruit growth phases are limited by auxin supply. Auxin application increases fruit growth rate and also has an effect on ripening time, colour, size and post harvest quality (Wittmann, 2002). The application of auxins to fruit with a sigmoid growth curve such as litchi, leads to a prolonged fruit growth period and bigger fruit (Wittmann, 2002).

Gao, Chen, Song, Hu & Huang (2001) found that no differences could be detected in growth of the pericarp, seed and aril of a large seeded, large sized cultivar and a small seeded, small sized cultivar, until 35 days after anthesis. After that, all the mentioned parts of the large seeded, large sized cultivar developed more rapidly, with heavier fruit mass than those of the small seeded, small sized cultivar. It was found that the pericarp growth of the small seeded cultivar lagged behind aril growth, while the cultivar with large pericarp developed into large fruit. A greater source of auxin was therefore displayed by the bigger seed with its bigger endosperm that attracted assimilates to the fruit. Fruit size is dependant on cell division and cell enlargement; therefore a faster and longer period of cell division leads to larger fruit (Wittmann, 2002). The big seeded cultivar had a higher source of auxins around anthesis and in the aril and the pericarp, leading to more active cell division and a bigger pericarp, and subsequently to more rapid aril enlargement.

Spraying with the auxin 3,5,6- TPA in Israel resulted in a significant increase of 14 - 30% in fruit mass in 'Mauritius', 'Floridian', 'Fei Zi Xiao', 'Kaimana' and 'Hei Ye' (Stern & Gazit, 1999; Stern *et al.*, 2000, 2001). The fruit mass increase occurred together with a significant increase in yield. However, these authors found that the main factor involved in the yield increase, was the increase in fruit number.

Increased competition between fruit usually has a negative effect on final fruit size, but results showed that an increase in yield did not prevent an increase in fruit size. The increase in litchi fruit size is therefore not related to fruit thinning, as is the case in apple (Dennis, 1986) and in citrus (Zaragoza, Trenor, Alonso, Primo-Millo & Agusti, 1992). Stern and Gazit (2003) are of the opinion that the treatment probably causes the fruit to be a stronger sink, as was found in a study on Clementine citrus by Agusti *et al.* (1995). The seed mass of fruit also showed an increase to reflect this effect. The TSS (Total Soluble Solids) level of fruit did not decrease with the increase in yield, indicating that the higher crop load did not deplete the reserves in the tree (Stern & Gazit, 2003). In Israel, a combined treatment of spraying 2,4,5-TP, followed about a week later by 3,5,6-TPA, gave the highest yield in 'Mauritius' and 'Floridian' (Stern *et al.*, 2000). These authors found that the increase in yield was the result of a reduction in fruitlet drop with application of both auxins and an increased fruit mass with the second 3,5,6 TPA treatment. These auxins are now being used in commercial litchi orchards in Israel in a two-stage application (Stern & Gazit, 2003). Firstly, 2,4,5-TP at a dosage of 67mg/ ℓ is applied at 5 weeks after full bloom when the fruitlet size is at the 2g-stage. Secondly, in week six (one week later), 3,5,6-TPA at a concentration of 20mg/ℓ is applied (Stern & Gazit, 2003).

However, the response of other cultivars to these auxins still needs to be investigated. The review article of Brown (1997) as well as the abovementioned authors suggests tailor-made protocols to be developed for each cultivar in order to realize the potential of auxins to significantly increase yield in litchi.

2.6.1.2 Cytokinins

Cytokinins are known to have an indirect effect on fruit abscission. Nutrients are mobilised and the sink activity of organs is promoted. In this way abscission is retarded and inhibited through promotion of vigour in the subtended organ (Addicott, 1983).

Cytokinins can also enhance cell division as well as cause larger fruit due to subsequent higher cell volume. This is only applicable if the cytokinins are applied in the early stage of fruit development when active cell division is occurring (Wittmann, 2002). However, no reports are available on the application of cytokinins on litchi and its effect on fruit size.

2.6.1.3 Ethylene inhibitors

Ethylene is one of several plant growth regulators that influence growth and developmental processes including ripening and senescence of fruit (Abeles *et al.*, 1992). It is a simple hydrocarbon that can diffuse into and out of plant tissues from endogenous (biological) or exogenous (non-biological) sources (Saltveit, 1999). For many years ethylene was not recognized as an important plant hormone, mainly because many physiologists believed that the effects of ethylene were due to auxin, the first plant hormone to be discovered. Work on ethylene was also hampered by the lack of chemical techniques for its quantification. Only with the introduction of gas chromatography in ethylene research was the importance of ethylene rediscovered, and its physiological significance as a plant growth regulator was recognized (Burg and Thimann, 1959). Fruit cells have ethylene binding receptors and ethylene naturally triggers these receptors. A molecule, 1-Methylcyclopropene, also known as 1-MCP, binds to ethylene receptors in fruits and plants and makes them insensitive to ethylene. Biochemical changes that lead to ripening and over - ripening do therefore not occur.

The development of 1-MCP provides a different approach for controlling ethylene production and thus ripening and senescence of fruit (Sisler & Serek, 1997). This gas prevents the action of ethylene in plants by binding permanently to ethylene receptors in the tissue. Results of work done by Serek *et al.* (1994), Fan & Mattheis (1999); Ku & Wills (1999) and Macnish *et al.* (1999), showed that 1-MCP can significantly delay ripening and senescence in a range of fruit, vegetables and ornamentals. Similar responses were found in avocado, custard apple and mango (Hofman *et al.*, 2001). Watkins (2006) mentions that a commercial breakthrough in 1-MCP application technology resulted from the formulation of 1-MCP as a stable powder in which it is complexed with y-cyclodextrin, so that 1-MCP is easily released as a gas when the powder is dissolved in water.

Although considerable research has been carried out on the effect of 1-MCP application as a post harvest technology, less work has been done on the use of 1-MCP as a pre- harvest management tool. Preliminary research and test results with Harvista® (developed by Rohm and Haas Company in the USA with 1-MCP as active ingredient) demonstrated that the use of the technology provides a tool for growers to bring ethylene management to the orchard, with a number of significant harvest benefits. Harvista® technology is a pre-harvest foliar spray to prevent the negative impact of ethylene triggered events in the orchard. Phobes (2008) mentioned one of the key benefits of Harvista® to be a selective delaying agent of harvest to allow time for additional fruit growth and colour development. Apples exhibited greater size and reduced fruit drop without negative impacts on colour development in that study.

Kruger & Botha (2007) found that orchard sprays of 1-MCP (Harvista®) slow down the on-tree ripening rate of 'Sunrise Solo' papayas. Results showed that the use of Harvista® allowed fruit to remain longer on the tree so as to reach a more advanced stage of ripening before being harvested. Appearance and internal quality of fruit were also improved.

In litchi, the objective will be to delay, rather than prevent ripening, in order to keep fruit on the tree for longer so as to reach a more advanced stage of development before being harvested.

Aminoethoxyvinylglycine, also known as AVG, is a naturally occurring fermentation product that blocks ethylene production in plants. AVG inhibits 1-aminocyclopropane-1-carboxylic acid (ACC) synthase, the key enzyme in the ethylene synthesis pathway and thus blocks ethylene biosynthesis in plant tissues (Silverman *et al.*, 2004). Application of AVG inhibits ethylene production and delays starch loss, fruit softening and abscission in apples (Kondo & Hayata, 1995; Greene & Krupa, 2000). Studies by Autio & Bramlage (1982), Bramlage *et al.* (1980) and Halder- Doll & Bangerth (1987) have all shown that AVG, when applied as a pre-harvest spray, will suppress ethylene biosynthesis in apple fruit. This delay in apple fruit maturation has led to the development of AVG as a commercial application (ReTain®) for harvest management and improvement of post-harvest fruit quality (Silverman *et al.*, 2004).

2.6.2 Pollen parent and fruit development

It is known that the pollen parent supplies half of the seed and a third of the endosperm genome. These tissues comprise the main portion of the seed; therefore the pollen parent may have a significant xenic effect on seed characteristics (Stern & Gazit, 2003). In nut crops, where the seed constitutes the harvested product, this phenomenon is well known (Sedgley & Griffin, 1989). The embryo and endosperm may exert a significant influence on the fruit maternal tissues (metaxenia). This phenomenon was first found in dates (Nixon, 1935; Reuveni, 1986), but is potentially widespread and may be discernible in many other fruit crops (Stern & Gazit, 2003).

The effect of the pollen parent on fruit and seed mass, as well as the occurrence of shrivelled seeds with aborted embryos, has been studied extensively in litchi. The identity of the pollen parent was determined by hand pollination (Mc Conchie, Batten & Vivian-Smith, 1991; Xiang *et al*., 2001) or by means of isozyme analysis of the fruit embryo (Stern *et al*., 1993; Degani *et al*., 1995). In Israel, self-pollinated and cross-pollinated embryos were identified by isozyme analysis in orchards containing only the cultivars 'Floridian' and 'Mauritius'. Cross-pollinated fruit were heavier and contained heavier seeds than selfed fruit. This effect was more pronounced in seed mass in self-pollinated 'Floridian' fruit. The Israeli researchers found that when seed mass (the xenic effect) is eliminated, the pericarp and aril (the metaxenic effect) from cross-pollinated fruit were heavier than fruit from self-pollinated fruit (Stern *et al*., 1993; Degani *et al*., 1995). In Australia, Batten & McConchie (1991) found that by hand pollinating 'Bengal' inflorescences with pollen from 'Salathiel', yield was increased, and that seed size was increased by pollinating the 'Bengal' flowers with 'Kwai May Red' pollen. Pollination with another cultivar is therefore very important in litchi, not only to improve yield, but also fruit quality.

2.7 PLANT NUTRITION AND FERTILISATION

Mineral nutrition plays a very important role in the growth and development of any fruit, and subsequently determines fruit quality and storage life (Ginsberg, 1985). Among the several factors associated with the production of litchi, balanced nutrition is considered to be the most important in determining productivity and quality (Singh & Babita, 2002). Response of litchi towards applied fertilizers varies, depending upon climatic conditions, soil types and cultivar (Kotur & Singh, 1993). Foliar fertilizers can be used supplementary to soil fertilizing in order to regulate tree growth and development.

In litchi, nitrogen (N) is the most important nutrient, followed by phosphorus (P), potassium (K), calcium (Ca) and magnesium (Mg). The micronutrients iron (Fe), boron (B), copper (Cu), zinc (Zn) and manganese (Mn) are required only in small quantities to maintain tree performance (Menzel, Simpson & Haydon, 1996). Characteristic symptoms appear in the leaves, stems, flowers or fruit when the concentration of a nutrient reaches abnormally high or low levels in a plant. Growth and yield are usually affected long before visible symptoms appear. It is therefore essential to monitor and maintain the concentrations of the nutrients in the plant and soil within the acceptable range determined from healthy, high yielding orchards (Menzel, 2002).

The role of different nutrients in litchi production is briefly discussed:

Nitrogen (N) - Nitrogen is one of the most important nutrients for litchi, since it affects the growth and productivity of the trees (Abercrombie, 2002). Nitrogen has a profound influence on fruit set, fruit retention, length, diameter and mass of fruit. Zhang *et al.* (1997) state that the requirement for nitrogen is dominant during the early stages of fruit development, while the plant requirement for potassium is much more than that of nitrogen at the mid-late stages of development. A deficiency of nitrogen results in stunted tree growth and yellowing of older leaves. Nitrogen is an integral part of proteins, amino acids, nucleic acids, auxins, chlorophyll and coenzymes (Kift, 2002).

Phosphorus (P) - Additional phosphorus applications of litchi trees are usually not necessary, since most orchards have high concentrations of the nutrient (Menzel *et al.*, 1996; Abercrombie, 2002; Menzel, 2005). Zhang *et al.* (1997) mentioned that the phosphorus level in leaves changes gradually during fruit development and that litchi fruit is not sensitive to the requirement of phosphorus. Although a phosphorus deficiency is very rare on litchi trees, symptoms include tip and marginal necrosis (coppery brown colour) of mature leaves proceeding towards the midrib. Responses to phosphorus applications are usually smaller than those recorded for nitrogen (Menzel, 2005).

Potassium (K) – Potassium has several important roles in plants, such as the regulation of water balance through controlling the opening and closing of stomata on leaves and the synthesis and movements of sugars, starches and oils, which may have a direct effect on fruit quality. Potassium also has a prominent effect on the conformation of enzymes and therefore regulates the activities of a large number of enzymes (Marschner, 1995). Potassium is important for fruit set, fruit development and fruit quality in litchi. It is reported by Menzel *et al.* (1996) that many litchi orchards in Australia have low leaf potassium levels (below 0.8 %). This is ascribed to potassium being transported to developing fruit. Potassium is the most abundant nutrient in litchi fruit and therefore removed in relatively large quantities by cropping (Menzel & Simpson, 1987b). Zhang *et al.* (1997) concluded that fruit drop at a late stage of development is related to low potassium levels, while early drop is more related to low nitrogen levels. Deficiency symptoms include yellowing of leaves, dying of leaf tips and later the bases of the leaves. Mature leaves can abscise prematurely, leaving a canopy of small terminal clusters of leaves (Abercrombie, 2002).

Calcium (Ca) - Calcium is an important component of the cell membrane structure and participates in maintaining the middle lamella (Clarkson & Hanson, 1980). Calcium is reported to affect fruit quality in litchi. Kift *et al.* (2002) found that correct usage of most commercial calcium products improved fruit set and yield, improved fruit size in HLH Mauritius, improved fruit firmness and reduced litchi fruit mass loss during cold storage. Bower (1985) found that calcium uptake by fruit is optimal during the first 6 weeks of fruit development and it is crucial that plants must be continuously supplied with calcium. Little or no redistribution occurs after accumulation (Clarkson, 1984). Calcium deficiencies are rare under orchard conditions since most commercial orchards receive calcium in the form of lime or super phosphate. However, fruit borne on vigorously growing trees can have a lower calcium content, because leaves and shoots have a greater physiological activity and transpiration rate and are therefore stronger sinks for calcium than fruit (Witney, Hofman & Wolstenholme, 1990). In culture solutions, where calcium deficiency has been induced, symptoms were small leaflets, leaf necrosis along margins, severe leaf drop, poor root development and no fruit set (Goldweber, 1959).

Magnesium (Mg) - Magnesium deficiencies have been observed in litchi plantings in Australia and in South Africa (Menzel *et al.,* 1996; Abercrombie, 2002) where trees are grown on sandy soils and high rates of nitrogen and potassium have been supplied without maintenance lime applications. Magnesium is not readily transported from the old leaves in litchi; therefore symptoms first become visible in the young leaves (Menzel, 2005). Deficiency symptoms include small leaves (Goldweber, 1959) with dead tips and centres, eventually dropping (Thomas *et al.,* 1995). Under severe deficiency conditions flowering is suppressed (Joiner & Dickey, 1961).

Boron (B) and *Copper (Cu)* - Boron is generally needed for satisfactory fruit set in plants. Deficiencies reduced pollination and flowering, with vegetative growth often unaffected (Hanson & Breen, 1985). According to Xu, Chen, Chen, Wang & Kang (1984) both soil and foliar applications increased litchi production in China. Boron and Copper deficiencies may occur simultaneously on litchi trees. Deficiency symptoms include small leaves, chlorosis between veins, death of growing points and misshapen fruit (Abercrombie, 2002; Menzel, 2005). Optimum boron and copper levels in South Africa are maintained by foliar application during spring and after harvest when trees are flushing with new leaf development.

Zinc (Zn) - Abercrombie (2002), stated that zinc deficiencies are common on most subtropical crops and can occur on most soil types. Deficiency can be intensified by heavy nitrogen fertilization (Menzel *et al.*, 1996). Zinc deficiency symptoms include general bronzing of leaflets, long internodes, smaller leaves and fruit with reduced flesh recovery and sugar content and eventual tree dieback if leaf concentrations reach a critically low level (Menzel *et al.*, 1996; Abercrombie, 2002).

Iron (Fe) and *Manganese (Mn)* - Iron and manganese deficiencies are not very common on litchi trees. It has however been noted in Australia that orchards established on sandy soils showed low iron levels, especially after excessive super phosphate applications which interfere with iron uptake of the roots (Menzel *et al.*, 1996). Zinc, iron and manganese deficiency symptoms are very similar with yellowing of leaves, spreading from young to older growth (Thomas *et al.*, 1995). Iron and manganese deficiencies may be rectified by foliar application on new flush (Abercrombie, 2002).

2.8 CULTURAL PRACTICES

There are many cultural practices that can be applied in orchards. Two specific practices were prominent in the literature with regard to manipulation of fruit size and quality, namely covering of fruit during development and girdling to increase litchi production.

Covering of fruit during development can reduce insect and disease damage and can improve quality at harvest in a number of fruit (Bentley & Viveros, 1992; Kitagawa, Manabe & Esguerra, 1992; Tyas *et al.*, 1998). This approach is used to obtain unblemished, high quality fruit in Japan (Kitagawa *et al.*, 1992) and on litchi in China (Zhang *et al.,* 1997), in Thailand (Sethpakdee, 2002) and in South Africa (Oosthuizen, 1991). In South Africa, brown paper bags were found to be the most suitable and also the most cost effective covering material (De Villiers, 2002). In China and Thailand, newsprint and unspecified paper bags are used as covering materials (Zhang *et al.,* 1997; Sethpakdee, 2002). Galan Sauco & Menini (1984) advised that litchi clusters should be bagged approximately 1.5 to 2 months prior to harvesting. In the Southern Hemisphere fruit clusters are bagged after the 'November drop' period (when fruit are at the 2g stage) to ensure the fruit in the bags will be retained and to prevent rotting inside the bag. It is also advisable to remove most of the leaves around the clusters; otherwise they will fall into the bags and rot. De Villiers (2002) mentions that not too many fruit should be placed in one bag; about 25 per bag are recommended. It is advisable to treat fruit with an appropriate insecticide before covering.

Covering experiments with 'Feizixiao' in China (Hu, Chen, Li, Ouyang, Gao, Wang & Dong, 2001) showed that colour development could be enhanced by diffuse light. Bags were made of adhesive-bonded fabric, with about 58% of the total sunlight being transmitted through the bag. Covering reduced potential damage to anthocyanins caused by direct sunlight. Under orchard conditions with direct sunlight on fruit, the shaded part of the fruit received almost no light and the synthesis of anthocyanins was inhibited, leading to uneven colouration of fruit. Enclosed litchis growing in the lower canopy in South Africa typically have better quality than exposed fruit (Huang, 2005).

Barritt, Rom, Guelich, Drake & Dilley (1987) and Tombesi, Antognozzi & Palliotti (1993) mentioned that radiation interception can have an influence on fruit size through overshadowing of leaves, as shown by smaller fruit in the lower canopy. Tyas *et al*., (1998) is of the opinion that radiation interception by the fruit itself may also be important, since it was concluded by Thorpe (1974) that reduced radiation interception (leading to lower fruit temperatures) may reduce their sink strength. Under above optimal radiation, shading may have the opposite effect of increasing fruit mass, because of reducing the negative effects of too much radiation. Covering can also affect fruit size by increasing relative humidity, and therefore reducing fruit water loss (Tombesi *et al.*, 1993).

Galan Sauco & Menini (1984) mentioned that the advantages of covering fruit bunches outweigh disadvantages such as cost of labour and material. Fruit quality is higher, colour and size is better and less labour is required for harvesting. The practice also protects the fruit from hail damage and from certain species of bat. Oosthuizen (1991) reported that an alternative, but labour intensive method of controlling fruit flies, litchi moth, fruit bats and birds is by tieing moisture resistant brown paper bags around litchi fruit bunches. It was found that bagged fruit have a much better quality, mainly because of the higher humidity (up to 30% higher than without bags on a hot summer day) inside the bag. The picking period of such fruit was also extended by up to two weeks, which can have the advantage of extending the harvesting period of a late cultivar like 'Wai Chee' even further.

Girdling has been used to increase litchi production in Israel, Australia, South Africa and China (Roe, Menzel, Oosthuizen & Doogan*,* 1997; Zhang *et al.,* 1997; Hieke, 2000; Stern *et al.*, 2001). Girdling resulted in the accumulation of carbohydrates above the cut by redirecting them towards the fruit. In addition, girdling also affects the downward movement of phytohormomes (Cohen, 1981; Monselise, 1986). The accumulation of carbohydrates in the canopy provides a rich source of energy for all the reproductive development stages: flowering, fruit set, fruit enlargement and ripening (Goren, Huberman & Goldschmidt, 2004). Lomax, Muday & Rubery (1995) mentioned that girdling disrupts the downward flow of photo assimilates and auxin from the leaves, which can reduce root growth. Girdling can be used to promote flowering, fruit set, retention, size, colour, quality, sugar content and ripening from a few days to a week earlier (Monselise, 1986; Mataa, Tominga & Kozaki, 1998).

Increase in fruit size with girdling has been reported in crops such as nectarines (De Villiers, 1990), peaches (Agusti *et al.,* 1998), apricots (Leopold, 1958), mangoes (Simmons, Hofman, Whiley & Hetherington, 1998) and avocado (Davie, Stassen, Van der Walt & Snijder, 1995). Weaver (1972) mentioned that girdling grapevines at anthesis is a well-known practice, used to improve set and increase berry size. The incidence of berry shatter was reduced and berry size was improved with girdling when 50% of the berries were 4-5 mm in diameter

(Wolf, Van der Merwe & Orth, 1991). In nectarine and peach, girdling is most effective prior to the pit hardening stage of fruit development. Girdling at this time shortened the stage and caused peak fruit growth rate to occur earlier in the season (Day & DeJong, 1990; Agusti *et al.*, 1998). Day & DeJong (1990) found that fruit mass was increased by 22-25% and also more than doubled the percentage of fruit in the larger fruit size category.

In apricots, it was found by Leopold (1958) that auxin applications increase fruit growth only after pit hardening, while girdling of branches increased the fruit growth rate during the whole fruit development period. Also in citrus, Fishler, Goldschmidt & Monselise (1983) found that girdling is effective throughout fruit development, while auxins and other plant growth substances are effective only during the early stage of fruit development (Ortola, Monerri & Guardiola, 1997).

Timing of girdling is also very important to influence fruit size, since early girdling during anthesis or a few weeks thereafter, increases the yield at the expense of fruit size in citrus and in litchis (Cohen, 1981; Wittmann, 2002). Cohen (1984) mentioned that only a small or no increase in fruit size is obtained when girdling is carried out too late, several weeks after the fruit drop period. Girdling at different times during the fruit growth phase increased fruit size. Fishler *et al.* (1983) and Cohen (1984) found that early summer girdling was the most effective. Girdling at the end of the summer fruit drop and beginning of autumn was found to increase final fruit size and mass in mandarin (Mataa *et al.*, 1998).

In China, girdling is used to control shooting and to promote flowering and also as a measure to retain fruit on young litchi trees (Zhang *et al.*, 1997). In Australia, yield of two litchi cultivars was increased when girdling about a month after fruit set. It was reported that the effect on average fruit size was slight (Menzel & Mc Conchie, 1996). In South Africa, Roe *et al.* (1997) found that yield and fruit mass of two litchi cultivars were improved when girdling was executed on branches two to four weeks after the full female flowering stage. Contrary to these findings, Oosthuizen (1993) mentioned lower yields from girdled trees than with control trees, but fruit ripened two weeks earlier with girdled trees when compared to maturity of the control fruit.

2.9 DISCUSSION AND CONCLUSION

The introduction of new cultivars to the South African litchi Industry necessitates research into factors that can influence production and quality aspects of such cultivars. The late cultivar 'Wai Chee', introduced from Australia, shows potential to stretch the short harvesting season in South Africa and thereby alleviating a very congested marketing period. The cultivar proved to be precocious with high yielding potential per hectare in close density plantings, but fruit size and quality still remain a problem. A review of the relevant literature indicates that there are various chemical, biological and cultural practices that can contribute towards the improvement of fruit size, yield and quality in litchis. However, none or very little information on the effects of these practices on 'Wai Chee' litchi is available.

Plant growth regulators such as auxins and ethylene inhibitors have shown satisfactory results on a number of crops such as citrus, apples and peaches. In litchi, the use of synthetic auxins showed increases in fruit size and mass and also increased yields of the 'HLH Mauritius' cultivar in Israel. These synthetic auxins were not tested on other cultivars and the potential of these plant regulators on 'Wai Chee' is therefore unknown.

Balanced nutrition is another aspect to be considered very important in determining productivity and quality in litchi. The response of 'Wai Chee' towards different fruit size and quality enhancing fertilizers therefore needs to be investigated.

Another factor found to be involved in yield and fruit size in litchi is the source of pollen in the fertilization process. Studies in Israel and in Australia showed that pollen parent can have an effect on yield, fruit- and seed mass in litchi. The effects of self- and cross pollination on yield and quality aspects in 'Wai Chee' are unknown and a better understanding of the benefits of different pollen donors should be researched.

Cultural practices, such as covering of fruit and girdling of branches were investigated in a number of crops, including litchi. The potential beneficial effects of these practices on 'Wai Chee' should be evaluated. However, for this study, fruit bunch covering was chosen as the most suitable cultural manipulation technique. Bunch covering is a technique known to have the potential of delaying litchi fruit maturity, whereas girdling is known to advance litchi harvest with early fruit maturity. An advanced harvest is not desirable in a late cultivar such as ‘Wai Chee’, while a delayed harvest could benefit from rising prices towards the end of the litchi marketing season in South Africa.

The objective of this study was therefore to investigate various horticultural methods for improving fruit size, quality and yield in ‘Wai Chee’ in order to address some of the problems restricting the commercial potential of the cultivar in South Africa.

CHAPTER 3

3 THE EFFECT OF DIFFERENT CHEMICAL TREATMENTS ON TREE-, FRUIT- AND QUALITY CHARACTERISTICS IN 'WAI CHEE' LITCHI

3.1 INTRODUCTION

Litchi (*Litchi chinensis* Sonn.) production has steadily grown in importance with the increasing popularity of exotic fruits on world markets. The South African Litchi Industry generally exports an average of 2500 tonnes of fruit, of which 95% will be marketed in the European Union (PPECB Export Directory, 2006). The demand is for large, bright red fruit with small seeds and crisp, sweet flesh (Menzel & Simpson, 1990).

One of the biggest constraints in the South African Litchi Industry is the lack of suitable cultivars to extend the very short harvesting season. Only two cultivars, namely 'HLH Mauritius' and 'McLean's Red', have been cultivated commercially for nearly a century in South Africa. This situation has led to a very short marketing period during a time when competitors such as Madagascar are flooding export markets with their big volumes, inevitably lowering returns for local growers. However, during the early 1990's, the South African Litchi Industry initiated efforts to import other promising cultivars, mainly from Australia and Israel. Special attention was paid to earlier and later cultivars than the existing commercial ones.

The late season litchi cultivar 'Wai Chee' was imported amongst others and proved to be precocious with high yielding potential per hectare in high density plantings. However, small fruit size and average internal quality restrict its potential. The fact that 'Wai Chee' is harvested at a time in South Africa when there is a gap in worldwide litchi production, makes the cultivar potentially very profitable. Methods to improve fruit size and quality, and thus to enhance the appeal of this cultivar on local and overseas markets are therefore necessary. Due to the relative novelty of the cultivar in South Africa, very little research on 'Wai Chee' has been conducted so far.

In the case of other cultivars and crops, considerable work has been done on the effects of chemical treatments for the improvement of yield, size and quality of fruit. Plant growth regulators such as auxins (synthetic, organic or endogenous) and ethylene inhibitors have shown satisfactory results on a number of crops such as litchi, avocados, apples and peaches (Autio & Bramlage, 1982; Serek *et al.,* 1994; Agusti *et al*., 1994, 1995; Fan & Mattheis, 1999; Ku & Wills, 1999; Macnish *et al.,* 1999; Stern *et al*., 2000, 2001; Hofman *et al.,* 2001).

Balanced nutrition is another aspect considered to be very important in determining productivity and quality in litchi (Aylward, 2002; Singh & Babita, 2002). The main nutrients involved in determining fruit size include nitrogen, potassium, phosphate and calcium.

The effects of these treatments are unknown in the case of ‘Wai Chee’ under South African conditions. In order to investigate the effect of different chemical treatments on tree-, fruit- and quality characteristics in ‘Wai Chee’ litchi, various foliar nutrition-, plant growth regulator- and ethylene inhibitor products were tested in a series of trials. During a first year of screening trials conducted during 2006, the aim was to identify the products with the best potential for further evaluation. The trial was repeated in a second season (2007) with the inclusion of only the best performers from the first years’ evaluation, as well as adding two programmes, where the best performing products were applied in sequence at different fruit developmental stages.

3.2 MATERIALS AND METHODS

3.2.1 Characteristics of the experimental plot

The study was conducted during 2006 and 2007 on a commercial farm in the Mpumalanga Province, South Africa. The experimental site consisted of ‘Wai Chee’ (*Litchi chinensis* Sonn.) trees, all grown from air-layers. The orchard was established in 2002 and is situated near Nelspruit (25°27’ S, 30°58’ E) in the eastern, subtropical Lowveld of Mpumalanga. The Nelspruit area is characterised by a cool winter and warm, humid summer. Soil in the orchard is sandy (Glenrosa type) with a pH of 6.9.

Trees were spaced 2.5 m apart within rows and 5 m between rows (800 trees/ha). Healthy, vigorous trees, uniform in canopy size and flower intensity, were included in the trial design (Figure 1).

Figure 1 Healthy, vigorous 'Wai Chee' five year old trees; uniform in canopy size, spaced 2.5 m apart within rows and 5 m between rows, were used in the trial. Trees are established on ridges with an open-hydroponics drip irrigation system.

Irrigation was supplied through an open-hydroponics drip irrigation system, scheduled three times per day in summer, for three hours, totalling nine hours per day. In winter, scheduling comprised of one irrigation cycle of three hours per day.

With the usage of three litres per hour (3ℓ/hr) drippers, 27 litres/tree/day were delivered on average in summer and 9 litres/tree/day in winter.

Commercially recommended orchard management practices for litchis in South Africa (Froneman & Husselman, 2002), including nutrition (fertiliser) applications based on soil and leaf analysis recommendations, were maintained throughout the duration of the trials.

3.2.2 Trial layout

3.2.2.1 Statistical design

The trial was laid out as a complete randomised block design. The study was done over two consecutive seasons by investigating a range of nutrition and plant growth regulators (PGR), applied alone or in combination (Table 1). All chemicals were applied according to the manufacturers' specifications.

Table 1 Treatments applied to 'Wai Chee' litchi at Nelspruit for fruit size and quality enhancement

Treatment #	Treatment	Concentration	Time of application
1	Control	-	-
2	Maxim®	40 ppm (0.4g/ℓ)	2-3g stage
3	Tipimon®	100 ppm (1.47l mℓ/ℓ)	2-3g stage
4	Tipimon® Maxim®	100 ppm (1.47mℓ/ℓ) 40 ppm (0.4g/ℓ)	2-3g stage 1 week later
5	Kelpak®	5 mℓ/ℓ (product)	FFB + 10 & 20 days later
6	Orca DS®	2 mℓ/ℓ (product)	FS + 5 weeks later
7	Harvista®	3 g/ℓ (product)	2 weeks before harvest
8	ReTain®	0.9 g/ℓ (product)	2 weeks before harvest
9	KNO_3	20 g/ℓ (product)	After FS + 2,4 & 7 weeks later
10	Ca Metalosate	2 mℓ/ℓ (product)	After FS + 2 & 4 weeks later
11	$CaNO_3$	20 g/ℓ (product)	After FS + 2 & 4 weeks later
12	Crop Plus®	2 mℓ/ℓ (product)	FS + 5 weeks later

FFB= Full Female Bloom *FS= Fruit Set*

Two nutrition treatments (KNO_3 and $CaNO_3$) and three plant growth regulator treatments (Harvista®, ReTain® and Tipimon®-Maxim® combination), containing different ethylene inhibitors and synthetic auxins, were selected for further evaluation in the second year. In addition, two different programmes, containing the best performing products of the first season's screening trials, were also included as treatments in the second season. These selected products were applied in sequence at different fruit developmental stages.

3.2.2.2 Statistical analyses

Data were statistically analysed as a randomised complete block design (RCBD). Analysis of variance (ANOVA) was used to test for differences between the selected chemical treatment effects. The data were acceptably normal with homogeneous treatment variances. Treatment means were separated using Fisher's protected t-test and least significant differences (LSD) at the 5% level of significance (Snedecor & Cochran, 1980). Calculated data were analysed using the statistical program GenStat® version 6 (VSN International Hermel Hempstead, UK).

3.2.3 Treatment details

3.2.3.1 Treatment details for Year 1

Eleven treatments and a control were imposed on 144 trees in a randomised complete block design. The blocks were assigned across rows. Each treatment was applied to three (3) consecutive trees to allow the proper application of chemicals in each of the four (4) replicate blocks. At least two (2) single guard (non-treated) trees separated each of the treatments within the row.

Figure 2 **Treatments were applied as a medium cover spray using a commercial motorised mist blower with hand lances at 2000 kPa and a 1mm nozzle at 8.0 litres per tree.**

The treatments, where applicable, were applied as a medium cover spray using a commercial motorised mist blower with hand lances at 2000 kPa and a 1mm nozzle at 8.0 litres per tree (Figure 2):

Control:

i. **Control trees** did not receive any treatments or sprays.

Nutrition treatments, containing nitrogen, potassium and calcium:

ii. **Calcium nitrate ($CaNO_3$)** containing 19.5% Ca and 15.5% N (manufactured by Sasol Nitro, South Africa). Applied at a rate of 20g/ℓ water immediately after fruit set and repeated after 2 and 4 weeks.

iii. **Potassium nitrate (KNO_3)** containing 38% K and 13% N (manufactured by Sasol Nitro, South Africa). Applied at a rate of 20g/ℓ water just after fruit set and repeated 2 , 4 and 7 weeks after fruit set.

iv. **Calcium Metalosate** containing 60g/kg Ca (manufactured by Ocean Agriculture, South Africa). The small molecular size of the product favours quick absorpsion across the leaf cuticle. Applied at a rate of 2 mℓ/ℓ water just after fruit set and repeated after 2 and 4 weeks.

v. **Crop Plus®**, containing nitrogen, phosphorus, potassium, sulphur and organic chelated micronutrients (manufactured by Cytozyme, USA). Applied at a rate of 2 mℓ/ℓ water at fruit set and repeated after 5 weeks.

Plant growth regulators, containing synthetic auxins:

vi. **Tipimon®** containing 6.8% 2,4,5 trichloro-phenoxy propionic acid (2,4,5-TP) formulated as a triethanolamine salt (manufactured by Tapazol, Israel). Applied at a rate of 1.47 mℓ/ℓ water when the embryo reached the heart shape (fruitlets weighed approximately 2-3g).

vii. **Maxim®** containing 10% 3, 5, 6 trichloro 2-piridyl-oxyacetic acid (3, 5, 6-TPA) as a free acid (manufactured by DowElanco, Spain). Applied at a rate of 0.4 g/ℓ water when the embryo reached the heart shape (fruitlets weighed about 2-3g).

viii. **Tipimon® -Maxim®** which is a combination of Tipimon® (2, 4, 5-TP) applied at 2-3g stage, and Maxim® (3, 5, 6-TPA) applied one week later. Same concentrations as with individual applications were used.

Plant growth regulators, containing naturally- occurring auxins and cytokinins:

ix. **Kelpak®** containing 11 mg/ℓ organic auxins and 0.0031 mg/ℓ organic cytokinins (manufactured by Kelp Products, South Africa). Applied at a rate of 5 mℓ/ℓ water during full female bloom (FFB), 10 days after FFB and 20 days after FFB.

x. **Orca DS®** containing 30 mg/kg cytokinins and 45 mg/kg auxins with added trace elements Manganese (0.25 g/kg), Copper (0.025 g/kg), Iron (0.712 g/kg), Zinc (0.2 g/kg), Boron (0.025 g/kg) and Molybdenum (0.025 g/kg) (manufactured by Madumbi-BCP, South Africa). Applied at a rate of 2 mℓ/ℓ water during fruit set and 5 weeks after fruit set.

Plant growth regulators, containing ethylene inhibitors:

xi. **ReTain®** containing aminoethoxyvinylglycine (AVG) (manufactured by Abbott laboratories, USA). Applied at a rate of 0.9 g/ℓ water, 2 weeks before harvest.

xii. **Harvista®** containing 1-Methylcyclopropene (1-MCP) (manufactured by Rohm and Haas, USA). Applied at a rate of 3g/ℓ water, 2 weeks before harvest.

3.2.3.2 Treatment details for Year 2

The treatments that showed the best promise to improve tree, fruit and quality characteristics during 2006 were selected for further evaluation in 2007. In addition two programmes were also tested, where the best performing products of 2006 were applied in sequence at different fruit developmental stages. The same experimental design was followed in both years.

Selected treatments from results of year 1

i. **Calcium nitrate ($CaNO_3$), Potassium nitrate (KNO_3), Tipimon® -Maxim®, ReTain®** and **Harvista®**. Applied at the same rates and time intervals as described in Table 1.

Programmes 1 and 2

ii. **Programme 1**, consisting of the nutrition treatment KNO_3, followed by the synthetic auxin combination treatment of Tipimon® and Maxim®, followed by the ethylene inhibitor ReTain®. Applied at the same rates and time intervals as described with single product treatments (Table1).

iii. **Programme 2**, consisting of the nutrition treatment $CaNO_3$, followed by the synthetic auxin combination treatment of Tipimon® and Maxim®, followed by the ethylene inhibitor ReTain®. Applied at the same rates and time intervals as described with single product treatments (Table 1).

3.2.3.3 General chemical enhancers in combination with all other applications in year 1 and 2:

- Commodobuff®, (manufactured by Villa Crop Protection, South Africa), a buffering agent for the correction of water pH to a reading of between 4 and 6, was used at a concentration of 50 mℓ/100 ℓ water with all products.

- Agral 90® (manufactured by Kynoch Agrochemicals, South Africa), a non-ionic surfactant containing alkylated phenol ethylene oxide, was used at a concentration of 12 mℓ/100 ℓ water with all products.

3.2.4 Data collection

Data relating to tree-, fruit- and quality characteristics of 'Wai Chee' litchi were collected at harvest:

- **Tree data:** Complete individual tree yields (total mass) were determined by weighing all the fruits at the time of harvest.
- **Fruit data:** Data from 'Wai Chee' trees were taken by collecting samples of 35 fruits per tree at random. Each fruit was weighed and its size was determined by measuring the shoulder width with an electronic hand calliper. Seed and peel of fruit were also weighed. Flesh mass, flesh-to-fruit and seed-to-fruit ratios were calculated from the measured data.
- **Quality data:** Total soluble solids (TSS) were determined with a temperature-compensating digital refractometer (Pal-1, manufactured by Atago®, Japan). Titratable acidity (TA) was determined by titration of a 10 ml aliquot of juice using 0.1 N NaOH to an endpoint with phenolphthalein as an indicator (Wardowski *et al.*, 1979). The internal quality of the fruit was determined by using the TSS: acid ratio of the pulp, as it is well correlated to mean eating quality (Underhill and Wong, 1990). Each of the 35 sample fruit was analysed separately.

3.3 RESULTS

3.3.1 Tree characteristics in Year 1

Figure 3 presents yields of 'Wai Chee' after application of eleven different nutrition and plant growth regulator treatments and a control. Treatments with ReTain®, and Crop Plus® significantly increased yields compared to the control by 67 and 56% respectively. Trends of increases and decreases in yield were obtained with the other chemical treatments but these were not statistically significant.

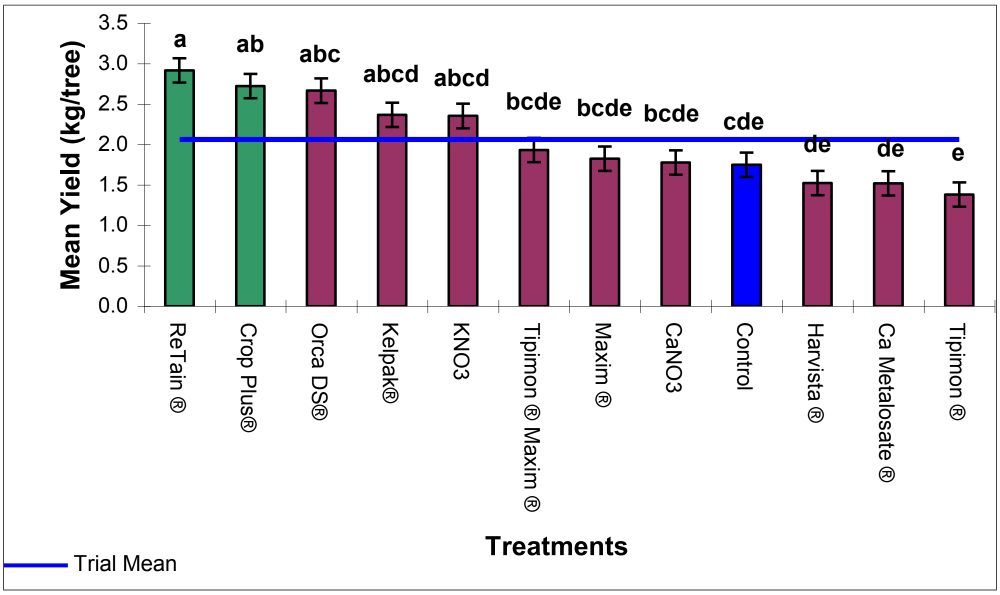


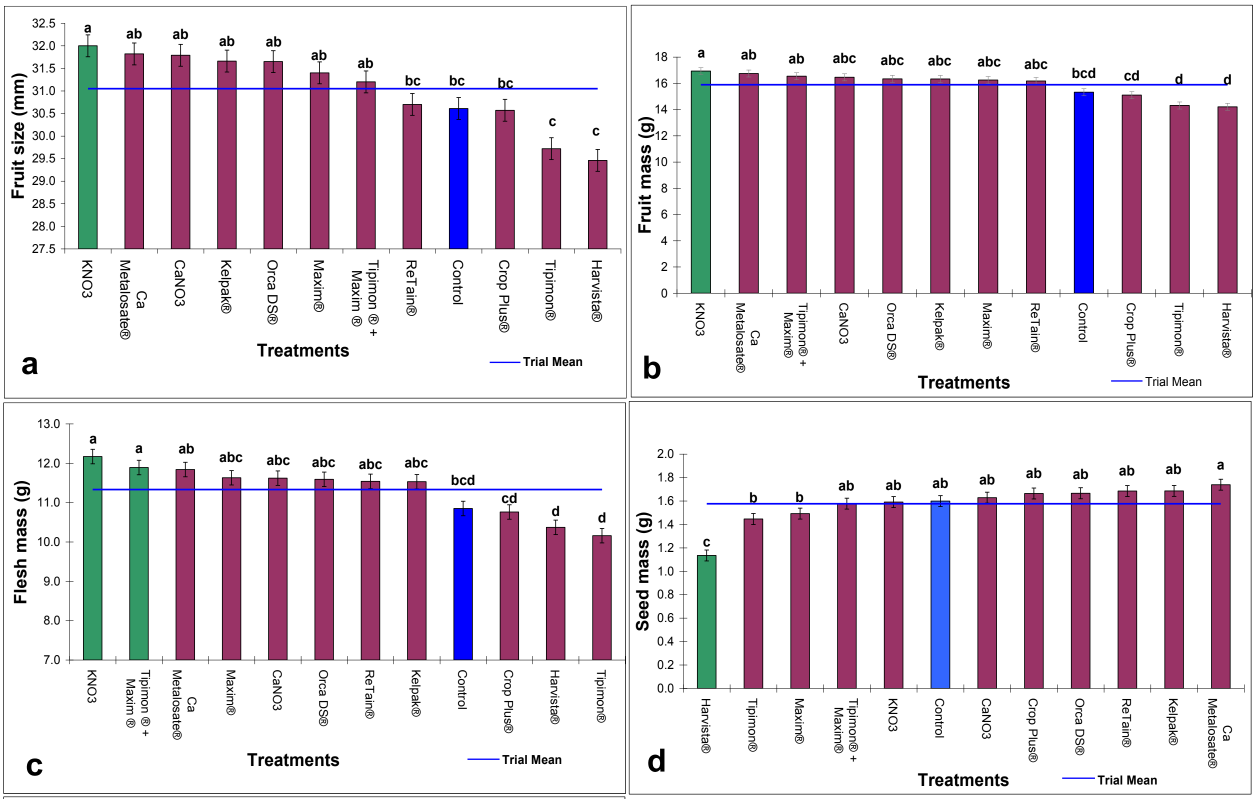
Figure 3 Wai Chee' yield (kg/tree) at Nelspruit as influenced by 11 different chemical applications in 2006. Different letters indicate significant differences at P≤ 0.05 (LSD=0.9681). Green bars were used to show the treatments that were significantly better than the control (blue bar).

3.3.2 Fruit characteristics in Year 1

The effects on fruit characteristics of 'Wai Chee' litchi after 11 nutrition and plant growth regulator applications, and a control are shown in Figure 4a-e.

The only significant increase in fruit size (Figure 4a) of 'Wai Chee' as opposed to control fruit occurred in the nutrition treatment of KNO_3. Although not statistically significant, there was nevertheless a tendency to size increase by application of nutrition treatments Ca Metalosate and $CaNO_3$ as well as with plant growth regulators Kelpak®, Orca DS®, Maxim® and the combination treatment of Tipimon® and Maxim®. Treatments with Crop Plus®, Tipimon® and Harvista®

tended to decrease fruit size compared to the control, although again, this was not significantly different.



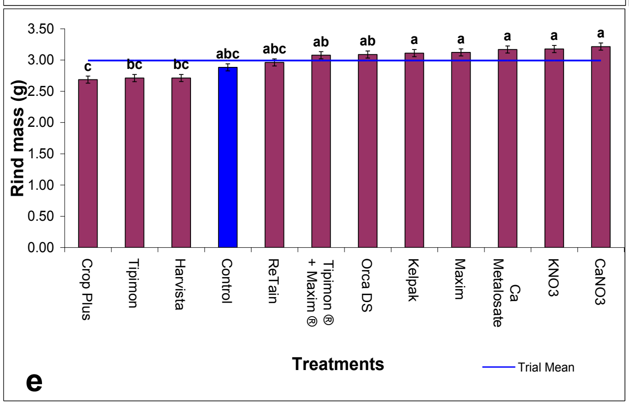


Figure 4a-e
Five different fruit characteristics of 'Wai Chee' litchi at Nelspruit as influenced by 11 different chemical applications in 2006.

Different letters indicate significant differences at P≤ 0.05. Green bars were used to show the treatments that were significantly better than the control (blue bar).

Regarding fruit mass (Figure 4b), the nutrition treatment of KNO_3 was the only treatment to significantly increase fruit mass as opposed to the control treatment. Although a tendency towards increasing fruit mass was shown by applications of Tipimon® in combination with Maxim®, Orca DS®, Kelpak®, Maxim® and Retain®, as well as the nutrition treatments of Ca Metalosate and $CaNO_3$, these were not significant. Crop Plus®, Tipimon® and Harvista® showed a tendency to decrease fruit mass, although again not statistically significant.

Flesh mass (Figure 4c) of 'Wai Chee' fruit was significantly increased by application of the nutrition treatment of KNO_3, and by the combination treatment of Tipimon® and Maxim®. A tendency to increased flesh mass (non significant) was shown by the nutrition treatments Ca Metalosate and $CaNO_3$ and by the plant growth regulators Maxim®, Orca DS®, Kelpak® and ReTain®. Treatments with Crop Plus®, Harvista® and Tipimon® tended to decrease flesh mass, although not statistically significantly.

Reduced seed mass (Figure 4d) and subsequent seed-to-fruit ratio to increase the proportionate edible portion of the fruit is an important parameter in determining quality in litchi. The only statistically significant decrease in seed mass was found with the plant growth regulator Harvista®. Treatments with the plant growth regulators Tipimon®, Maxim® and the combination treatment of Tipimon® and Maxim®, as well as the nutrition treatment KNO_3 also tended to decrease seed mass, but not statistically significant. On the other hand, a tendency to increase seed mass was obtained with nutrition treatments $CaNO_3$, Crop Plus® and Ca Metalosate, and with the plant growth regulators Kelpak®, Orca DS® and ReTain®, although not statistically significant.

With rind mass (Figure 4e), no statistically significant differences were found between the different treatments when compared to the control. However, the three treatments displaying the highest rind mass, namely nutrition treatments Ca NO_3, KNO_3 and Ca Metalosate, were also among treatments with the highest fruit mass. The treatments with a reduction in rind mass, namely Harvista®, Tipimon® and Crop Plus®, also reflected treatments with the lowest fruit mass. A strong positive correlation (r^2=0.74) was found between rind mass and fruit mass over all the different treatments (Table 2).

3.3.3 The correlation amongst tree- and fruit characteristics

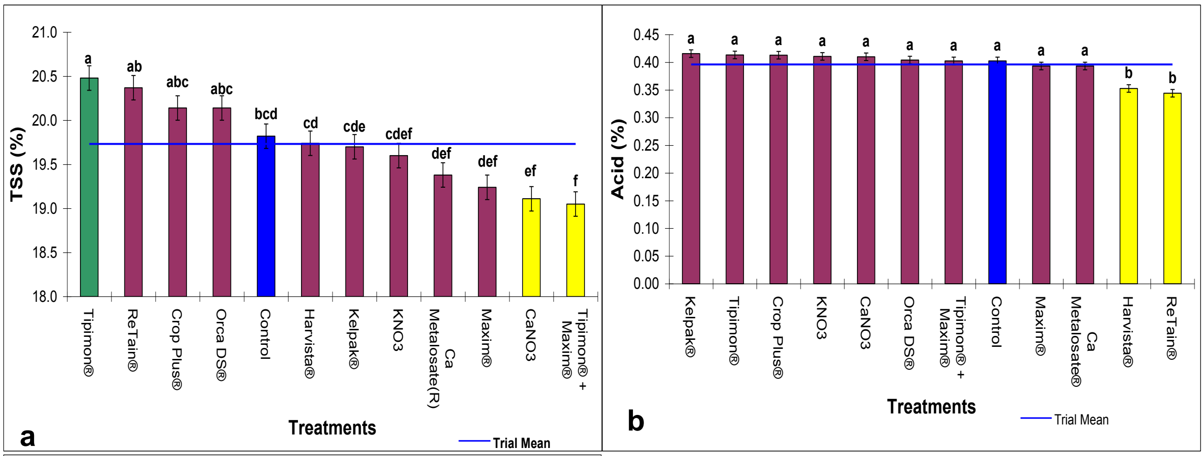
Correlations were calculated for the 6 characteristics discussed in Figures 3 and 4. Data in Table 2 indicate that increased yields did not negatively influence fruit size and mass for all the different treatments. A significant correlation was found between fruit mass and fruit size (r^2=0.88) as well as between fruit mass and flesh mass (r^2=0.93). A lower, but significant correlation was also found between fruit mass and rind mass (r^2=0.74). Although significant, correlations (r^2=0.39) between seed mass and fruit mass in all the treatments were low.

Table 2 Extraction from the correlation matrix of yield and physical fruit characteristics of 'Wai Chee', showing r^2 values for six different characteristics. Significant correlation values are highlighted in blue.

Fruit size (mm)	**1.00**					
Fruit mass (g)	**0.88**	**1.00**				
Rind mass (g)	**0.72**	**0.74**	**1.00**			
Flesh mass(g)	**0.76**	**0.93**	**0.55**	**1.00**		
Seed mass(g)	**0.42**	**0.39**	**0.28**	**0.21**	**1.00**	
Yield (kg/tree)	**0.07**	**0.09**	**0.01**	**0.10**	**0.10**	**1.00**
	Fruit size (mm)	**Fruit mass(g)**	**Rind mass(g)**	**Flesh mass(g)**	**Seed mass(g)**	**Yield (kg/tree)**

3.3.4 Quality characteristics in Year 1

Figure 5a-c shows quality characteristics of 'Wai Chee' litchi fruit flesh as influenced by eleven different chemical applications.



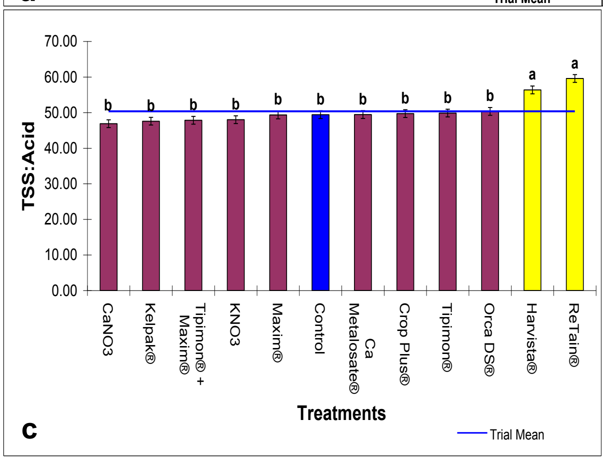


Figure 5a-c

Quality characteristics fruit characteristics of 'Wai Chee' litchi fruit at Nelspruit as influenced by eleven different chemical applications in 2006. Different letters indicate significant differences at P≤ 0.05. Green bars and yellow bars indicate the treatments that were respectively significantly higher and lower than the control (blue bar).

The only statistically significant increase in TSS (Figure 5a) as compared to the control was found with the growth regulator Tipimon®. Tendencies to increase

TSS percentages were also found with plant growth regulators ReTain® and Orca DS®, as well as with nutrition treatment Crop Plus®, but these increases were not statistically significant. The combination treatment with Tipimon® and Maxim®, and $CaNO_3$ showed a statistically significant decrease in TSS, while Harvista®, Kelpak®, Maxim®, KNO_3 and Ca Metalosate also showed only slight, non-significant decreases in TSS percentages.

The only statistically significant decrease in acid (TA) concentrations (Figure 5b) when compared to the control, occurred in plant growth regulator treatments Harvista® and ReTain®. None of the other treatments delivered significant increases or decreases in acid levels.

Regarding the Total Soluble Solids to Acid ratio (Figure 5c), the plant growth regulators ReTain® and Harvista® displayed statistically significantly higher ratios compared to the control ratio. This increased TSS: TA ratio is reflected in the acid determination of treatments, with the same two ethylene inhibitors also displaying low acid levels. Slight increases and decreases in ratios were found with the other chemical treatments, but none of these were statistically significant.

3.3.4.1 Summary of the effect of 11 different chemical applications on 'Wai Chee' tree-, fruit- and quality characteristics at Nelspruit.

The overall effect of the 11 different chemical treatments in comparison with the non-chemical treated control on 'Wai Chee' fruit at Nelspruit in year 1 (2006) is summarized in Table 3. Characteristics that were measured included tree-, fruit- and quality characteristics.

Table 3 Summary of the effect of different chemical treatments compared to a non-chemical treated control, on 'Wai Chee' tree-, fruit- and quality characteristics at Nelspruit in 2006.

Treatments	Yield (kg/tree)	Fruit Size (mm)	Fruit Mass (g)	Rind mass (g)	Flesh Mass (g)	Seed Mass (g)	TSS (%)	TA (%)	TSS:TA
KNO_3	Same	Higher	Higher	Same	Higher	Same	Same	Same	Same
$CaNO_3$	Same	Same	Same	Same	Same	Same	Lower	Same	Same
Maxim ®	Same	Same	Same	Same	Same	Same	Same	Same	Same
Tipimon ®	Same	Same	Same	Same	Same	Same	Higher	Same	Same
Tipimon® & Maxim ®	Same	Same	Same	Same	Higher	Same	Lower	Same	Same
Crop Plus®	Higher	Same	Same	Same	Same	Same	Same	Same	Same
Ca Metalosate ®	Same	Same	Same	Same	Same	Same	Same	Same	Same
Kelpak®	Same	Same	Same	Same	Same	Same	Same	Same	Same
Orca DS®	Same	Same	Same	Same	Same	Same	Same	Same	Same
Harvista ®	Same	Same	Same	Same	Same	Lower	Same	Lower	Higher
ReTain ®	Higher	Same	Same	Same	Same	Same	Same	Lower	Higher

(TSS = Total Soluble Solids, TA = Titratable Acids)

3.3.5 Results in Year 2 of single product applications selected from Year 1

The treatments of the 2007 trial were selected from those that performed best during the 2006 screening trial. No statistical significant enhancing effect could be found in 2006 with the application of naturally-occurring auxins and cytokinins, and were therefore not considered for further evaluation in 2007. The results reported under point 3.3.5.1 to 3.3.5.4 are thus a verification of a portion of the 2006 results, where the selected products were again applied as single applications as in the first year. Additionally, two programmes were included in this trial to determine the interaction and combined effect of single products. The results reported under point 3.3.6.1 to 3.3.6.3 depict the effect of the single products versus their combined effect in a programme. Verification of results of the best performing treatments from Year 1 with a second year's evaluation on different tree-, fruit- and quality characteristics in 'Wai Chee' litchi, is shown in Figures 6- 8.

3.3.5.1 Tree characteristics, in Year 2, of single product applications selected from Year 1

Figure 6 presents yields per tree at harvest of 'Wai Chee' litchi after selected nutrition and plant growth regulator treatments over two consecutive seasons. Treatment with the ethylene inhibitor ReTain® significantly increased yields in

2006, with an increase of 67% compared to the control. Contrary to these results, application of ReTain® tended to decrease yield in 2007. Applications of KNO_3, $CaNO_3$ and the Tipimon®-Maxim® combination improved yields in both seasons compared to the control, although not significantly. None of the treatments applied during the second year differed significantly from the control.

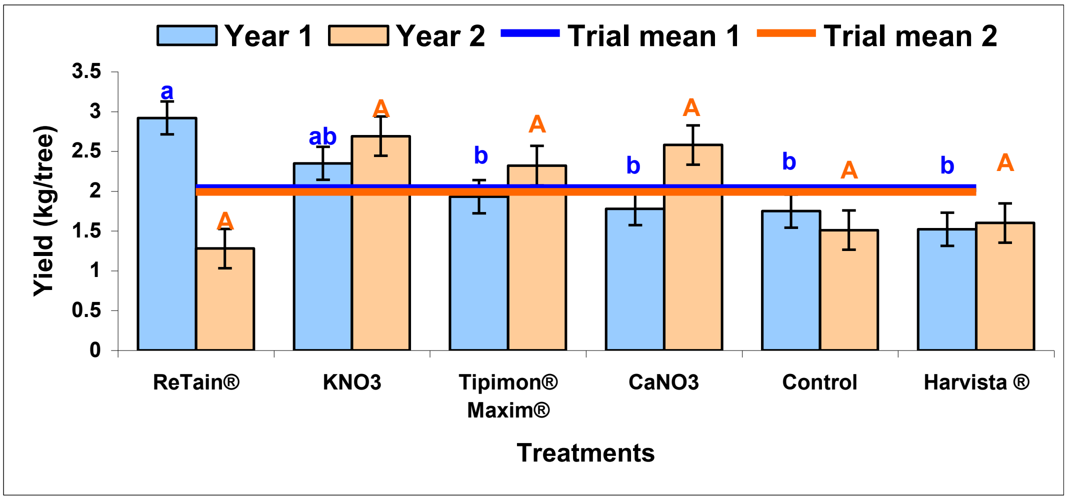
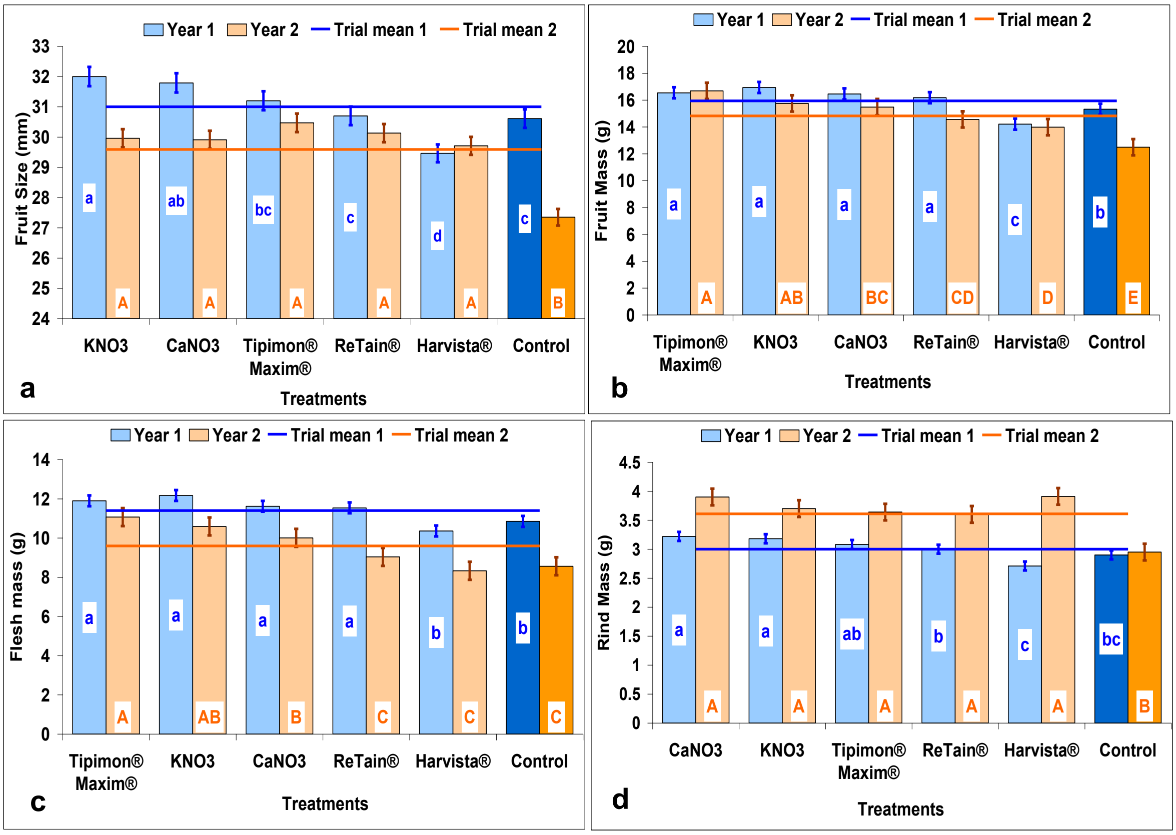


Figure 6 **Yield (kg/tree) at harvest of 'Wai Chee' litchi at Nelspruit as influenced by five different chemical applications repeated over two consecutive seasons during 2006 and 2007. Different letters indicate significant differences at P≤ 0.05. Lower case letters represent Year 1 (2006) and upper case letters Year 2 (2007).**

3.3.5.2 Fruit characteristics, in Year 2, of single product applications selected from Year 1

The effects of five single chemical applications and a control on fruit characteristics of 'Wai Chee' litchi during two consecutive seasons are shown in Figure 7a-e.

The fruit characteristics relating to fruit size, fruit mass and flesh mass (Figure 7a-c) were all lower in the 2007 trial than in the 2006 trial, whereas rind- and seed mass (Figure 7d-e) on the other hand, were higher for all products. Although measurements of fruit characteristics were lower in 2007 than in 2006, all of the chemical applications applied in the second year caused significant increases in all fruit characteristics when compared to the control. The only exception occurred in flesh mass (Figure 7c), where no significant difference could be detected between the control and applications with Retain® and Harvista®.



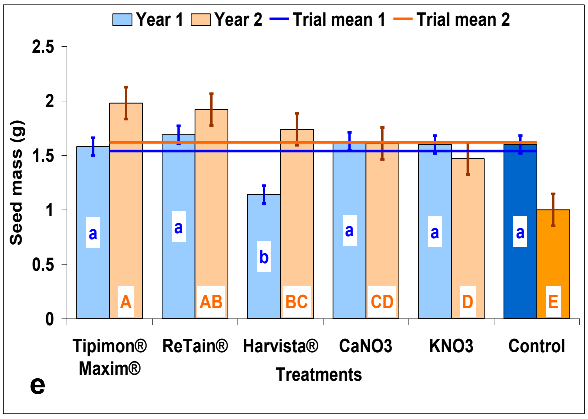


Figure 7a-e

Five different fruit characteristics (fruit size and mass, flesh mass, rind mass and seed mass) of 'Wai Chee' litchi at Nelspruit as influenced by five different chemical applications repeated during two consecutive seasons (2006 and 2007).

Different letters indicate significant differences at P≤ 0.05. Lower case letters represent Year 1 (2006) and upper case letters Year 2 (2007).

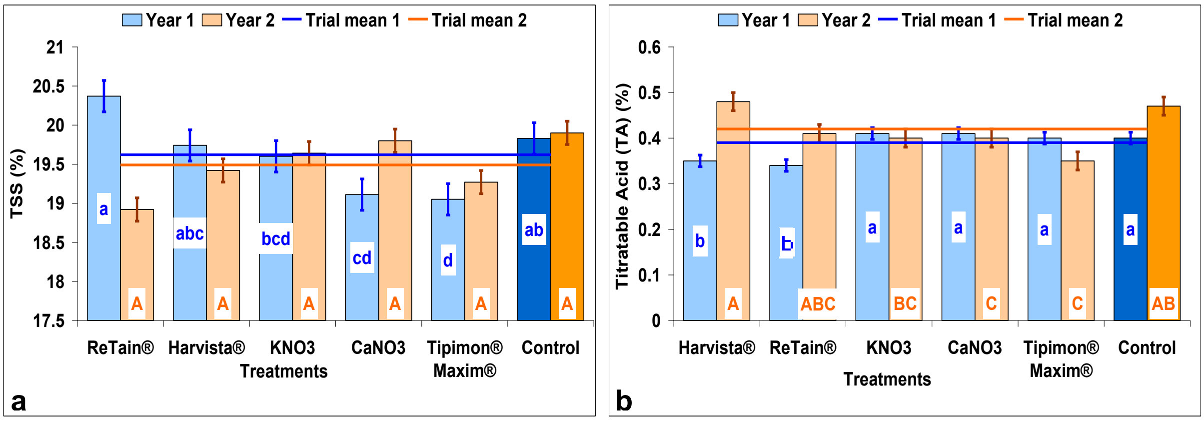
Harvista® resulted in significantly decreased seed mass (Figure 7e) in the 2006 trial, but other chemicals applied were more effective at enhancing fruit -, flesh - and rind mass as well as fruit size. This trend was not repeated in the 2007 trial, with application of Harvista® resulting in increased seed mass. Although fruit size, flesh - and fruit mass were also increased by Harvista® in 2007, all other products were once again more effective at enhancing these fruit characteristics.

In both seasons, KNO_3 and $CaNO_3$ were the only two treatments that significantly increased four of the five fruit characteristics, namely fruit size, flesh-, rind- and fruit mass. The Tipimon®-Maxim® combination treatment managed to significantly increase fruit- and flesh mass in the two consecutive seasons. Other fruit characteristics were also increased by the treatment, although only those increases obtained in 2007 were significant.

3.3.5.3 Quality characteristics, in Year 2, of single product applications selected from Year 1

Figure 8a-c shows quality characteristics of 'Wai Chee' litchi fruit flesh as influenced by different chemical applications over two consecutive seasons.

During 2006 ReTain® resulted in the only tendency to increase TSS percentage (Figure 8a) compared to the control. However, this was not statistically significant. During the 2007 season none of the treatments showed increases in TSS percentage.



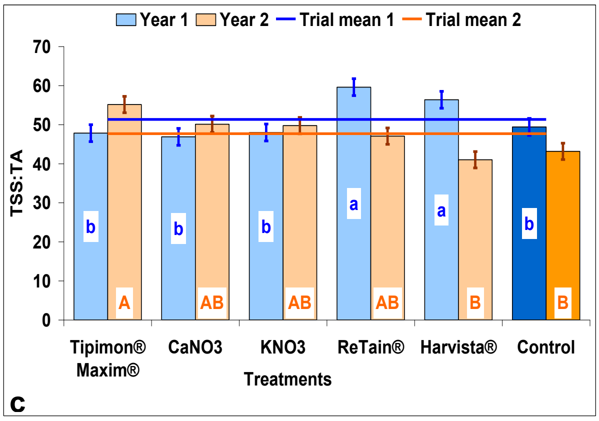


Figure 8a-c
Quality characteristics (TA, TSS and TSS: TA ratio) of 'Wai Chee' litchi fruit at Nelspruit as influenced by five different chemical applications repeated during two consecutive seasons (2006 and 2007).

Different letters indicate significant differences at P≤ 0.05. Lower case letters represent Year 1 (2006) and upper case letters Year 2 (2007).

(TSS=Total Soluble Solids and TA=Titratable Acidity)

Regarding titratable acid (TA) as a quality parameter (Figure 8b), no significant increase in TA was found with any of the treatments applied during 2006. A statistically significant decrease in TA, occurred with the ethylene inhibitor treatments Harvista® and ReTain® in the 2006 season, when compared to the control. During the 2007 season, TA percentage was slightly higher with the

Harvista® treatment and slightly lower with the ReTain® treatment compared to the control. However, these higher and lower TA percentages were not statistically significant. Significantly lower TA compared to the control was found with $CaNO_3$ and the combination of Tipimon® - Maxim®, applied during the 2007 season.

During the 2006 season, the ethylene inhibitors ReTain® and Harvista® resulted in significantly higher ratios of TSS:TA (Figure 8c) when compared to the control. The increased TSS: TA ratios of the ReTain® and Harvista® treatments are as a result of the lower TA levels in these two treatments. All other treatments tended to display lower ratios than the control, however, these ratios were not statistically significant. During the 2007 season only the Tipimon® - Maxim® combination application caused a significant increase in the TSS:TA ratio compared to the control. The same treatment also displayed significantly lower TA levels than the control. However, no significant differences in TSS:TA ratio were detected in fruit treated with Tipimon® - Maxim® compared to the other treatments. Harvista® showed a lower TSS: TA ratio than the control.

3.3.5.4 Summary of the effect of five selected single chemical applications on 'Wai Chee' litchi, applied over two consecutive seasons at Nelspruit.

The overall effect of the five selected chemical treatments in comparison with the non-chemical treated control on 'Wai Chee' fruit at Nelspruit in 2006 and 2007, is summarized in Table 4. Characteristics that were measured included tree-, fruit- and quality characteristics.

Table 4 Summary of the effect of selected treatments applied to 'Wai Chee' litchi at Nelspruit for yield, fruit size and quality enhancement in comparison with the control, repeated over two years (2006 and 2007).

Product	Tipimon® - Maxim®		KNO_3		$CaNO_3$		Harvista®		ReTain®	
Year	2006	2007	2006	2007	2006	2007	2006	2007	2006	2007
Yield	Same	Same	Same	Same	Same	Same	Same	Same	Higher	Same
Fruit Size	Same	Higher	Higher	Higher	Higher	Higher	Lower	Higher	Same	Higher
Fruit Mass	Same	Higher	Higher	Higher	Higher	Higher	Lower	Higher	Same	Higher
Rind mass	Same	Higher	Higher	Higher	Higher	Higher	Same	Higher	Same	Higher
Flesh Mass	Higher	Higher	Higher	Higher	Higher	Higher	Same	Same	Higher	Same
Seed Mass	Same	Higher	Same	Higher	Same	Higher	Lower	Higher	Same	Higher
TSS	Lower	Same	Same	Same	Lower	Same	Same	Same	Same	Same
Acid	Same	Lower	Same	Same	Same	Lower	Lower	Same	Lower	Same
TSS:acid	Same	Higher	Same	Same	Same	Same	Higher	Same	Higher	Same

3.3.6 Programmes combining the best chemical products

Figures 9 to 11 show a comparison between four single chemical applications and their combined effect when included in two sequential programmes on different tree-, fruit- and quality characteristics in 'Wai Chee' litchi. The two programmes included selected nutrition-, synthetic auxin- and ethylene inhibitor treatments of the 2006 trial, applied in sequence at different fruit developmental stages. The time of application for each product is shown in Table 1. Programme 1 consisted of KNO_3 followed first by a combination application of Tipimon® and Maxim® and then by Retain®, while Programme 2 consisted of $CaNO_3$ followed first by a combination application of Tipimon® and Maxim® and then by Retain®.

3.3.6.1 Tree characteristics as influenced by two different chemical application programmes

The effects of four single chemical applications, compared to their combined inclusion in two chemical programmes on yield, as a tree characteristic of 'Wai Chee' litchi at Nelspruit in 2007, are shown in Figure 9. There were no significant differences in yield between any of the treatments.

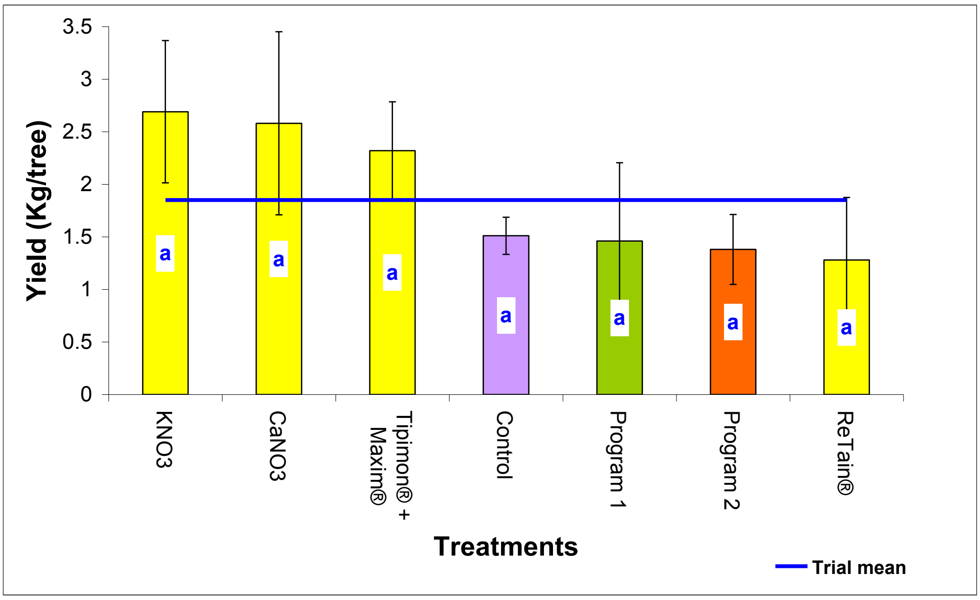
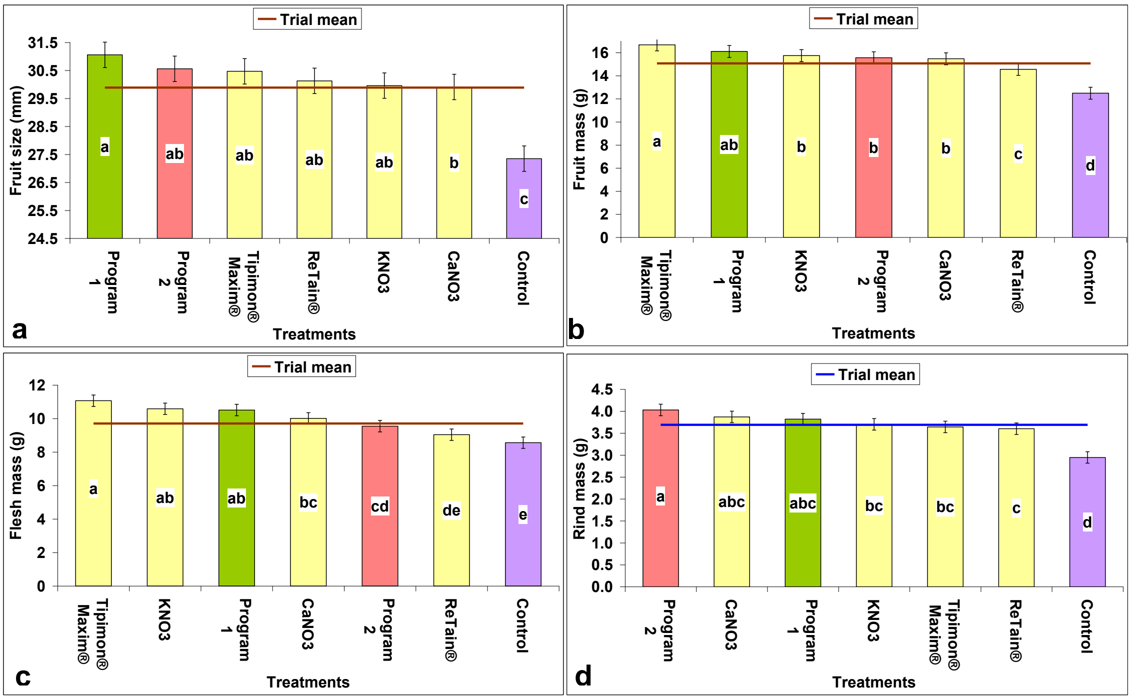


Figure 9 Yield (kg/tree) of 'Wai Chee' litchi at Nelspruit as influenced by four single chemical applications and their combined inclusion in two chemical programmes in 2007. Programme 1 consisted of KNO_3 followed first by Tipimon®-Maxim® and then by Retain®. Programme 2 consisted of $CaNO_3$ followed first by Tipimon®-Maxim® and then by Retain®. Different letters indicate significance at $P \leq 0.05$ (LSD=4.900). The green, red and purple bars represent Programmes 1, 2 and control respectively.

3.3.6.2 Fruit characteristics as influenced by two different chemical application programmes

The effects of four single chemical applications, compared to their combined inclusion in two chemical programmes on the fruit characteristics at harvest, are shown in Figure 10a-e. All of the applied treatments had a significant effect on the improvement of fruit size, fruit mass, rind mass and seed mass as compared to the control. This is also true for flesh mass, except with regard to the Retain® treatment, where there was no significant difference when compared to the control (Figure 10c).



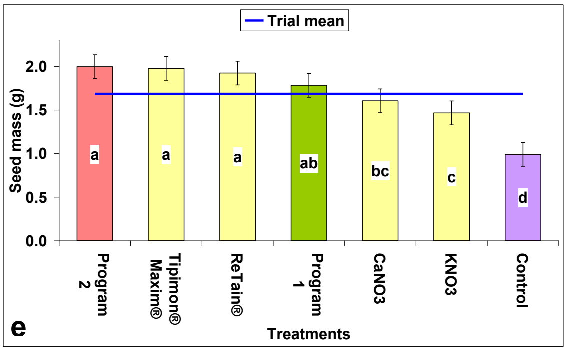


Figure 10a-e

Effect of four single chemical applications and their combined inclusion in two chemical programmes on five different fruit characteristics of 'Wai Chee' litchi at Nelspruit in 2007.

Different letters indicate significance at $P \leq 0.05$ (LSD= 4.900). The green, red and purple bars represent Programmes 1, 2 and control respectively.

There were also no significant differences between Programme 1 and 2 in all instances, except with regard to flesh mass (Figure 10c), where fruit from Programme 1 resulted in a flesh mass significantly higher than fruit from Programme 2.

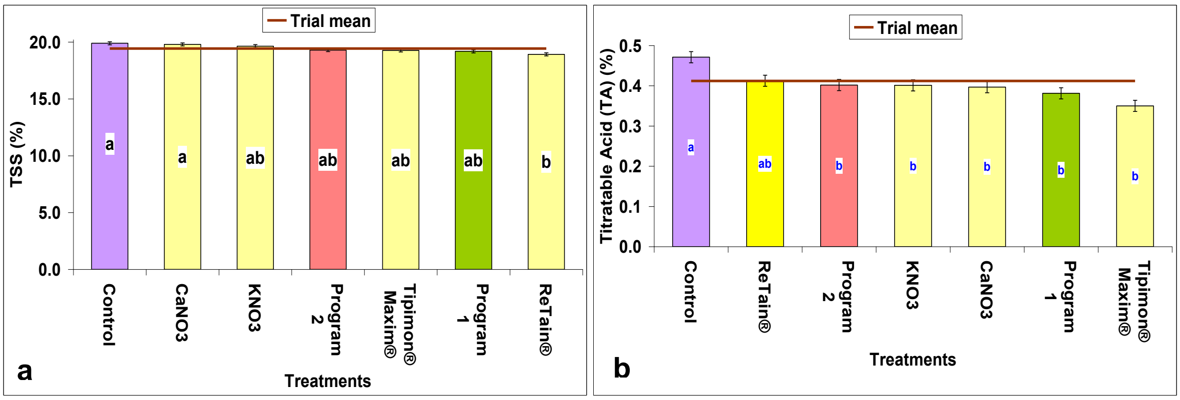
Programme 1, as opposed to its three single chemicals KNO_3, Tipimon®-Maxim®, and Retain®, did not differ significantly with regard to increasing the fruit size (Figure 10a) and rind mass (Figure 10d). With fruit mass (Figure 10b) and flesh mass (Figure 10c), there were also no significant differences amongst Programme 1 and the single applications of Tipimon®-Maxim® and KNO_3. However, a significantly lower fruit- and flesh mass (Figure 10b-c) were obtained by the single application of Retain®. With regard to seed mass (Figure 10e), it was only KNO_3 of the three single applications that caused a significantly lower seed mass than Programme 1.

Programme 2, as opposed to its three single chemicals $CaNO_3$, Tipimon®-Maxim®, and Retain®, did not differ significantly with regard to increasing the fruit size (Figure 10a). Fruit mass and flesh mass (Figure 10b-c) were significantly higher with the single applications of Tipimon®-Maxim® as opposed to the combined Programme 2. The $CaNO_3$ treatment results did not differ significantly from that of Programme 2 except for having a significantly lower seed mass (Figure 10e). Single applications of Retain® caused significantly lower fruit- and rind mass (Figure 10b & 10d). Rind mass (Figure 10d) of the single applications of Tipimon®-Maxim® and Retain®, although not significantly different from each other, were significantly lower than Programme 2, but not significantly lower than $CaNO_3$.

3.3.6.3 Quality characteristics as influenced by two different chemical application programmes

The influence of four single chemical applications and their combined inclusion in two chemical programmes on the quality components (TSS and TA) of ‘Wai Chee’ litchi at Nelspruit is shown in Figure 11.

There were no significant differences between Programme 1 and 2 in all instances. None of the applied treatments had a significant effect on the improvement of TSS percentage (Figure 11a) as opposed to the control. Only ReTain® had a significantly lower TSS reading than control fruit. Significant decreases in TA percentage (Figure 11b) when compared to the control were caused by all applications except for Retain®. With regard to TSS:TA ratio (Figure 11c), Tipimon®-Maxim® was the only treatment with a significantly higher ratio than the control.



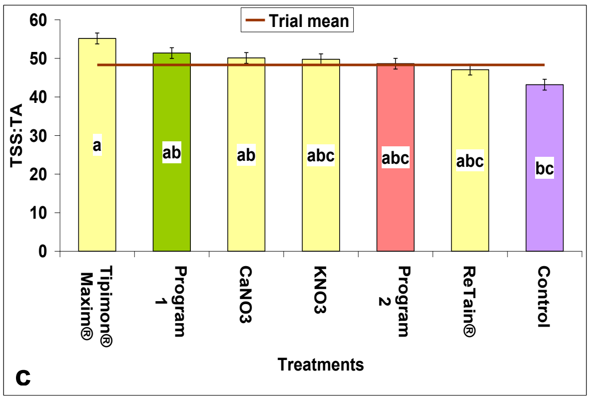


Figure 11a-c

Effect of four single chemical applications and their combined inclusion in two chemical programmes on quality characteristics of 'Wai Chee' litchi at Nelspruit in 2007.

Different letters indicate significance at P≤ 0.05 (LSD= 4.900). The green, red and purple bars represent Programmes 1, 2 and control respectively. (TSS=Total Soluble Solids and TA=Titratable Acid)

3.4 DISCUSSION

3.4.1 Single product applications

3.4.1.1 Tree characteristics

A significant increase in yield (on a per tree basis) of 'Wai Chee' litchi at Nelspruit was obtained in 2006 with the ethylene inhibitor ReTain®. As ReTain® was only applied two weeks prior to harvest it could not have had a significant influence on fruit set. The increased yield can therefore only be due to better fruit retention, or to an increase in fruit mass. However, results of the study showed no significant increase in fruit mass with applications of ReTain®. As fruit drop was not specifically monitored, it is assumed that the higher yield was the result of better fruit retention. Zhang *et al*. (1997) mentioned a fruit abscission period in certain litchi cultivars in China occurring prior to harvest. ReTain® acts as an ethylene inhibitor by blocking ethylene biosynthesis in plant tissues and as fruit ethylene is known to accelerate abscission of leaves, stems, flowers and fruit, it is quite possible that fruit drop was minimised where ReTain® was applied. However, the same increase in yield with ReTain® was not repeated after applying this chemical during the 2007 season. However, maximum temperatures during the last part of the fruit growth period in 2006 were on average two degrees higher than in the same period in 2007 (data not shown). ReTain® might therefore have reduced fruit abscission prior to harvest in 2006, which can be caused by stress due to high temperatures. There were also no statistical differences in yields of the two applied programmes (where ReTain® was included in the sequential programme in 2007) compared to the control. Further studies on the effect of ethylene inhibitors on yield are therefore necessary to confirm the findings in 2006.

No significant differences were found in the yields of any of the other treatments in both years of applying the chemicals. However, the tendencies towards increased yields in both years with nutrition treatments (KNO_3 and $CaNO_3$) and treatments with synthetic auxins (combination treatment of Tipimon® and Maxim®), may indicate a positive response of 'Wai Chee' litchi towards the application of these substances. This is in accordance with findings of Menzel (2005), indicating the decline of yields when the concentration of elemental nutrients is not at optimal levels. Menzel (1984) also mentions internal nutritional imbalances as a reason for the premature abscission of fruit, thereby reducing the yield potential. The tendencies towards increased yields in both years, although not significant, of treatments with plant growth regulators containing synthetic auxins, confirms findings of Stern *et al.* (2000), showing that auxins can be used to increase litchi yields through higher fruit retention. Different concentrations and optimal timing should therefore be investigated further, since 'Wai Chee' litchi may not react in exactly the same way as the 'HLH Mauritius' cultivar to these plant growth regulators. Further investigations should also consider the difference in environment between South Africa and conditions in Israel and Australia, where most of the previous work was done.

The age of the 'Wai Chee' trees used in the current study was 5 years with completion of the trials in 2007. As the trials were done on young trees in the first commercial 'Wai Chee' orchard in South Africa, a better indication of yield potential may be obtained when using mature trees in full production in follow-up studies.

3.4.1.2 Fruit characteristics

Nutrition treatments, containing nitrogen, potassium and calcium:

Compared to the control, the nutrition treatments of KNO_3 and $CaNO_3$ during both years led to significantly better fruit characteristics such as fruit mass, fruit size, flesh mass and rind mass, indicating the enhancing effects of these nutrients on 'Wai Chee' litchi fruit dimensions. The importance of these elements was mentioned by Zhang *et al.* (1997), emphasising the requirement for nitrogen during early stages of fruit development due to its effects during cell division and the requirement for adequate potassium during the mid-late stages of fruit development. Menzel & Simpson (1987) found potassium to be the most abundant nutrient in litchi fruit. The nutrient was found to be removed in relatively large quantities by cropping, indicating the importance of adequate potassium in a heavy bearer like 'Wai Chee'. With calcium, it was found by Bower (1985) that uptake in fruit is optimal during the first six weeks of fruit development, since little or no redistribution occurs after accumulation. Work done by Kift *et al.* (2002) showed that the correct usage of most commercial calcium products during the first six weeks of fruit development improved yield, fruit set and size of litchis, further supporting the findings in the current study. The inclusion of additional foliar applications of KNO_3 and $CaNO_3$ during the period of fruit development may therefore help to prevent shortages of these nutrients, which may have a negative effect on fruit quality.

Plant growth regulators, containing synthetic auxins:

Among the plant growth regulators containing synthetic auxins, the combination treatment of Tipimon® (applied at the 2-3g stage) and Maxim® (applied one week later), showed a tendency for increasing fruit size and fruit mass, with a significant increase in flesh mass obtained after the 2006 season application. During the 2007 season all of these parameters were significantly increased, thereby verifying the enhancing effect of the treatment on fruit dimensions.

The tendency towards increased yield in both years with this combination treatment, although not statistically significant, did not prevent the increase in fruit size and mass in 'Wai Chee' litchi. This confirms work of Wittmann (2002) on the 'HLH Mauritius' cultivar in South Africa. Similar results were obtained by Stern *et al.* (2001) in China, finding that the two auxins combined showed an increase in yield as well as fruit mass in the cultivars 'Fei Xi Xiao' and 'Hei Ye'. However, the researchers also found a significant increase in fruit size and mass after application of Maxim® alone in several litchi cultivars (Stern *et al.,* 2000, 2001). Stern *et al.* (2000) also found that Tipimon® alone did not have any effect on litchi fruit size, although fruit drop was reduced in 'Mauritius' litchi. This suggests that the combination effect of these two products has the potential to increase yield in 'Wai Chee' litchi, without having a negative effect on fruit size.

Stern *et al.* (2000) found in Israel that Maxim® increased the percentage of heavy seed in fruits, while Tipimon® increased the percentage of light seeds. With the Nelspruit study, it was found after the 2006 seasons' application that the combination treatment of Tipimon® and Maxim® slightly decreased seed mass, while after the second application in 2007 seed mass significantly increased. This significant increase in seed mass in the second year may indicate that the seed mass increasing effect of Maxim® might have an overriding effect on the ability of Tipimon® to reduce seed mass when the products are used in combination. Overall, the combination application of Tipimon® and Maxim® proved to have a better effect on yield and fruit size than with their individual effect, suggesting that future investigations could focus on different dosages and application times for the combined treatment.

Plant growth regulators, containing ethylene inhibitors:

With the plant growth regulator containing the ethylene inhibitor 1-MCP, the 2006 application significantly decreased fruit size and –mass, whilst the 2007 application significant increased the size and mass of fruit. Rind and seed mass were also significantly increased by applications of Harvista® in the 2007 year. Litchi is a non-climacteric fruit and it is therefore expected that the ethylene inhibitors would have no physiological effect on the aril. However, it has been shown by Hu *et al.* (2005), that 1-MCP does have an effect on the activity of anthocyanase in the litchi pericarp, thereby delaying abscission. It may therefore be possible that Harvista® may slow down the maturation rate/senescence of the pericarp whilst fruit are on the tree. This can possibly explain the increase in rind mass in the 2007 year with both ethylene inhibitors. However, the significant decrease in seed mass with the Harvista® treatment after the 2006 application, as opposed to the significant increase after the 2007 application, warrants further investigation.

With the other ethylene inhibitor, ReTain®, only slight increases in fruit size and mass occurred after the 2006 application, whilst after the second year's application significant increases for these parameters as well as for rind- and seed mass occurred. Considering the overall yield increase due to ReTain®, it can be postulated that the higher yield was possibly the result of better fruit retention. This argument is supported by the higher seed mass with ReTain®. Studies in China by Stern *et al.* (2001) indicated that well-developed embryos may help to prevent pre-harvest fruit abscission. It was also shown that the mass of the aril and whole fruit are correlated with the mass of the pericarp, irrespective of the fruit having a normal or aborted seed. The higher seed mass obtained with ReTain® may therefore have caused the increases in fruit size and mass. However, further studies are necessary to support this hypothesis.

3.4.1.3 Quality characteristics

All the treatments had a TSS: TA ratio of between 31:1 and 60:1 in both years, indicating good eating quality, According to standards determined by Underderhill and Wong (1990), fruit in such ratios should have good eating quality.

No significant differences as compared to the control were detected among treatments with nutrition and synthetic auxins regarding the TSS:TA ratio after the 2006 application. However, a significant increase in the ratio was found with the ethylene inhibitor treatments Harvista® and ReTain® after the 2006 application. The two ethylene inhibitor treatments also unexpectedly showed significantly lower values of titritable acids when compared to the control. Ethylene inhibitor treatments should prevent or block the production of ethylene during the ripening process, slowing down ripening and allowing fruit to further increase size and quality. However, in 2006, both treatments of Harvista® and ReTain® had significantly lower TA values and also significantly higher TSS:TA ratios, indicating an advanced stage of fruit maturity. This advanced maturity can possibly be the result of data collection during the 2006 season stretching over four days to accommodate 12 treatments, with fruit samples of the ethylene inhibitors being the last to be collected. On the contrary, when it was possible to collect fewer treatments in two days during the 2007 year, no significant difference with the ethylene inhibitors as opposed to the control were observed.

Further research on the effect of ethylene inhibitors on fruit maturity in litchis is therefore necessary, since results of the two seasons did not correspond. The response of other cultivars can also be investigated in order to gain a better understanding of the effect of ethylene inhibitor treatments on litchis in the orchard. A new avenue of research to test the effect of ethylene as opposed to ethylene inhibitors on litchi fruit on the tree can be opened.

The synthetic combination treatment of Tipimon® and Maxim® was the only treatment applied during the 2007 season to have resulted in a significantly higher TSS:TA ratio than the control. Although an increasing tendency was also found during the 2006 year, this was not statistically significant. However, the main reason for applying synthetic auxins is the reduction of fruit drop and to increase fruit mass in litchi, with its effect on internal quality being less important.

3.4.2 Programmes combining the best chemical products

Fruit characteristics of ‘Wai Chee’ were significantly improved with applications of Programmes 1 and 2, consisting of the best performing chemicals, included in a sequential programme. However, when comparing the performance of single products on their own with when they are included in the sequential programme, it is evident that their performance does not differ significantly from each other. However, the margin of performance was found to be bigger with the programme treatments compared to the single product treatments. Whether this additional margin of performance obtained is economical, is a matter for further investigation. Regarding yield and quality characteristics of fruit, no clear advantage was obtained by using either sequential programme as opposed to the use of single products.

3.5 CONCLUSION

The results in this study have indicated a positive response of ‘Wai Chee’ litchi towards different foliar applications of nutrition and plant growth regulators containing synthetic auxins. Additional foliar nutrient applications of nitrogen, potassium and calcium during the early stages of fruit set and development can improve fruit set and subsequently yield, and can also increase fruit mass, fruit size and flesh mass. Potassium nitrate (KNO_3) and Calcium nitrate ($CaNO_3$) proved to be the most enhancing nutrient applications, being important in the early to mid stages of fruit development. From a commercial point of view, Potassium nitrate (KNO_3) and Calcium nitrate ($CaNO_3$) showed a clear beneficial result over two seasons by significantly enhancing fruit characteristics such as fruit size, fruit mass, rind mass and flesh mass of ‘Wai Chee’. The positive effect of these treatments on different tree-, fruit- and quality characteristics showed that these applications can be beneficial when included in an orchard management programme for ‘Wai Chee’ litchi.

The combination application of the synthetic auxins Tipimon® and Maxim® during the 2-3 g stage of fruit development improved fruit size, fruit mass, rind mass and flesh mass, although the improvements were only significant after the second season’s application in 2007. At the same time the flesh-to-fruit ratio can be increased through reduced seed mass, as was found by studies done in Israel on ‘HLH Mauritius’. However, this reduction in seed mass with synthetic auxins was not found in the current study on ‘Wai Chee’ litchi, suggesting that different cultivars have a different response towards these chemicals. Different concentrations and timing of synthetic auxins also need to be further evaluated.

The influence of naturally-occurring auxins and cytokinins showed some tendencies on most of the parameters in 2006, but these influences were not statistically significant. These products were therefore not considered for further evaluation in the following year. However, different concentrations and/or application times could be investigated further in order to determine the value of these substances towards litchi production practices.

The potential of ethylene inhibitors to delay harvesting time and thereby improving size and quality is not clear at this stage. Conflicting results over two seasons necessitates further, more in-depth research in this field.

No clear advantage was found with the use of the sequential programmes as opposed to the use of single products on tree-, fruit- and quality characteristics in ‘Wai Chee’. Although the margin of performance was found to be greater with the programme treatments compared to the single product treatments on some of the evaluated characteristics, the economic viability of this additional margin of performance does not justify the use of programme applications.

CHAPTER 4

4 EFFECT OF DIFFERENT POLLEN PARENTS ON FRUIT RETENTION AND FRUIT CHARACTERISTICS IN 'WAI CHEE' LITCHI

4.1 INTRODUCTION

Litchi *(Litchi chinensis,* Sonn.), which belongs to the Sapindaceae family, has gained much popularity on international markets where it is considered exotic and scarce. The demand is for large, bright red fruit with small seeds and crisp, sweet flesh (Menzel & Simpson, 1990).

In South Africa, substantial growth in the local litchi industry was hampered by a lack of suitable cultivars to extend the very short harvesting season. For nearly a century only two cultivars, namely 'HLH Mauritius' and 'McLean's Red', were cultivated commercially. The commercial viability of litchi farming in South Africa was under constant pressure increasing competition from other Southern Hemisphere countries such as Madagascar, producing during the same time and flooding export markets with their volumes. During the early 1990's the South African Litchi Industry decided to address this situation by importing promising litchi cultivars, mainly from Israel and Australia.

'Wai Chee', the latest commercial litchi cultivar available, was imported amongst others and proved to be precocious and high yielding, but produced medium-sized fruit of average quality. However, the cultivar is harvested at a time in South Africa when there is a gap in worldwide litchi production, thereby increasing its profitability. An obvious field of study would therefore be to investigate ways of improving fruit size and quality to enhance the appeal of 'Wai Chee' on local and overseas markets.

One of the factors found to be involved in yield and fruit size in litchi is pollen source. It is known from pollination studies done in Israel by Stern *et al.* (1993) and by Degani *et al.* (1995) that pollen parent can have an effect on yield, fruit- and seed mass of litchi fruit. It was also determined in other fruit crops such as avocado that certain cultivars are better pollen donors for optimal fruit-set than others (Gazit & Gafni, 1986; Degani *et al.*, 1989).

Before studies on the effects of self- and cross-pollination can be conducted, it is essential to have a clear understanding of litchi flower morphology: A litchi inflorescence bears three different flower types that open in succession on the same panicle (Joubert, 1985; Goren *et al.,* 1998; Kift, 2002b). The first flowers that open are functionally male with well-developed stamens, an aborted ovary and a degenerated pistil. The second wave of flowers to open is functionally female with a well-developed stigma, style, ovary and stamens with very short filaments. The third wave of flowers to open is functionally male with well-developed anthers and rudimentary non-functional female parts. According to Stern & Gazit (2003) there is little overlap of the different flower types on the same inflorescence.

Studies on the effect of self- and cross-pollination in Israel showed that the litchi flowering pattern tends to promote cross-pollination (Stern & Gazit, 2003). However, the partial overlap between the female flowering stage and the two male stages enables pollination among flowers on the same tree, or among trees of the same cultivar, thereby providing an opportunity for self-pollination (Joubert, 1986; Stern & Gazit, 1996). Litchi is considered to be self-fertile, since single cultivar litchi orchards are capable of producing good yields, as in the case of 'Brewster' in Florida (Campbell & Malo, 1968), 'HLH Mauritius' in South Africa (Joubert, 1986), 'Wai Chee' in China (McConchie & Batten, 1989) and 'Haak Yip' in Taiwan (Batten & McConchie, 1992). Hand-pollination studies in South Africa (Fivaz & Robbertse, 1995), Israel (Stern & Gazit, 1998), and Australia (Batten & McConchie, 1992) confirmed the self-compatibility of several litchi cultivars. The use of self- or cross-pollination had in most cases no significant effect on fruit set. However, some pollen parents increased fruit set in certain cultivars in Australia (Stern & Gazit, 2003). Hand-pollination in 'Bengal' with self- and four cross-pollinating cultivars resulted in noticeable differences in fruit mass among the five progeny. One was heavier and two were lighter than the selfed progeny (McConchie *et al.*, 1991), indicating that cross pollination is not always more effective than self-pollination (Stern & Gazit, 2003).

Determination of the pollen parent's identity by isozyme analysis confirmed the self fertility of 'Floridian' and 'Mauritius' in commercial orchards in Israel (Stern *et al.*, 1993; Degani *et al.*, 1995). However, it also demonstrated the importance of cross-pollination in litchi. Almost all fruit on trees close to a second cultivar were hybrids. In addition, there was a pronounced selective abscission in favour of hybrid fruit retention over self-pollinated ones. Stern & Gazit (2003) stated that about a month after fruit set, the hybrid rate in a 'Mauritius' plot was 30 per cent, increasing to 76 per cent at harvest. They also stated that the survival percentage of the hybrid fruitlets may derive from inbreeding depression. Sedgley and Griffin

(1989) found that self-pollinated progeny often have less vigorous embryos than cross-pollinated progeny.

'Mauritius' yield was not correlated with proximity or hybrid percentage to 'Floridian' trees, indicating that cross-pollination by 'Floridian' has no advantage over self-pollination. In contrast, 'Floridian' yield was higher close to 'Mauritius' and a significant positive relationship was observed between the yield and hybrid percentage or proximity to 'Mauritius', indicating a beneficial effect of cross-pollination by 'Mauritius' (Degani *et al*., 1995).

The aim of this study was to investigate the effect of different pollen donors on fruit retention and improvement of fruit characteristics in 'Wai Chee' litchi.

4.2 MATERIALS AND METHODS

4.2.1 Characteristics of the experimental plot

The study was conducted in 2007 on the Agricultural Research Councils' Institute for Tropical and Subtropical Crops' (ARC-ITSC) research farm in the Mpumalanga Province, South Africa. The experimental site consisted of 'Wai Chee' (*Litchi chinensis* Sonn.) trees, all grown from air-layers. The orchard was established in 1994 and is situated near Nelspruit (25° 27' S, 30° 58' E) in the eastern, subtropical Lowveld of Mpumalanga. The Nelspruit area is characterised by a cool and dry winter and a warm, humid summer. Soil in the orchard is a sandy Glenrosa type (Macvicar *et al.,* 1991) with a pH of 6.8.

Trees were spaced 7m apart within rows and 7m between rows (204 trees/ha). Healthy trees, uniform in canopy size and flower intensity, were included in the trial design.

A micro-irrigation system supplied water, scheduled once a week for 8 hr in summer and once every two weeks for 8 hr in winter. Therefore, an average of 80 ℓ/tree/day in summer and 40 ℓ/tree/day in winter was delivered with 70ℓ/hr micro jets.

Soil applications of nitrogen, phosphate and potassium were applied at rates of 3500 g LAN (limestone ammonium nitrate), 1000 g Supers (phosphate) and 1000 g

KCl (potassium chloride) per tree per annum. Leaf applications of zinc, boron and copper were applied annually in August at rates of 200g Zinc oxide/100ℓ water, 100g Solubor/100 ℓ water and 200g Copper oxychloride/100 ℓ water.

Commercial orchard management practices for litchis (Froneman & Husselman, 2002) were maintained throughout the duration of the trial.

4.2.2 Treatment details

4.2.2.1 Trial layout

The trial was laid out as a complete randomised design. Twenty panicles, five each chosen at random per pollen donor, were marked on a single 'Wai Chee' tree that was used as the female parent. Pollen of male donor parents 'HLH Mauritius', 'McLean's Red', 'Fay Zee Siu' and 'Wai Chee', were each applied to all receptive female flowers per panicle, after which the panicle was enclosed to exclude cross contamination. Replications comprised of nine single trees. The aim was to determine the effect of self- and cross-pollination on fruit retention and fruit characteristics in 'Wai Chee' litchi.

4.2.2.2 Pollen collection

Pollen of male parents 'HLH Mauritius', 'McLean's Red', 'Fay Zee Siu' and 'Wai Chee' was collected by placing ten inflorescences of each cultivar on separate clean glass surfaces after which it was left overnight. Pollen grains of each cultivar, that shed overnight onto the glass were collected in the morning and placed in separately marked glass vials for the day's pollination. Fresh pollen, not older than 24 hours (Figure 1), was used every day throughout the trial duration.

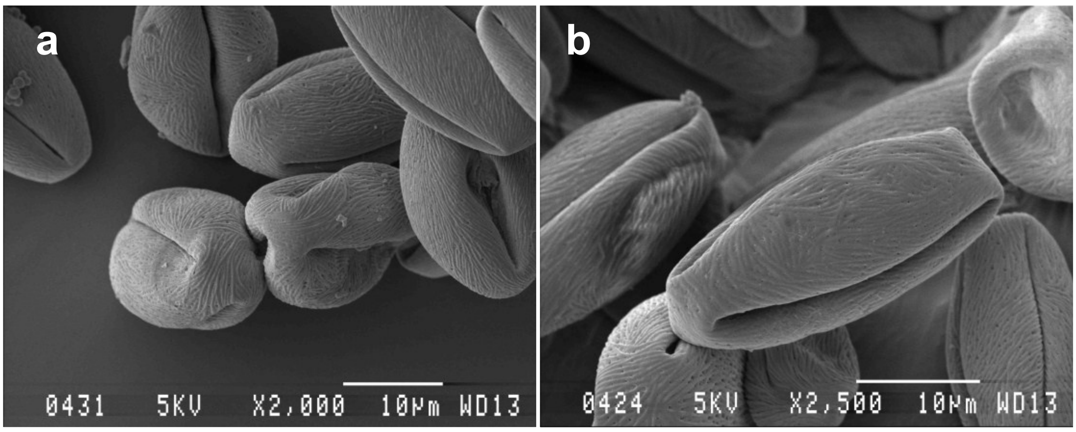


Figure 1 Electron Microscope (SEM) pictures showing Male 2 pollen of (a) 'Wai Chee' and (b) 'HLH Mauritius' flowers, less than 24 hours after collection.

Pollen was primarily obtained from M2 flowers of the pollen donor cultivars, coinciding with the late female bloom of ‘Wai Chee’. According to Mustard *et al.* (1953), about three times more M2 pollen germinated than M1 pollen in the hanging-drop test. Similar effects in several cultivars were also reported in South Africa (Fivaz *et al.,* 1994) and in Israel (Stern & Gazit, 1998).

4.2.2.3 Pollination methods

Flower panicles of ‘Wai Chee’ were enclosed with perforated nylon pollination bags in order to prevent unwanted cross-pollination by insects or wind. The panicles were covered before any of the flowers on the panicles were open. Pollination was performed at anthesis when the surface of the stigma of female flowers was shiny white. The stigma of an individual female flower remains receptive until the colour turns to white-brown, usually after two to three days (McConchie & Batten, 1989). During daily inspections, bags were removed and receptive female flowers were hand-pollinated with a small artist’s paint brush (Figure 2).

Figure 2 Hand pollination of receptive female flowers (a) and the subsequent fruit set induced by each male parent (b).

If any newly opened male flowers were noticed, these flowers were mechanically removed from the inflorescence to prevent self-pollination. This was seldom necessary, since the three sexes were separated and were produced successively on a given inflorescence. After pollination, inflorescences were covered again until the procedure was repeated after 24h during the following morning. This routine was continued for all inflorescences until all receptive female flowers on individual inflorescences were pollinated.

4.2.3 Data collection

4.2.3.1 Fruit set and retention

Data from 'Wai Chee' panicles was taken by counting the number of fruit set per replication after the pollination process on a daily basis. Initial fruit set was regarded as all the fruit per panicle at the end of male 2 flowering. After initial fruit set, the drop of fruitlets on individual panicles was monitored by counting the number of fruitlets retained per panicle on a daily basis until harvest to determine fruit retention.

4.2.3.2 Fruit characteristics evaluation

At harvest, fruit retained on hand-pollinated panicles were evaluated to determine the effect of pollen donor on different fruit characteristics of 'Wai Chee'. Fruit from the different pollination treatments was weighed and fruit size was determined by measuring the shoulder width with an electronic hand caliper (accurate up to 0.01mm). Seed and peel of fruit were also weighed.

Total soluble solids (TSS) were determined with a temperature-compensating digital refractometer (Palette PR-101) and titratable acidity (TA) by titration of a 10 ml aliquot of juice using 0.1 N NaOH to an endpoint with phenolphthalein as an indicator (Wardowski *et al*, 1979). The TSS: TA ratio of the pulp is known to be well correlated to mean eating quality (Underhill & Wong, 1990).

4.2.4 Experimental design and statistical analyses

Data were statistically analysed as a completely randomised design with 4 treatments and 9 replications. Analysis of variance (ANOVA) was used to test for differences between the four pollen treatment effects. The data were acceptably normal with homogeneous treatment variances. Treatment means were separated using Fisher's protected t-test least significant difference (LSD) at the 5% level of significance (Snedecor & Cochran, 1980). Data were analysed using the statistical program GenStat (2003).

4.3 RESULTS

4.3.1 Initial fruit set and final fruit retention

A trend of pollen donor effect on fruit set and subsequent fruit retention of 'Wai Chee' on 45 flower panicles per treatment is demonstrated in Figure 3 and the statistical differences are illustrated in Figure 4.

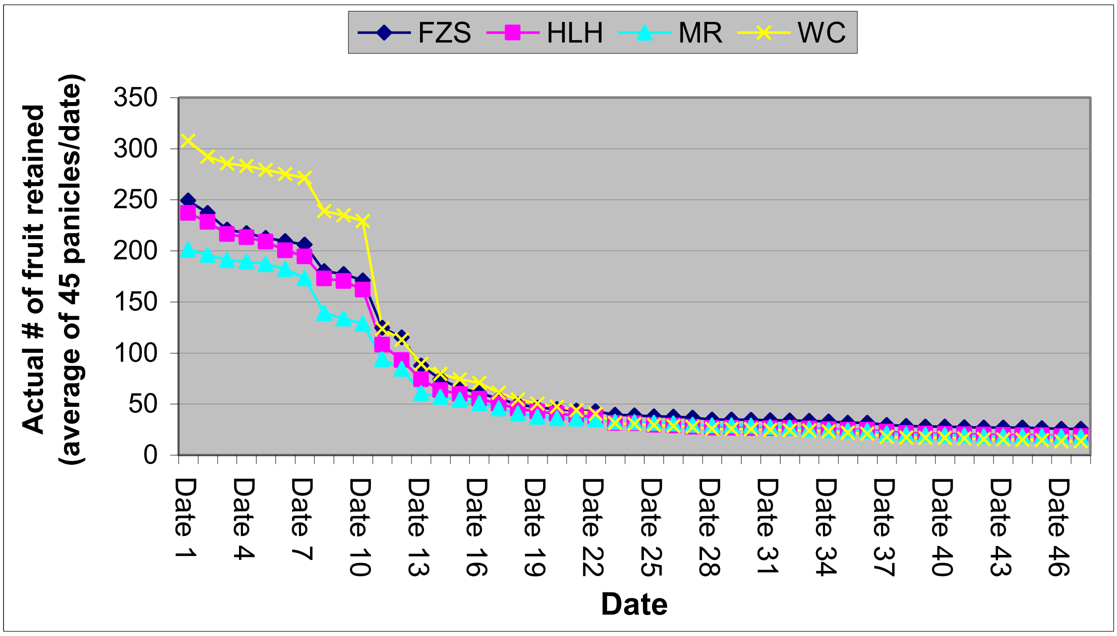


Figure 3 **Trend of pollen donor effect on number of fruit retained daily on 'Wai Chee' (WC) panicles from fruit set (Date 1) to harvest (Date 47). Data are means of 45 panicles per treatment (5 panicles x 9 trees) of male parents 'HLH Mauritius' (HLH), 'McLean's Red' (MR), 'Fay Zee Siu' (FZS) and 'Wai Chee' (WC).**

Initial fruit set in 'Wai Chee' panicles was lower in all cross-pollinating treatments compared to initial fruit set in self-pollination with 'Wai Chee' (Figure 4). However, the opposite was found in final fruit retention, with higher retention in all the cross-pollinating treatments as opposed to self-pollination with 'Wai Chee' (Figure 4).

Compared to self-pollination with 'Wai Chee' pollen, a significantly lower ($P<0.05$) initial fruit set (33% and 30%) was found with pollen donors 'McLean's Red' and 'HLH Mauritius' respectively (Figure 4). However, 'Wai Chee' as pollen donor retained significantly less fruit at harvest than cross-pollinating treatments 'Fay Zee Siu' and 'McLean's Red'.

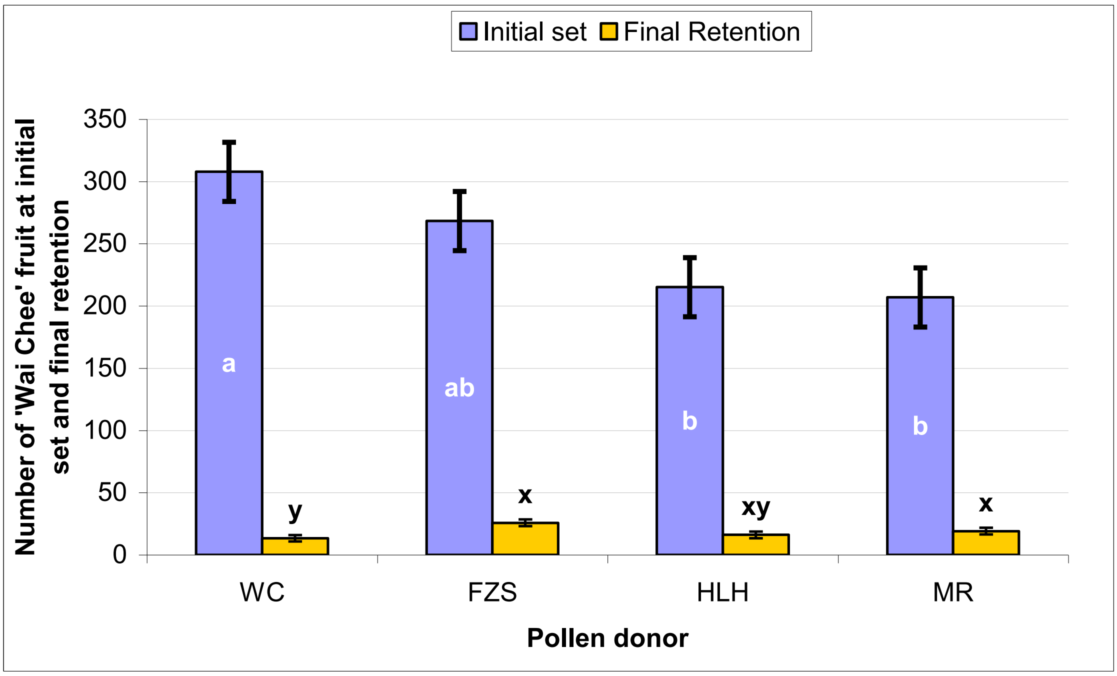


Figure 4 **Effect of pollen donor on the initial fruit set and final fruit retention of the cultivar 'Wai Chee' (WC). Data are means of 45 panicles per treatment (5 panicles x 9 trees) of male parents 'HLH Mauritius' (HLH), 'McLean's Red' (MR), 'Fay Zee Siu' (FZS) and 'Wai Chee' (WC). Within the initial set and final retention respectively, bars of means denoted by different letters differ significantly ($P \leq 0.05$).**

Pollen donors 'Fay Zee Siu' and 'McLean's Red' retained significantly more fruit compared to self-pollination with 'Wai Chee', showing increases of 48% and 30% respectively in final retention. 'HLH Mauritius' as donor did not significantly retain more fruit than 'Wai Chee' or less fruit than 'Fay Zee Siu' and 'McLean's Red' (Figure 4).

4.3.2 Influence of pollen donor on 'Wai Chee' fruit retention during the fruit development period

In order to determine the significance of pollen donor between dates 2 and 47, the percentage fruit retained on each date relative to initial fruit set was analysed. In Figure 3 the fruit drop between dates 9 and 17 seems to be pronounced, but after statistical analysis of the percentage fruit retained on each date relative to initial fruit set as displayed in Figure 5, it became clear that pollen donor had no significant influence during this period. However, in the periods between date 3 and 6, date 26 and 32 and date 36 to 47, pollen donor did have a significant influence on fruit retention. From Figure 5, it is evident that the initial high percentage of fruit retained with self-pollinating donor 'Wai Chee' decreased to the lowest percentage fruit retained at harvest as opposed to higher fruit retention related to the cross-pollinating donors. During the peak fruit drop period from date 9 to date 17, the percentage fruit drop of pollen donor 'Wai Chee' was also more pronounced than fruit drop in the cross-pollinating donors, although the differences in this period were not statistically significant.

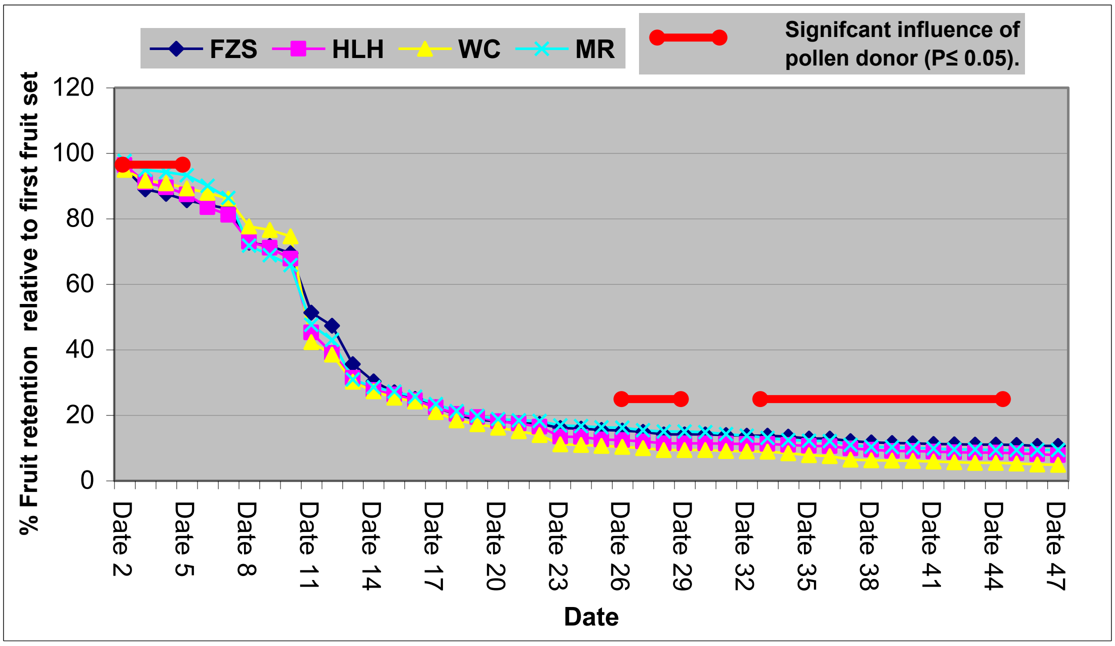


Figure 5 **Effect of pollen donor on percentage fruit retained in 'Wai Chee' (WC) relative to the initial fruit set. Percentages were calculated as the number of fruit retained relative to the initial fruit set on Date 1. This was done on a daily basis until harvest (Date 47). Data are means of 45 panicles per treatment (5 panicles x 9 trees) of male parents 'HLH Mauritius' (HLH), 'McLean's Red' (MR), 'Fay Zee Siu' (FZS) and 'Wai Chee' (WC). Red horizontal bars denote the dates of which the means differ significantly ($P \leq 0.05$) for the percentage of fruit retained.**

4.3.3 Pollen donor and fruit characteristics

4.3.3.1 Fruit size

Though it was not significant, slight differences in average fruit size of 'Wai Chee' was found with cross-pollinating donors 'McLean's Red' and 'Fay Zee Siu' , while 'HLH Mauritius' tended to produce the same average fruit size than the self-pollinating donor 'Wai Chee' (Figure 6). Fruits originating from cross-pollination were larger by 8% for 'McLean's Red' and 0.2% for 'Fay Zee Siu' compared to fruit originating from self-pollination.

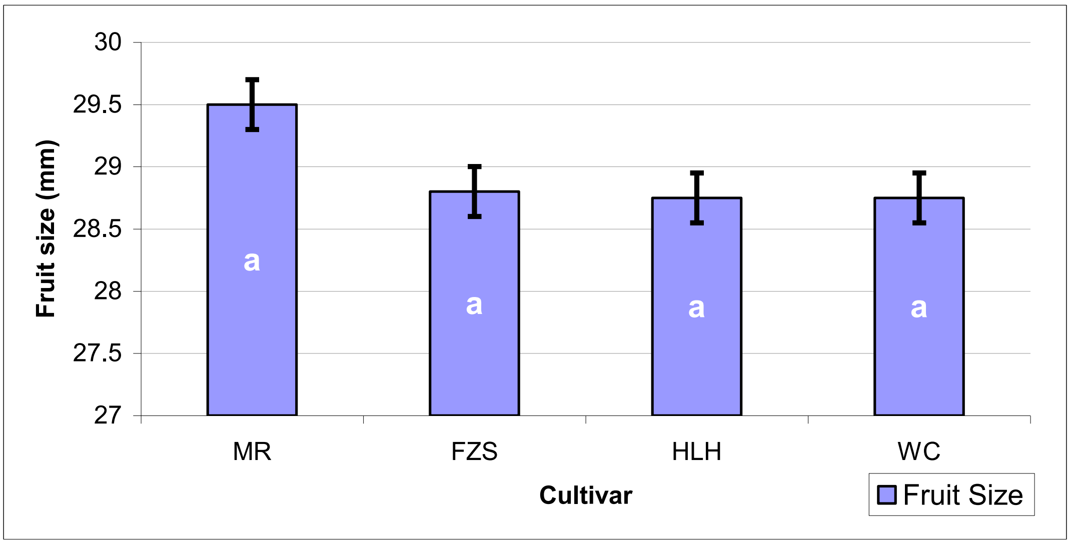


Figure 6 **Effect of pollen donor on fruit size in 'Wai Chee' (WC). Different letters indicate significance (P≤ 0.05). Data are means of fruit retained on panicles (5 panicles x 9 trees) pollinated by male parents 'HLH Mauritius' (HLH), 'McLean's Red' (MR), 'Fay Zee Siu' (FZS) and 'Wai Chee' (WC).**

There was a tendency, although not significant, for a small increase in average 'Wai Chee' fruit mass with cross-pollinating donors 'HLH Mauritius', McLean's Red' and 'Fay Zee Siu' (Figure 7). As was found in fruit size, fruit produced through self-pollination with 'Wai Chee' had the lowest average fruit mass. Fruits originating from cross-pollination were heavier by 8% for 'McLean's Red', 5% for 'HLH Mauritius' and 3% for 'Fay Zee Siu' compared to fruit originating from self-pollination.

Similar tendencies for a slight increase in flesh mass were reflected in treatments with cross-pollinating donors, while the self-pollinating donor 'Wai Chee' again produced fruit with the lowest average flesh mass. Increases in flesh mass of 9% for 'HLH Mauritius', 9% for 'McLean's Red' and 2% for 'Fay Zee Siu' were obtained in fruit originating from cross-pollination when compared to fruit originating from self-pollination. These increases were however not significant.

No clear effect of pollen donor on average rind- and seed mass could be determined, with only slight, non-significant increases or decreases among the different pollen donor treatments. The highest average rind- and seed mass were from pollen donors 'McLean's Red' and 'Fay Zee Siu' and the lowest from 'Fay Zee Siu' and 'HLH Mauritius' respectively.

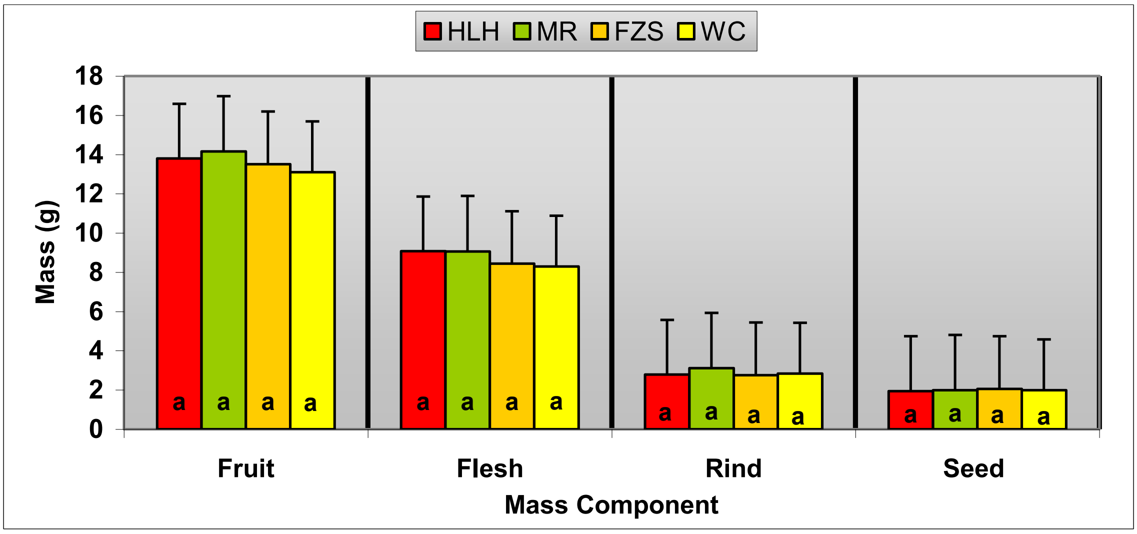


Figure 7 **Effect of pollen donor on fruit-, flesh-, rind- and seed mass in 'Wai Chee' (WC). Different letters indicate significance (P≤ 0.05). Data are means of fruit retained on panicles (5 panicles x 9 trees) pollinated by male parents 'HLH Mauritius' (HLH), 'McLean's Red' (MR), 'Fay Zee Siu' (FZS) and 'Wai Chee' (WC).**

4.3.3.2 Fruit quality

Figure 8 shows total soluble solids (TSS) and acid percentages in resulting 'Wai Chee' fruit after pollination of female flowers with different male donors. Although there seemed to be a trend towards increases in TSS percentage in fruit from cross-pollinating donors 'McLean's Red' and 'HLH Mauritius' as opposed to lower TSS concentrations in fruit from the self-pollinating 'Wai Chee' donor, the differences were not significant. Titratable acid concentrations in fruit resulting from the 'HLH Mauritius' pollen donor appeared to be slightly higher, while fruit from the 'McLean's Red' pollen donor were slightly lower in acid than in fruit from the self-pollinating 'Wai Chee' donor, but again, none of the differences were statistically significant.

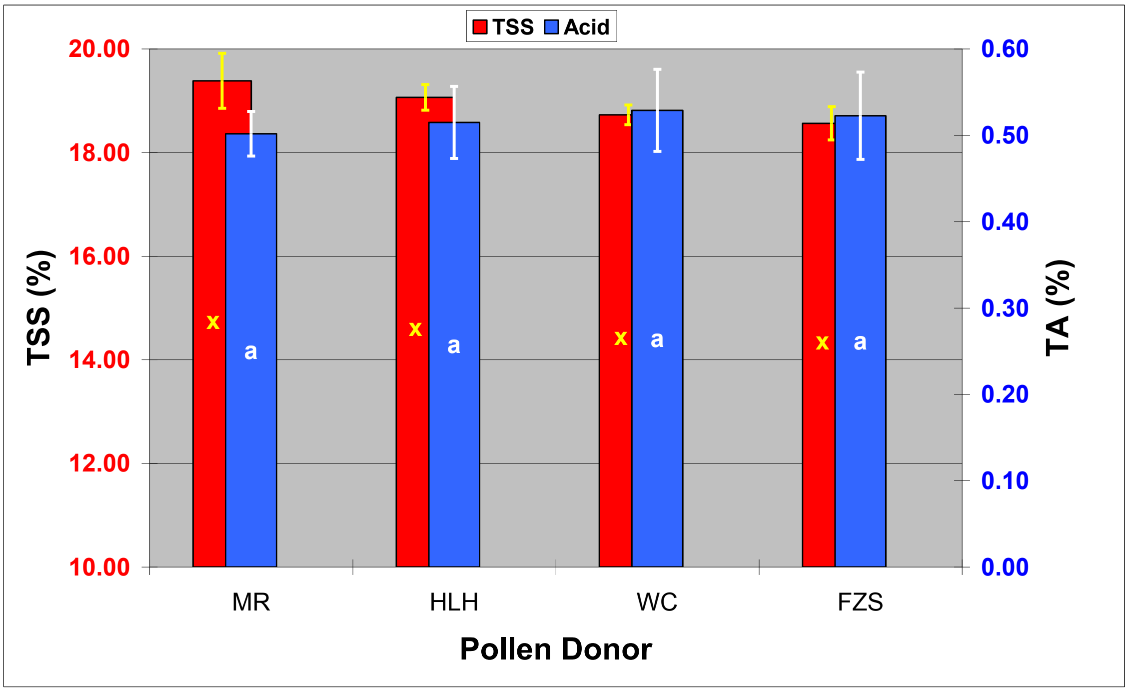


Figure 8 **Effect of pollen donor on Total Soluble Solids (TSS) and Titratable Acid (TA) in 'Wai Chee' (WC). Different letters indicate significance (P≤ 0.05). Data are the means of TSS and TA of fruit retained on panicles (5 panicles x 9 trees) pollinated by male parents 'HLH Mauritius' (HLH), 'McLean's Red' (MR), 'Fay Zee Siu' (FZS) and 'Wai Chee' (WC).**

4.4 DISCUSSION

4.4.1 Effect of pollen donor on fruit set and -retention

In litchi, far more fruit are set than eventually develop to maturity. Usually a great number of fruit drop prematurely, thereby reducing the crop potential. This fruit drop pattern was also clear in 'Wai Chee' after hand pollination of female flowers. The significant effect of pollen donor on litchi fruit set and retention in 'Wai Chee' became evident as the number of fruits that survived throughout the development period was monitored.

Significant initial fruit set from self-pollination in 'Wai Chee' indicated a high level of self-compatibility of the cultivar. In Israel, self-compatibility of litchi cultivars was confirmed with isozyme analysis by Stern *et al.* (1993), mentioning significant fruit set from self-pollination in 'Mauritius' and 'Floridian' orchards. Results in this study showed that self-pollinating 'Wai Chee' gave the highest initial fruit set, but fruitlets emanating from these self-pollinations abscised at a higher rate than the fruitlets resulting from cross-pollination. Contrary to the initial high fruit set obtained with self-pollination, final fruit retention with cross-pollinating donors was all higher than in retention with self-pollination. The significant increases in retention obtained with ‘Fay Zee Siu’ and ‘McLean’s Red’ pollen may be an indication of potential enhancing effects of cross-pollinators on yield in ‘Wai Chee’.

When the percentage fruit retained relative to initial fruit set was analysed, it was found that pollen donor had a statistically significant effect on fruit retention during certain stages of fruit development. Retention of self-pollinated fruitlets was highest in the period just after fruit set, but after a pronounced fruit drop period of three weeks, cross-pollinating donors had a significantly higher influence on fruit retention during the later stages of fruit development. Degani *et al.* (1995) reported that in-bred fruit often abscise early, supporting the findings of this study. The better fruitlet retention with cross-pollination may also be explained by embryo degeneration and abortion found in self-pollinated fruit due to inbreeding depression (Sedgley & Griffin, 1989). Final fruit retention at harvest was higher in all cross-pollinating donors, demonstrating the potential ability of cross-pollinated fruitlets to outcompete self-pollinated fruitlets for available tree resources.

Pollen from M2 flowers of the different pollen donors was used to pollinate ‘Wai Chee’ flowers in this study. However, a different fruit set may have been achieved with pollination by the M1 bloom under orchard conditions. Stern & Gazit (1996) found that M2 pollen is more fertile than M1 pollen. It was further mentioned by the authors that pollination rate is very slow at the start of the ‘Mauritius’ female bloom in Israel when only M1 pollen is available. Similarily, M1 flowers of ‘Tai So’ produced no pollen in the early bloom in Queensland, Australia (Batten & McConchie, 1992), demonstrating the disadvantage of the M1 bloom in litchi fertilization. The greater fertility of M2 pollen, coupled with the greater attractiveness of M2 flowers to insect pollinators, make the M2 bloom more effective for pollination than the M1 bloom (Stern & Gazit, 2003). Therefore, when considering a pollen donor to increase the productivity of another litchi cultivar, the M2 bloom of the pollen donor should overlap with the female bloom of the other cultivar. In the case of ‘Wai Chee’, with a late flowering morphology, it is possible for the M2 flowering period of earlier cultivars such as ‘HLH Mauritius’, ‘Fay Zee Siu’ and ‘McLean’s Red’ to overlap with the female flowering stage in ‘Wai Chee’. Results in the study therefore indicated that interplanting of cultivars to encourage pollen transfer from M2 pollen donors to female flowers could have beneficial effects on litchi production.

4.4.2 Effect of pollen donor on fruit characteristics

Although not significant, fruit characteristics such as fruit size, fruit mass and flesh mass in 'Wai Chee', tended to be increased by the use of cross-pollinating donors. Fruit produced from self-pollination with 'Wai Chee' had the lowest averages regarding these fruit size measurements, indicating potential advantages with cross-pollinating donors. Further studies should therefore verify these positive tendencies, since similar studies done in Israel (Stern *et* al, 1993; Degani *et al,* 1995) and Australia (McConchie *et al.*, 1991) suggested some beneficial effects on fruit mass with cross-pollination. Even though 'Wai Chee' had a significantly higher initial set with self pollination, no significant correlation could be found between fruit set and fruit size.

In the current study, pollen donor had no clear effect on average rind- and seed mass of fruit, with only slight, non-significant increases or decreases among the different pollen donor treatments. Studies on pollen parent effect on fruit characteristics of 'Floridian' and 'Mauritius' litchi in Israel showed that, in both cultivars, fruit- as well as seed mass were affected by the pollen parent. Fruit resulting from cross-pollination were heavier and contained heavier seeds than selfed ones (Degani *et al.*, 1995). In the current study, similar, although non-significant tendencies with cross-pollinators were detected regarding the improvement of fruit mass, but with a less pronounced effect on seed mass.

Stern *et al.* (1993) found that seeds from self-pollinated flowers are more likely to abort than seeds from cross-pollination. This was not noticed with 'Wai Chee' in this study, with normal seeds containing viable embryos at maturity in self- and cross-pollinated fruits. However, 'Wai Chee' usually has a low incidence of aborted seeds, contrary to cultivars such as 'Nuomici' and 'Feizixiao' with higher proportions of aborted seeds. The effect of cross-pollination may therefore be more pronounced on seed mass in 'aborted-' or 'variable seeded' cultivars as was found by Xiang *et al.* (2001) in 'Nuomici' and 'Guiwei' in China.

No significant differences were found in total soluble solids and acid percentages in fruit from self- or controlled pollination. However, tendencies towards increased

TSS percentages in fruit emanating from cross-pollinating donors ‘McLean’s Red’ and ‘HLH Mauritius’ were noticed. Further research on the effect of pollen donor on quality characteristics in litchis is, however, necessary before any meaningful conclusions can be made.

It may well be possible that, with the use of other pollen donor cultivars than the ones used in the current study, different effects on the various fruit characteristics in ‘Wai Chee’ could be found. In view of the beneficial effects obtained in fruit retention with cross-pollination, a repeat of this experiment may therefore be worthwhile in order to further investigate the potential for enhancing fruit dimensions of ‘Wai Chee’ litchis.

4.5 CONCLUSION

Results obtained in this study have shown a positive response of ‘Wai Chee’ towards different pollen donor applications aiming to enhance fruit retention and certain fruit characteristics in the cultivar. Final fruit retention with cross-pollinating donors was all higher than in retention with self-pollination, with significant increases in retention caused by some cross-pollinating cultivars. Results therefore indicated that the inclusion of cross-pollinators in orchards to encourage pollen transfer from fertile M2 flowers of cross-pollinators to the female flowers of ‘Wai Chee’ could have beneficial effects on production. Fruit characteristics such as fruit size, fruit- and flesh mass may also be enhanced by the use of cross-pollinators. Although tendencies towards enhanced fruit characteristics were found in this study, these were not significant. Additional work is therefore necessary in order to confirm the possible beneficial effects of cross-pollinators on fruit characteristics of ‘Wai Chee’, since adequate fruit size is more of an issue in this cultivar than its yielding potential.

Work in this study was mainly focused on the ‘Wai Chee’ cultivar. However, it would be worthwhile to include some other important litchi cultivars in the South African litchi Industry in future investigations to determine the effect of cross-pollination on these cultivars. Due to environmental effects it could also be worthwhile to repeat this trial over consecutive seasons to determine the economic impact of the pollen donor on various fruit characteristics over a period of time.

CHAPTER 5

5 EFFECT OF DIFFERENT BUNCH COVERING MATERIALS ON FRUIT SIZE AND MATURATION OF ‘WAI CHEE’ AND ‘HLH MAURITIUS’ LITCHI

5.1 INTRODUCTION

With the increasing popularity of exotic fruits on world markets, the importance of litchi production has increased. The South African Litchi Industry generally exports an average of 2500 tonnes of fruit (60% of production), of which 95% are marketed in the European Union (PPECB Export Directory, 2006). The demand is high for large, bright red fruit with small seeds and crisp, sweet flesh (Menzel & Simpson, 1990).

In the early 1990's, the South African Litchi Industry initiated efforts to import some promising cultivars, mainly from Australia and Israel. Special attention was given to earlier and later cultivars than the existing commercial ones in order to address a very congested marketing period in South Africa. The late season litchi cultivar 'Wai Chee' was subsequently imported amongst others and proved to be precocious with high yielding potential per hectare in high density plantings. However, small fruit size and average internal fruit quality restrict the potential of this cultivar. The fact that 'Wai Chee' is harvested at a time in South Africa when there is a gap in worldwide litchi production, makes the cultivar potentially very profitable.

One of the methods used to improve fruit size and quality in litchis is the bagging of fruit bunches after fruit set. Quality benefits of bagging have been previously documented in 'Mauritius' litchi by Tyas *et al.* (1998). The study found that bagging has the potential to improve fruit quality and red colouration with no negative impact on yield. Quality loss during fruit growth can also be attributed to insect- and bird damage as well as skin browning. Oosthuizen (1989) reported that solar injury, insect damage and fruit cracking were markedly reduced when 'HLH Mauritius' litchi bunches were covered. Galan Sauco & Menini (1989) mentioned that the advantages of covering fruit bunches outweigh disadvantages such as cost of labour and material because fruit quality, colour and size is better and less labour is required for harvesting. This practice also protects the fruit from hail damage and from certain species of bat.

Galan Sauco & Menini (1989) advised that litchi clusters should be bagged approximately 1.5 to 2 months prior to harvesting. In the Southern Hemisphere fruit clusters are bagged after the 'November drop' period (when fruit are past the 2g. stage) to ensure that the fruit in the bags will be retained and to prevent fruit from rotting inside the bag. It is also advisable to remove most of the leaves around clusters; otherwise they will fall into the bags and rot. De Villiers (2002) mentioned that not too many fruit should be placed in one bag; about 25 per bag are recommended. It is advisable to treat fruit with an appropriate insecticide before bagging.

In South Africa, brown paper bags (Figure 1) were found to be the most suitable and also the most cost effective covering material (De Villiers, 2002).

Figure 1 Litchi trees showing brown paper bags on fruit clusters.

Oosthuizen (1991) reported that an alternative, but labour intensive method for controlling fruit flies, litchi moth, fruit bats and birds, is tying brown paper bags around litchi fruit bunches. It was found that bagged fruit have a much better quality, mainly because of the higher humidity inside the bag. Oosthuizen (1991) stated that humidity on a hot summer day can be up to 30 per cent higher inside the bags than is the case with exposed fruit. It was mentioned that the picking period of such fruit is also extended by up to two weeks, which can have the additional advantage of extending the harvesting period of a late cultivar like 'Wai Chee' even more.

In China and Thailand, newsprint and unspecified paper bags are used as bagging material (Zhang *et al.,* 1997; Sethpakdee, 2002). Bagging experiments with 'Feizixiao' in China (Hu *et al.,* 2001) showed that colour development can be

enhanced by diffuse light. Bags were made of adhesive-bonded fabric, with about 58% of the total sunlight being transmitted through the bag. Potential damage to anthocyanins caused by direct sunlight was reduced by bagging. Under orchard conditions with direct sunlight on fruit, the shaded part of the fruit received almost no light and the synthesis of anthocyanins was inhibited, leading to uneven colouration of fruit. Enclosed litchis growing in the lower canopy in South Africa was found to have better quality than exposed fruit (Huang, 2005).
Barritt *et al*. (1987) and Tombesi *et al*. (1993) mentioned that radiation interception can have an influence on fruit size through overshadowing of leaves, as shown by smaller fruit in the lower canopy. Tyas *et al*. (1998) suggested that radiation interception by the fruit itself may also be important, since it was concluded by Thorpe (1974) that reduced radiation interception (leading to lower fruit temperatures) may reduce their sink strength. Under very high levels of radiation, shading may have the opposite effect of increasing fruit mass, because of reducing the negative effects of too much radiation. Bagging can also affect fruit size by increased relative humidity, and therefore reduced fruit water loss (Tombesi *et al.*, 1993).

The present study aimed at investigating the potential enhancing effects of different bagging materials on maturation and fruit size in ‘Wai Chee’ litchi. As Total Soluble Solids (TSS) in the fruit aril will be used as an indicator of maturation, fruit quality assessment will only be based on visual assessment of skin colour. The ‘HLH Mauritius’ cultivar was included together with ‘Wai Chee’ in the trials in order to investigate possible variation among cultivars in their response to bunch covering.

5.2 MATERIALS AND METHODS

5.2.1 Characteristics of the experimental plot

The study was conducted in 2007 on a research farm of the Agricultural Research Council in the Mpumalanga Province, South Africa. The experimental site consisted of ‘Wai Chee’ and ‘HLH Mauritius’ trees (*Litchi chinensis* Sonn.), all grown from air-layers. The orchard was established in 1994 and is situated near Nelspruit (25° 27’ S, 30° 58’ E) in the eastern, subtropical Lowveld of Mpumalanga. The Nelspruit area is characterised by a cool and dry winter and a warm, humid summer. Soil in the orchard is sandy (Glenrosa type) with a pH of 6.8.

Trees were spaced 7m apart within rows and 7m between rows (204 trees/ha) for both cultivars. Only healthy trees, uniform in canopy size and fruit set, were used for the trial.

A micro-irrigation system supplied water, scheduled once a week for 8 hr in summer and once every two weeks for 8 hr in winter. Therefore, an average of 80 ℓ/tree/day in summer and 40 ℓ/tree/day in winter was delivered with 70ℓ/hr micro jets.

Soil applications of nitrogen, phosphate and potassium were applied at rates of 3500 g LAN (limestone ammonium nitrate), 1000 g Supers (phosphate) and 1000 g KCl (potassium chloride) per tree per annum. Leaf applications of zinc, boron and copper were applied annually in August at rates of 200g/ 100ℓ water Zinc oxide, 100g/100ℓ water Solubor and 200g/ 100ℓ water Copper oxychloride.

Commercial orchard management practices for litchis (Froneman & Husselman, 2002) were maintained throughout the duration of the trial.

5.2.2 Treatment details

5.2.2.1 Trial layout and experimental design

The trial was laid out as a complete randomised block design with 4 blocks and 10 replicates per treatment. Five different treatments, namely a commercially used "Closed Brown" paper bag (with nominal basis mass of 65g/m²), a "Thin Brown" paper sleeve (60g/m²), a "Thick Brown" paper sleeve (80g/m²), a "White Water Resistant" paper sleeve (70g/m²) and a control consisting of uncovered bunches were applied to fruit bunches of 'Wai Chee' and 'HLH Mauritius' respectively. Paper sleeves of 30 cm x 60 cm and closed bags of 20 cm x 37 cm were used to cover whole bunches. Paper sleeves were tied closed with twine above the bunch of fruit, with the open end of the sleeve extending 20 cm below the base of the bunch. In order to retain a higher humidity, the base of the sleeve was closed with a single staple. This also allowed for rotten or abscised fruit to roll out on either side of the staple. The constitution of the overlapping sleeve allowed for closure after such fruit were dispatched. Bunches were covered after mid- November when the peak fruit abscission period for the respective cultivars was completed, when fruit mass was between 3 and 6 g. Treatments were evenly distributed in all wind directions and from top to bottom of the tree canopy. The aim of the trial was to determine the effect of different covering materials on maturity and fruit size in 'Wai Chee' and 'HLH Mauritius' litchi.

5.2.2.2 Data collection

Harvesting: Litchi bunches were harvested on 17 December 2007 for 'HLH Mauritius' and on 14 January 2008 for 'Wai Chee'. After harvest all the fruit were kept in cold storage at 4ºC and were analysed within two days. No mass loss was found during the time of storage.

Evaluation of fruit characteristics: Fruit retained on bunches were evaluated to determine the effect of different bagging materials on maturity, fruit size and quality aspects. Fruit from the different bagging treatments were weighed with a laboratory scale (Shimadzu UX 6200H) to determine fruit mass. Fruit size was determined by measuring the fruit length and the fruit width with an electronic hand calliper (accurate up to 0.01mm). Two measurements on fruit width were taken, namely on the broad (width 1) and on the narrow side (width 2). Total soluble solids (TSS) in the aril were determined with a temperature-compensating digital refractometer (Palette PR-101).

5.2.2.3 Statistical analyses

Data were statistically analysed as a complete randomised design with 5 treatments and 10 replications. Analysis of variance (ANOVA) was used to test for differences between the treatment effects. The data were acceptably normal with homogeneous treatment variances. Treatment means were separated using Fisher's protected t-test least significant difference (LSD) at the 5% level of significance (Snedecor & Cochran, 1980). Data were analysed using the statistical program GenStat (2003).

5.3 RESULTS

5.3.1 Fruit mass

The effect of different bagging materials on fruit mass of 'HLH Mauritius' and 'Wai Chee' is shown in Figure 2. The only significantly increase in fruit mass occurred in 'HLH Mauritius' with the "White Water Resistant" (70 g) sleeve when compared to the uncovered control fruit. All the other bagging treatments showed a tendency for fruit mass increase in 'HLH Mauritius' compared to the control, although these increases were not statistically significant. The same tendency was also observed in 'Wai Chee', with all bagging treatments showing slight, non-significant increases.

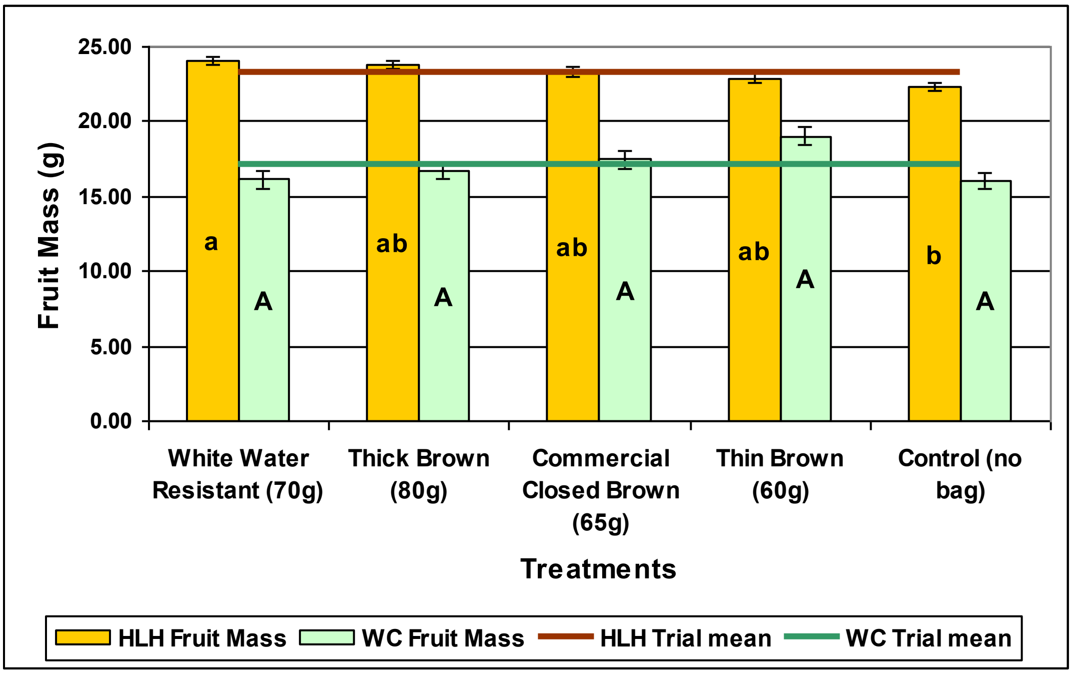


Figure 2 'Wai Chee' (WC) and 'HLH Mauritius' (HLH) fruit mass (g) at Nelspruit as influenced by five different physical treatments. Different letters in the bars indicate significant differences at $P \leq 0.05$. Lower case letters represent HLH and upper case letters represent WC.

5.3.2 Fruit size

5.3.2.1 Fruit length

Figure 3 shows that length (mm) of 'HLH Mauritius' fruit was significantly increased when covered with the "Thick Brown" sleeve (80 g) and "White Water Resistant" sleeve (70 g) when compared to control fruit. All other treatments showed an enhancing effect on fruit length of 'HLH Mauritius', although not statistically significant. With 'Wai Chee', all the treatments showed a tendency to increase fruit length as opposed to the uncovered control fruit. However, these increases were again not statistically significant.

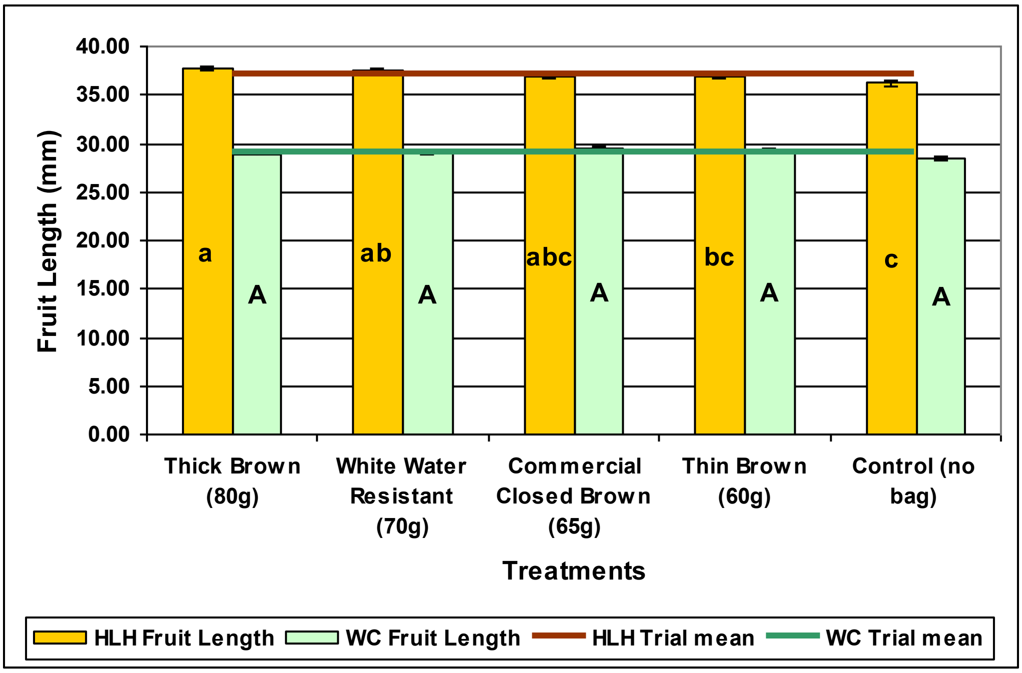


Figure 3 **Wai Chee' and 'HLH Mauritius' (HLH) fruit length (mm) at Nelspruit as influenced by five different physical treatments. Different letters in the bars indicate significant differences at P≤ 0.05. Lower case letters represent HLH and upper case letters represent WC.**

5.3.2.2 Fruit Width 1

Figure 4 presents the width of fruit (mm) with four different covering treatments and a control as measured on the broad side of fruit (width 1). Treatment with the "Thick Brown" sleeve (80 g) significantly improved fruit width1 in 'HLH Mauritius' when compared to the fruit width1 of uncovered control fruit. All the other treatments used with 'HLH Mauritius' showed tendencies of enhanced fruit width1, but these were not significant. In 'Wai Chee', treatments with the commercial "Closed Brown" bag (65 g) and the "Thin Brown" sleeve (60 g) showed a significant increase in width1 as opposed to the control. All the other covering treatments showed enhancing tendencies on fruit width, however not significant.

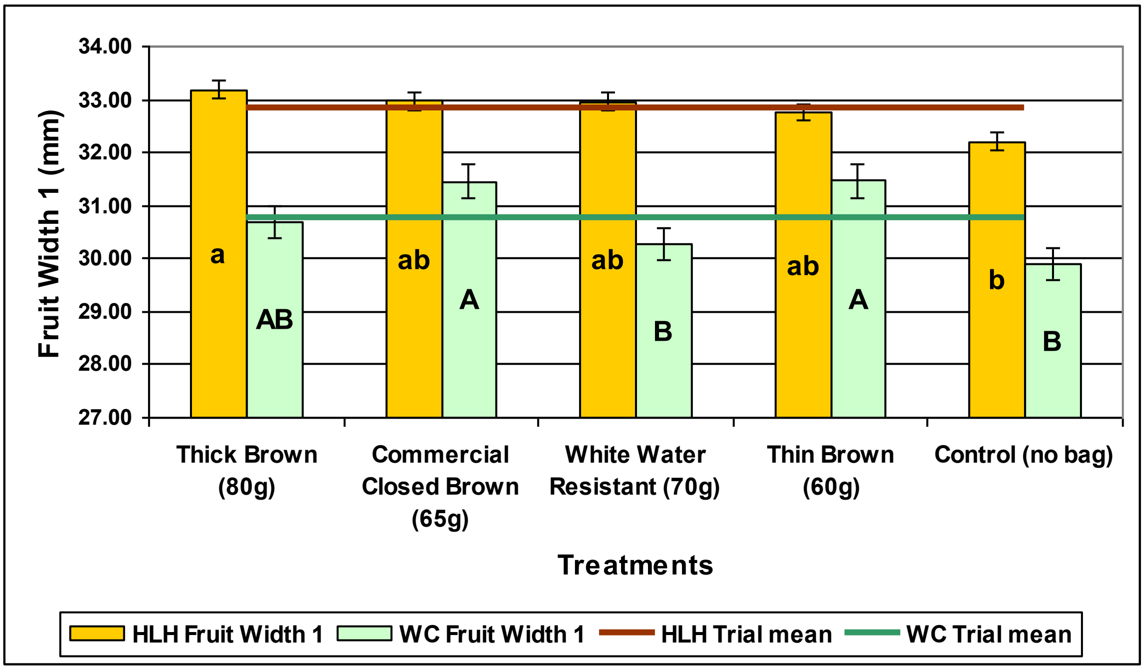


Figure 4 **Wai Chee' (WC) and 'HLH Mauritius' (HLH) fruit width1 in mm (measured on the broad part of the fruit) at Nelspruit as influenced by five different physical treatments. Different letters in the bars indicate significant differences at P≤ 0.05. Lower case letters represents HLH and upper case letters represent WC.**

5.3.2.3 Fruit Width 2

Figure 5 presents the width of fruit (mm) with four different covering treatments and a control as measured on the narrow part of the fruit (width 2). In 'HLH Mauritius', no significant differences were detected among the different covering treatments as compared to the control.

Although not statistically significant, all of the treatments showed a tendency for improvement of fruit width 2 when compared to the control. In 'Wai Chee', only the treatment with the "Thin Brown" sleeve (60 g) significantly differed from the uncovered control. Although not significant, all other treatments nevertheless either enhanced or showed similar effects on fruit width 2 when compared to the control.

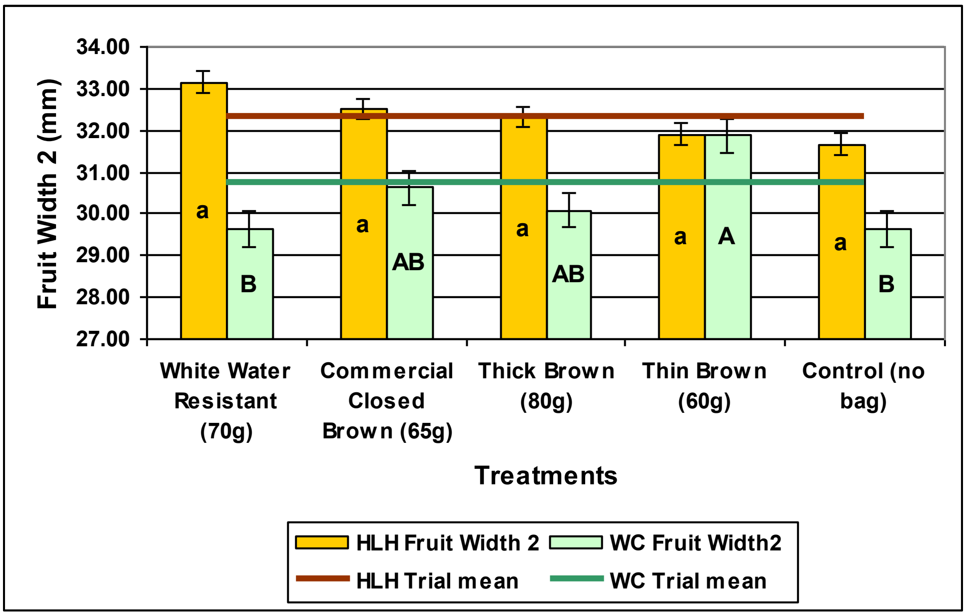


Figure 5 **'Wai Chee' (WC) and 'HLH Mauritius' fruit width2 in mm (measured on the narrow part of the fruit) at Nelspruit as influenced by 5 different physical treatments. Different letters in the bars indicate significant differences at $P \leq 0.05$. Lower case letters represent HLH and upper case letters represent WC.**

5.3.3 Maturity as represented by Total Soluble Solids (TSS)

The pulp juice Total Soluble Solids (TSS) percentage of the fruit pulp after treatment with different covering treatments is shown in Figure 6. No significant differences were found in the TSS percentages of the different treatments as compared to the control in 'HLH Mauritius'.

However, there was a tendency, although not significant, towards slightly decreased TSS with all the treatments, indicating a delayed maturity. With 'Wai Chee', significantly decreased TSS was found in fruit of the "White Water Resistant" sleeve (70 g) and the "Thick Brown" sleeve (60 g) when compared to uncovered control fruit. All other treatments also showed decreased TSS in fruit, with the exception of the commercial "Closed Brown" bag (65 g), showing a slight, non-significant increase as opposed to the control.

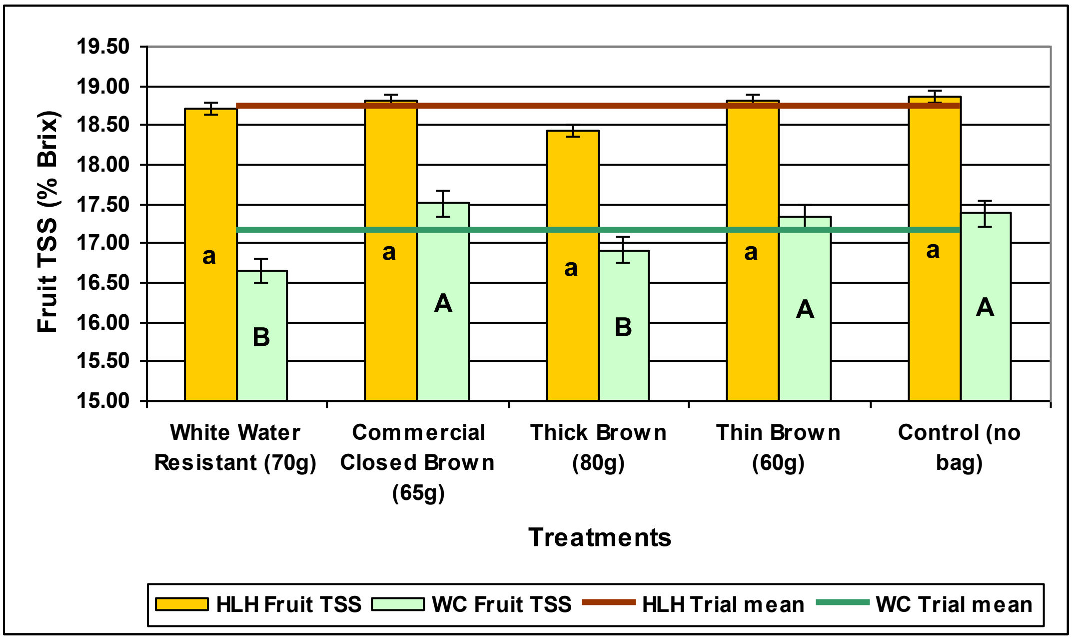


Figure 6 **Wai Chee' (WC) and 'HLH Mauritius' (HLH) fruit TSS (%Brix) at Nelspruit as influenced by five different physical treatments. Different letters in the bars indicate significant differences at P≤ 0.05. Lower case letters represent HLH and upper case letters represent WC.**

5.3.4 Fruit quality as influenced by different physical treatments.

Although the effect of covering on fruit colouration was not an objective in the current study, visual observations illustrated the benefits of covering on 'HLH Mauritius' skin colouration as can be seen in Figure 7, as well as on 'Wai Chee' in Figure 8.

It was clearly visible that covering enhanced the colour over the control in both cultivars. Although it appears to the naked eye as if there is a colour discrepancy amongst the different closing materials, visual assessment can be very inaccurate. However, with chromameter technology, being able to consistently analyse colour with continuous data that are statistically analysable, this is a trend that merits further investigation.

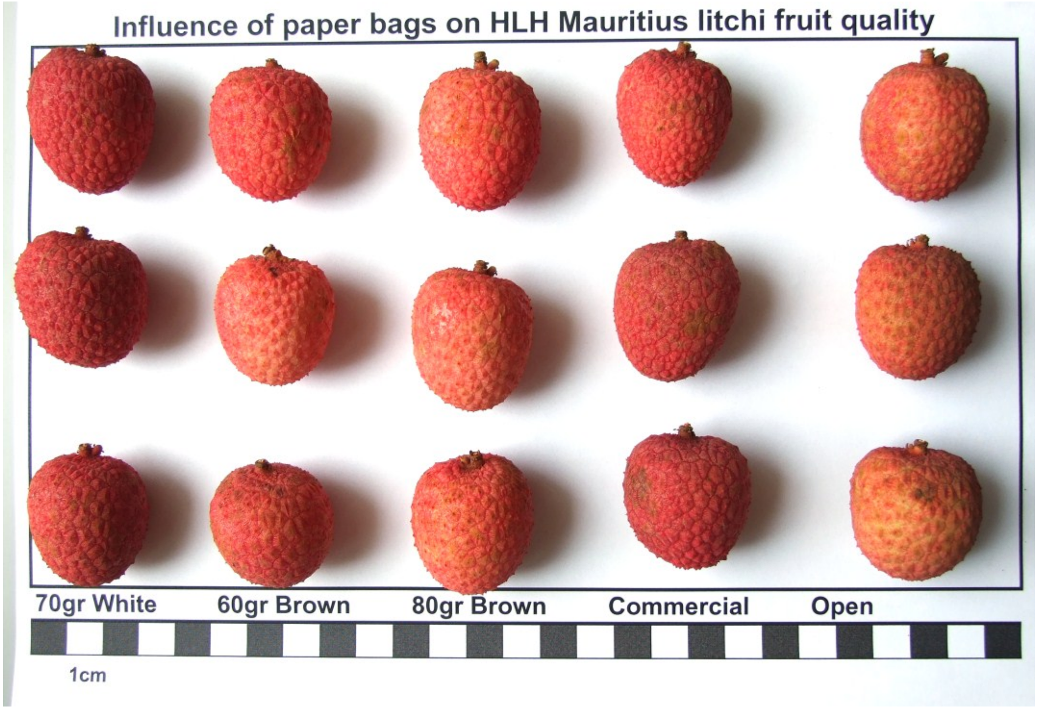


Figure 7 **'HLH Mauritius' litchi skin colour at Nelspruit as influenced by five different fruit covering treatments.**

Figure 8 **'Wai Chee' litchi skin colour at Nelspruit as influenced by five different fruit covering treatments.**

5.4 DISCUSSION AND CONCLUSION

Results in the study indicated that the use of bagging may be an effective way of enhancing fruit mass and -size in litchis. All litchi covering treatments in the study showed a trend towards enhancing fruit mass and -size on both cultivars used in the trials. Statistical proven beneficial effects were obtained with treatments of the "White Water Resistant" paper sleeve (70 g) and "Thick Brown" paper sleeve (80 g) on fruit mass as well as fruit size dimensions in 'HLH Mauritius'. In the case of 'Wai Chee', treatments with the "Thin Brown" paper sleeve (60 g) and commercial "Closed Brown" paper bag (65 g) proved to have clear enhancing effects on fruit size dimensions. These findings may indicate a variation in the response of cultivars towards various covering materials. Fruit size can be influenced by either too low or too high radiation interception. It was mentioned by Barritt *et al*. (1987) and Tombesi *et al*. (1993) that reduced radiation interception due to overshadowing leaves can have a negative effect on fruit size. Conversely, very high radiation levels may also be detrimental to optimal fruit development, especially on days where temperatures rise between 30 and 40 ºC, as is possible in litchi producing areas of South Africa. These negative effects are manifested in skin colouration or blemish as a result of direct exposure to solar radiation on hot summer days with low humidity (Oosthuizen, 1991). It was found by Tyas *et al*. (1998) that under such conditions of high radiation, shading may have the effect of increasing fruit mass by reducing the negative effects of too much radiation as well as increasing relative humidity inside the bag, and thereby reducing fruit moisture loss.

Distinct cultivar response to covering, as illustrated by the beneficial effects obtained with thicker covering on 'HLH Mauritius' (70 and 80g/m²) as opposed to a better response of 'Wai Chee' with thinner covering (60 and 65g/m²), could be due to varied sensitivity of the cultivars towards radiation levels. One explanation for the varied sensitivity towards radiation could be that less protection from radiation is needed by 'Wai Chee', having a thick, leathery skin, than what is the case with 'HLH Mauritius', with a thinner, more susceptible skin for sunburn.

Although no statistical proof was presented, literature indicated maturation of covered litchi fruit bunches to be delayed. One such study by Oosthuyse (2007) found a delayed maturation in 'Wai Chee' with the use of unspecified surface coated, white paper bags. In the current study, Total Soluble Solids (TSS) as an indicator of maturity, it is suggested that covering may influence fruit maturation rate. In this study, all treatments were harvested on the same day as the control. Only two treatments significantly delayed fruit ripening, and only in ‘Wai Chee’. TSS values of the thicker covering materials (70g/m² and 80g/m²) in ‘Wai Chee’ showed statistically significant reductions in TSS indicating delayed maturation. Although the other covering treatments on both cultivars had lower TSS readings than uncovered fruit, indicating possible delayed ripening, none of these were significant.

Again the thicker skin of ‘Wai Chee’ together with the thicker covering materials (70 and 80g/ m²), implies reduced radiation interception caused by excessive shading, thereby retarding accumulation of solids.

Although fruit size components in ‘Wai Chee’ were improved in the current study by the use of thinner covering materials (60 and 65g/ m²), it can be argued that if fruit were left on the tree for longer in the thicker covering material, further fruit development could have taken place, resulting in larger fruit size. The lower TSS readings in fruit of these thicker covering treatments indicate that fruit could have been left on the tree for longer.

It may well be possible that thicker covering materials could also have significant beneficial effects on fruit size, should fruit be left in the bags for longer. It is quite possible that this gain in fruit size may be cancelled out by too low acid levels, resulting in reduced quality. It may therefore be worthwhile to repeat the experiment on ‘Wai Chee’ in order to investigate the effect of thick, stronger covering materials on delayed maturity and the enhancement of fruit size in the cultivar, without compromising fruit quality, i.e. taste. Contrary to the current trial layout where single fruit per bunch was evaluated, it will then be possible to evaluate whole bunches for TSS as well as acids to obtain a more accurate indication of quality.

For South Africa, a later harvest date or an extended harvesting period would be beneficial for marketing litchis, since higher prices are paid for litchis towards the end of the season when competitor countries have finished their marketing season.

Although the effect of covering materials on fruit colouration was not an objective in the current study, visual observations illustrated potential benefits of covering on skin colouration. Because anthocyanin (the principal pigment responsible for the red colour in litchi) is closely related to intense red colouration, the relation between litchi skin colour and radiation would be relatively strong. Hu *et al.* (2001) showed that potential damage to anthocyanins caused by direct sunlight was reduced by bagging of litchis. Increased colour intensity with covering litchi bunches was reported by Oosthuizen (1991) and De Villiers (2002), supporting the observations in the current study. However, Oosthuyse (2007) found that skin colouration of 'Wai Chee' was not elevated as a result of covering fruit. In that study, skin colouration was quantified by estimating, in terms of a percentage, the extent to which the skin had developed an intense red colouration. These contradicting results necessitate more research on colouration as a result of closing. Scientific colour assessment is difficult if done with scores or colour charts, which is subject to environmental and human influences. It is therefore suggested that chromameter technology be applied to accurately verify the differences in colour caused by different covering materials.

The current study showed that results obtained with one cultivar cannot be used as a benchmark for all cultivars regarding different covering materials and additional cultivars should therefore be included in future investigations.

Various other aspects, such as other materials than paper, colour of covering material, water resistance of material and size of bag/sleeve relevant to bunch size, could also play a role in the enhancing effects on litchi fruit. However, the economic viability of the litchi bunch covering practice should also be determined.

CONCLUSION

AND

FINAL RECOMMENDATION

CONCLUSIONS

The main aim of the study was to find ways of improving the appeal of 'Wai Chee' in order to realise its potential as a commercial, late harvesting cultivar in South Africa. Potentially, cultivation of the cultivar can be very profitable, since fruit is harvested in South Africa at a time when there is a gap in worldwide litchi production. However, small fruit size and questionable quality with a poor TSS: acid ratio restrict this potential.

Different horticultural manipulation techniques to improve tree-, fruit- and quality characteristics were therefore considered so that the future of this cultivar in South Africa could be ensured. The study started out by considering various chemical, biological and cultural practices that could be administered to address the fruit size and quality issue in 'Wai Chee'.

Investigation into suitable chemical products to attain this goal has led to the application of various nutrition- and plant growth regulator substances. Previous research had indicated the decline of yields and fruit quality in litchis when elemental nutrients are not optimal (Singh & Babita, 2002; Menzel, 2005). In this study, foliar nutrient applications of nitrogen, potassium and calcium during the early stages of fruit set and -development improved fruit set and subsequently yield, and also increased fruit mass, fruit size and flesh mass. Treatments with potassium nitrate (KNO_3), calcium nitrate ($CaNO_3$) and calcium metalosate proved to be the most enhancing nutrient applications in this regard.

Work done by Stern *et al.* (2001) and by Wittmann (2002) showed the advantages of synthetic auxins in obtaining increases in yield and in fruit mass of litchis. The use of synthetic auxins has already become part of commercial orchard management practices for litchis in Israel (Stern & Gazit, 2000). In this study, applications of synthetic auxins and auxin-like substances during the 2-3g stage of fruit development improved fruit size, fruit mass and flesh mass in 'Wai Chee'. The combination treatment of Tipimon® (2,4,5-TP), applied at the 2g stage, followed by Maxim® (3,5,6-TPA) a week later, yielded the best results in this regard.

The influence of exogenous applications of naturally-occurring auxins and cytokinins had a positive influence on most of the parameters, but these influences were not statistically significant. Future studies on different concentrations and timing of applications may investigate these trends further. The potential of ethylene inhibitors to delay harvesting time and thereby improve fruit size and quality could not be verified. Conflicting results necessitates more in-depth research in this field regarding application time and concentration of chemicals. The use of ethylene inhibitors in a late cultivar such as ‘Wai Chee’ can potentially be a useful tool to delay harvesting in order to benefit from the high returns for end-of- season litchi fruit.

Improved yield, fruit size and quality with application of certain foliar nutrients and plant growth regulators suggest that their inclusion in a litchi orchard management programme for ‘Wai Chee’ may be beneficial. Two different sequential programmes, including the best performing chemicals applied at various stages of fruit development, showed no significant advantage when compared to the use of single products.

With biological practices, pollination was found to play an important role in litchi tree- and fruit characteristics. In Israel and in Australia improvements were reported on the positive effects of cross-pollination on yield and fruit size (Batten & Mc Conchie, 1991; Degani *et al*, 1995). To determine the effects of self- and cross-pollination on fruit retention and fruit characteristics in ‘Wai Chee’, pollen of four different cultivars were used to hand pollinate enclosed ‘Wai Chee’ flowers. Initial fruit set in ‘Wai Chee’ was lower in all cross-pollinating treatments compared to the self-pollinating treatment. However, final fruit retention was higher with all cross-pollinators compared to self-pollination, with pollen of the cultivars ‘Fay Zee Siu’ and ‘McLean’s Red’ showing significant increases of 48% and 30% respectively in fruit retention. Increases in fruit size, fruit- and flesh mass in ‘Wai Chee’ were observed with cross-pollination, but these increasing trends were not significant. Pollen donor effect on quality parameters such as acid- and Total Soluble Solids content of fruit were also not significant. Results of this trial indicate that the inclusion of cross-pollinators in orchards to encourage pollen transfer of their Male 2 flowers to the female flowers of ‘Wai Chee’ may have beneficial effects on production. However, another season of data will be required in order to reach a meaningful conclusion on the effects of cross-pollination on fruit size and quality aspects.

Cultural practices such as bunch covering are used in litchis to improve fruit quality aspects such as size and colour (Galan Sauco & Menini, 1989; Oosthuizen, 1991). It was found that factors such as radiation levels and humidity are important for good quality fruit. Additionally, maturity of litchi fruit in bags can be delayed for up to two weeks (Oosthuizen, 1991), which can have the advantage of extending the harvest of a late cultivar like 'Wai Chee' even further. The effect of different bunch covering materials on size and maturation rate of fruit was subsequently investigated in the litchi cultivars 'Wai Chee' and 'HLH Mauritius'. Beneficial effects on fruit size were obtained with thicker covering materials having a nominal mass of 70 and 80 g/m² on 'HLH Mauritius' as opposed to uncovered bunches, while with 'Wai Chee', thinner covering materials (60 and 65 g/m²) as opposed to no covering, showed enhancing effects regarding fruit width. Maturation rate was significantly delayed only on 'Wai Chee' with the use of thicker covering materials (70 and 80g/m²). Although not the primary objective of the study, visual observations also indicated potential benefits of bagging on skin colouration. Differences in colour were detected amongst different covering materials, but these should be verified with Chromameter technology in future studies. The data indicated that bagging could be a viable option for improving fruit size and for extending the harvesting period of litchis. Especially for 'Wai Chee', a late cultivar, better fruit size and a later harvest date would be beneficial, since better prices can be realised towards the end of the season.

The work has indicated that it is possible to enhance fruit size and quality aspects in the 'Wai Chee' litchi cultivar. The horticultural manipulation techniques investigated in this study interact with the plant at different stages of fruit development and the best result of each manipulation technique could therefore be included in an existing commercial litchi orchard management programme of 'Wai Chee' litchis. However, in spite of scientific proof, a technique is only commercially viable if it has an economic impact, which was not the focus of this study and should be explored further before these techniques can be applied commercially.

FINAL RECOMMENDATIONS

The 'Wai Chee' cultivar has the potential to fill a very profitable gap in the international litchi market. If the fruit size and quality problems of the cultivar can be successfully addressed, the appeal of 'Wai Chee' will be enhanced, resulting in better competitiveness on local and overseas markets.

Horticultural manipulation techniques investigated in this study interact with the plant at different stages of fruit development. It is therefore recommended that the best result of each manipulation technique be included in an existing commercial orchard management programme of 'Wai Chee' litchi. The logical approach would be to manipulate pollination of 'Wai Chee' litchi through cross- pollination, followed by additional nutrient applications of nitrogen, potassium and calcium in sufficient quantities, especially during the critical early stages of fruit set and -development. Coupled to this is the application of synthetic auxins during the 2-3g stage of fruit development, to prevent abscission of fruit and to increase fruit mass. After fruit set, fruit bunches can be covered to improve size and quality of fruit, and more importantly, to delay maturity and thereby extending the harvesting season of 'Wai Chee'.

However, in spite of scientific evidence indicating the best performer per technique, the economic impact, as part of a commercial orchard management plan, needs to be investigated in a new study before commercial application.

In such a study the following should be taken into account:

- For the practical implementation of cross-pollinators, the advantage of inter-planting with cross-pollinating cultivars needs. to be investigated further, since this is a common practice in many other fruit crops. Various studies have already indicated a beneficial effect of cross-pollination on yields of litchis.
- The cost of the chemicals applied should be compared with its impact on the profit margin.
- In view of producer resistance to using covering materials in other litchi cultivars, the financial advantage of covering a dwarfed, late cultivar such as 'Wai Chee' should be investigated. The choice of covering material for 'Wai Chee" warrants more investigation on a commercial scale to

determine the effects of different covering materials regarding the colour, size and water resistance of such materials. Chromameter technology should be used in future studies to accurately verify skin colour differences between various treatments.

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